**001**

# Reducing nitrous oxide emissions from grazed grasslands

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**Application** Grazed grassland in Ireland and the UK use substantial amounts of nitrogen fertiliser. Changing fertiliser type and rate can reduce nitrous oxide emissions while maintaining grass yields and nitrogen recovery.

**Introduction** Fertiliser nitrogen (N) is a cornerstone input in many intensive agricultural systems including those prevalent in Irish temperate grassland. However, N fertiliser is associated with environmental loss of the potent greenhouse gas nitrous oxide ($N_2O$). Agriculture faces the challenging EU target of reducing greenhouse gas emissions by 30% by 2030 while also remaining economically competitive. The objective of the study was to investigate the effect of N fertiliser form and the use of N stabilisers on yield, N uptake, $N_2O$ and ammonia ($NH_3$) emissions.

**Material and methods** Five N fertiliser formulations; 1. calcium ammonium nitrate (CAN), 2. urea, 3. urea+NBPT (urease inhibitor), 4. urea+DCD (nitrification inhibitor) and 5. urea+NBPT+DCD were evaluated for agronomic and environmental loss parameters on three Irish grassland soils over 2 years. The study followed a randomised block design with 6 replicates per treatment. Emissons were measured using static chambers ($N_2O$) and wind tunnels ($NH_3$). Data were analysed using Proc Glimmix (SAS) and the factors in the model were site-year, fertilizer N type and their interactions as fixed effects with block as a random.

**Results** Emissions of the greenhouse gas $N_2O$ were highest ($P<0.05$) and most variable for CAN which had an emission factor (EF) of 1.49%. Emissions for the urea treatments were lower at all site-years, mean EFs were 0.25, 0.4, 0.11 and 0.11% for urea, urea+NBPT, urea+NBPT+DCD and urea+DCD, respectively. $N_2O$ emissions from urea fertilisers were less variable than CAN with the coefficient of variation ranging for urea based treatments 14-38% compared to 61% for CAN. Thus urea based fertilisers reduced $N_2O$ emissions by 58-87% (Harty *et al*. 2016). All fertiliser options gave similar grass dry matter annual yields across the sites and years, with the exception of urea+DCD which had significantly lower yield than the other treatments at three site-years (Forrestal *et al*. 2017, Harty *et al* 2017). Urea and urea+DCD had significantly lower apparent fertiliser N recovery efficiency than CAN and urea+NBPT which were consistently equal. The urea+NBPT treatment had significantly lower $NH_3$ emissions compared with urea; on average 78.5% lower (Forrestal *et al*. 2016). Urea stabilised with NBPT matched the yield performance and $NH_3$ emissions of CAN while having similar $N_2O$ emissions to straight urea.

**Conclusion** Fertiliser formulation including the use of N stabilisers can help farmers meet agronomic and environmental goals without reducing the N rates that underpin production. The use of NBPT with urea does not substantially increase $NH_3$ emissions. Urea with NBPT matches the grass yield and N fertiliser recovery of CAN on Irish grassland soils while reducing $N_2O$ emissions and not increasing $NH_3$ emissions.

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**References**

Forrestal P J, Harty M A, Carolan R, Lanigan G J, Watson C J, Laughlin R J, McNeill G, Chambers B J and Richards K G 2016. Soil Use and Management 32 S1, 92–100.
Forrestal P J, Harty M A, Carolan R, Watson C J, Lanigan G J, Wall D P, Hennessy D and Richards K G 2017. Soil Use and Management 33, 243–251.
Harty M A, Forrestal P J, Carolan R, Watson C J, Hennessy D, Lanigan G J, Wall D P and Richards K G 2017. Journal of Agronomy 109, 1-9.
Harty M A, Forrestal P J, Watson C J, McGeough K L, Carolan R, Elliot C, Krol D, Laughlin R J, Richards K G and Lanigan G J 2016. Science of the Total Environment 563–564, 576–586.

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002

# Livestock insurance: a tool to reduce economical loss of farmers from climate change related hazards

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**Application** Climate change brings extreme events like drought, landslide, flood and anticipated more constraint to profitable livestock production causing huge economical loss in the livestock sector of the country. Deaths of livestock and damage to farms and farm infrastructure is causing a huge loss, small livestock holders are highly venerable to such climatic hazards. So to cope with these uncertain climatic hazards livestock insurance is the one of the best strategies.

**Introduction** Nepal has a very diverse and fragile topography; it is the 20th topmost disaster prone country in the world. It is ranked on 4th place with regard to vulnerability to climate change, and 30th for flood related calmatives (MOHA, 2015). Livestock rearing is a major source of income, food and nutrition, manure for the farmers of Nepal. Studies on livestock and climate change revealed that climate change adversely affects the animal health and livestock production. An increase in extreme climate events, such as droughts and floods, is anticipated more constraint to profitable livestock production (Christensen *et al.,* 2007). Every year farmers bear large economical loss due to climatic hazards. In the year 2013 (1,535 animals) and in year 2014(5282) were killed by climatic hazards. (MOHA, 2015). Recent flood in the August, 2017 in terai of Nepal the livestock sector has incurred a loss of around Rs. 1 Billion due to deaths of livestock and damage to farms and other infrastructure (MoLD, 2017). This uncertainty in the weather and weather related hazards is causing huge economical loss to the livestock farmers so livestock insurance is the best way to reduce the economic loss of the livestock holders.

**Material and methods** The study place includes three districts of Nepal. Jhapa from terai, Dolakha from the mountain and Kaski from the hilly region of Nepal. These Districts were selected purposively as livestock rearing is major source of income for the livelihood of the people. From each district 100 households were selected using purposive simple random sampling technique. A total of 300 households were used for the survey. The primary data was collected through household survey using pretested semi structured questionnaire via face to face interview during July 2016 to February 2017. The questionnaire survey was focused on perception of the climate change, major effect on livestock due to changing climate and possible adaptation measures, knowledge about livestock insurance policies, constrains of insurance and livestock insurance a way to mitigate with changing climate were assessed. All quantitative data were entered in the Statistical Package for Social Science (SPSS 16 version). Microsoft Word, Microsoft Excel, and SPSS were used for data processing, analysis, and interpretation of information.

**Results** A perception of the change in climate was done by questioner survey. Change in the temperature, rainfall pattern, intensity of the rainfall, drought, flood, pest infection and hailstone are the major climate change issue people have noticed. 62% of the respondent has observed the change in climate. Disease and pest infection, drought, flood and hailstone are the major factors affecting livestock in these districts. Occurrence of calamities are seen more in the mountain and hill nearly 68.0% of total respondents perceived the increase in temperature, rainfall intensity, climatic calamities like flood, landslide, hailstone, disease and pest as compared to last five years. 47% of farmer has insured their livestock in Kaski district, 33% in Jhapa and 20 % in Dolakha district. Beside the proper vaccination, deworming, management and feeding of animals people are insuring their livestock assets due to uncertain in the climatic pattern and its consequences.

**Conclusion** Deaths of livestock and damage to farms and farm infrastructure is causing the farmers a huge loss, small livestock holders are highly venerable to such climatic hazards. So to cope with these uncertain climatic hazards livestock insurance is the one of the best strategies. All though lack of knowledge of livestock insurance, lack of the interest of the insurance company to work in the remote area is also the major constrains. Combining data of climate and weather, livestock supply, and insurance market can be use to formulate a well suited livestock index insurance by the government to prevent the economical loss of the livestock holders.

**References**

Christensen L 2004. Vulnerability of the Asian typical steppe to grazing and climate change. Climate Change 63, 351-368.
MOAD. 2013. Agriculture and livestock insurance policy.
MOAD. 2014. Agriculture and livestock insurance premium subsidy policy.
MOHA. 2009. Nepal Disaster Report, 2015, Ministry of Home Affairs, Kathmandu.
MOHA. 2010. b: Draft of Disaster Management Bill 2010, Ministry of Home Affairs, Kathmandu .
MOHA. 2010. Disaster Scenario in Nepal (Unpublished data), Disaster Management section, Ministry of Home Affairs.
MOLD. 2017. Report on the livestock death due to recent flood in Terai.
Shrestha, A.B, Wake, C.P, Mayewski, P.A, Dibb, J.E.1999. Maximum temperature trends in the Himalaya, an analysis based on temperature records from Nepal for the period 1971-94. Journal of Climate 12, 2775-2787.

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**003**

# How will the nutritional quality of European pastures be affected by climate change?

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**Application** Changes in pasture quality impact the productivity of ruminant animals that rely on grass as a main component of their diet. Understanding these changes is essential for farmers and will enable them to adjust their management practices appropriately.

**Introduction** Climate change will have significant effects throughout Europe. Some areas, particularly in the north-east, will become considerably wetter (+W), while southern Europe will become drier (-W). All areas are predicted to experience increased temperatures (+T), as well as heightened atmospheric $CO_2$ concentrations (+C) (EEA 2017). The objectives of this research are to quantify the effects of these changes on the quality of pastures across different European regions and for different plant functional groups (PFGs). This is done by means of a meta-analysis of previous studies.

**Material and methods** Studies for the meta-analysis were selected through a systematic review of the available literature, including the Web of Science database, grey literature, bibliographies of key review articles and other meta-analyses. To be included a study must: (i) be conducted in Europe or else in laboratory conditions representative of European weather conditions; (ii) include a common European forage species; (iii) consider the effect of +C, +T, +W and/or –W on plant nutritional quality (e.g. carbon/nitrogen (C/N) ratio, N concentration, sugar and starch content); (iv) provide the mean, standard deviation (or equivalent) and sample size. 144 studies were selected as being relevant; the average +C was 284±79ppm (mean ± sd), +T: 3.2±1.7°C, +W: 117±96% more water than control treatments, -W: 79±26% less water than control treatments. The effect size used was the natural logarithm of the response ratio (result under test conditions/result under control conditions). Fixed effects models were implemented in WinBUGS, which employs a Bayesian methodology using Markov Chain Monte Carlo simulations. Results were grouped where region/PFG was not found to have a significant effect or there was insufficient data for a division. There are no results for the northern region under –W as this is not an expected consequence of climate change. Analyses were only performed where data from at least five studies was available.

**Results** Plant N concentration is a measure of protein concentration; it generally decreased for all treatments (Table 1) with the exception of +W, where there was a mean increase of 10.5%. Under +T conditions, graminoids showed the smallest decrease in N concentration (-4.2%) and forbs the greatest (-18.0%), with shrubs and legumes in-between. There was no significant effect between PFGs for +C, +W or –W. C/N ratios under +C tended to increase, the lowest increase was for legumes (7.7%), followed by forbs (19.5%) and then graminoids (25.9%); no data was available for shrubs. There was insufficient data to divide the results for the other climatic changes or for other quality measures by PFG.

**Table 1** Percentage change in N concentration, C/N ratio, soluble sugar content and starch content under +C, +T and –W conditions for different European regions. Values are mean ± standard deviation.

| | N concentration | | | C/N ratio | | Soluble sugars | | Starch |
|---|---|---|---|---|---|---|---|---|
| | +C | +T | -W | +C | +T | +C | -W | +C |
| Alpine | -8.2±4.0 | | -16.2±6.1 | 1.4±5.7 | | | | |
| Atlantic | -21.5±3.2 | | -2.6±7.9 | 19.0±2.8 | | | | |
| Continental | -13.2±2.6 | -10.5±1.8 | -10.3±6.0 | 18.6±3.6 | 11.9±3.3 | 17.4±10.5 | 1.3±5.0 | 39.6±14.1 |
| Southern | -2.1±4.8 | | -20.0±6.9 | 33.2±12.1 | | | | |
| Northern | 7.0±6.6 | | N/A | No data | | | N/A | |

**Conclusion** While northern Europe may experience a slight improvement in the protein (N) concentration of its pastures, other regions will see reductions, particularly when conditions are warm and dry. On the other hand, non-structural carbohydrates (sugars and starch) will increase, providing more energy for grazing livestock. It seems likely that irrigation will become increasing necessary in drier areas, to counteract the negative effects of reduced water availability, particularly as droughts become more frequent; alternatively sheep and goat production may have to move to other areas. It is also probable that livestock will require more concentrate feed in the future, to mitigate the loss of protein. With different PFGs responding in different ways, it is also very likely that there will be changes in pasture species composition.

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**References**

EEA. 2017. Climate change, impacts and vulnerability in Europe 2016. European Environment Agency, Luxembourg.

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004

# Effects of grassland sward diversity on nitrous oxide emissions following fertiliser N application under ambient and wet soil conditions

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**Application** Timing of N fertiliser application based on weather and soil moisture may help reduce $N_2O$ emissions from grassland swards.

**Introduction** Nitrous oxide from fertilised grassland pastures are an important source of anthropogenic greenhouse gas (GHG) emissions. Numerous factors have been shown to influence $N_2O$ emissions from soil, such as; moisture content, temperature, organic matter, and fertiliser source and rate (Liang *et al*. 2016). Mixed species swards are known to enhance sustainability indicators like invertebrate abundance and diversity. However, N fertiliser use and sward legume content interactions under differing environmental conditions are complex. Niklaus *et al.* (2016) found that species richness reduced $N_2O$ emissions over time except from plots with increasing legume content when fertiliser was added. The aim of the present study was to investigate the effect of grassland sward diversity on $N_2O$ fluxes following fertiliser N application under two contrasting soil moisture regimes.

**Material and methods** This study used plots from a multispecies trial (Smartgrass) established in September 2013 at UCD Lyons Research Farm on a silty loam Gleysol. The experiment was a Simplex design with each plot (2 m x 4 m) containing a mix of three plant functional groups (grasses, legumes and herbs) at varying ratios. The plot mixes are represented as; G: L: H, in the results below. The plots were maintained at 90 kg N $ha^{-1}$ $yr^{-1}$ but had not been fertilised since the end of the growing season in 2016. Plots continued to be harvested monthly during 2017. In July 2017 eight plots were selected for study. Two static chamber collars were deployed in each plot. The area within and around one of the static chambers in each plot was wetted to increase the soil moisture content. Soil moisture contents were monitored throughout the experiment using a Theta probe. Rewetting was carried out if the soil moisture content of the wet half of the plot was within 5% of the dry (ambient) half of the plot. A liquid urea fertiliser was applied to the base and surrounding area of the chambers at a rate of 40 kg N $ha^{-1}$ on 11/07/2017. Gas sampling took place once prior to fertiliser application. Following fertiliser application, gas sampling took place 4 times per week in the first 2 weeks, then twice per week for the following 2 weeks and once per week for the following month. Three sampling times were used to estimate fluxes; 0, 30 and 60 minutes. $N_2O$ concentrations were measured using a Bruker Scion 456GC with a 63Ni electron capture detector (ECD). Daily $N_2O$ fluxes were calculated using the following equation: $(\Delta C/\Delta t) \times ((M \times P)/(R \times T)) \times (V/A)$ and then extrapolated to units of g $N_2O$-N $ha^{-1}$ $d^{-1}$ (De Klein and Harvey, 2012). Daily fluxes were used to estimate the cumulative $N_2O$ emissions from each sward type over the 8-week sampling period.

**Results** $N_2O$ fluxes ranged from -3.11 to 86.86 g $N_2O$-N $ha^{-1}$ $d^{-1}$. The mean overall $N_2O$-N loss from dry and wet soil conditions was 0.99 and 1.87 kg $N_2O$-N $ha^{-1}$ $d^{-1}$, respectively. The largest flux occurred after heavy rainfall from the 14G: 7.2L: 0H sward under wet soil conditions. Figure 1 shows cumulative $N_2O$-N loss as a percentage of fertiliser N applied, for all the sward and soil condition treatments.

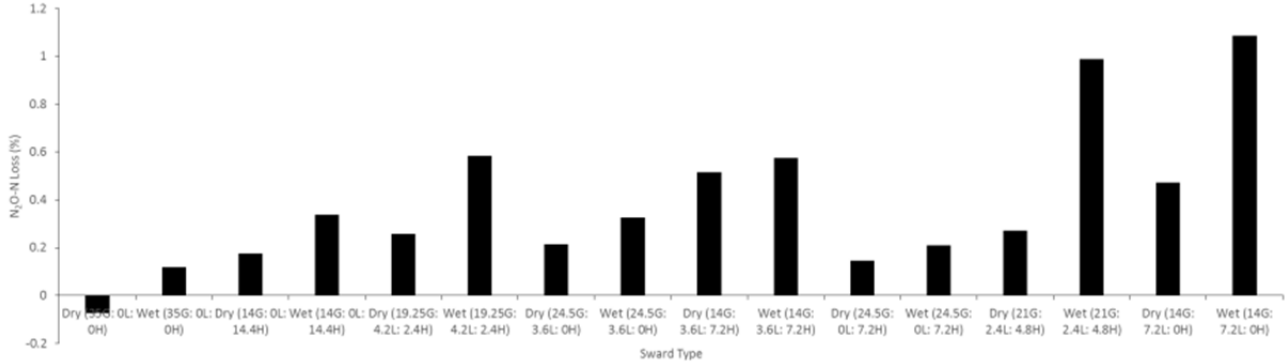


**Figure 1** $N_2O$-N loss from different sward types and soil conditions as a percentage of nitrogen rate applied.

**Conclusion** Greater $N_2O$ loss occurred from wet soil than dry soil conditions across all sward types. N fertiliser timing based on forecasted weather and antecedent soil conditions could help reduce $N_2O$ losses.

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**References**

De Klein, C. A. M. and Harvey, M. 2012. Nitrous oxide chamber methodology guidelines.
Liang, L. L., Grantz, D. A. and Jenerette, G. D. 2016. Global Change Biology 22, 1286-1298.
Niklaus, P. A., Le Roux, X., Poly, F., Buchmann, N., Scherer-Lorenzen, M., Weigelt, A. and Barnard, R. L. 2016. Oecologia 181, 919-930.
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**005**

# Novel metrics for assessing, monitoring and improving dairy farm sustainability

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**Application** Development of novel dairy farm efficiency benchmarking metrics to help solve practical farm management problems. Focus on the identification of cost-saving and profit-making opportunities for farmers.

**Introduction** There is an ever-increasing need to develop metrics able to capture the numerous socio-economic and environmental dimensions of (dairy) farming and thus help achieve 'sustainable intensification' of UK agriculture. Measurement and monitoring of farm performance is typically effected via benchmarking to identify best-practice management for a given farming system, to guide farmers on how to produce more sustainably (Fraser and Cordina, 1999). 'Conventional' benchmarking based on average values or ratios may mislead when performance and profitability are determined by interrelated multifactorial processes (Cooper *et al*., 2007). By contrast, the benchmarking method Data Envelopment Analysis (DEA; Cooper *et al*., 2007), accounts for several processes simultaneously, identifies benchmarks for *each* farm and indicates the adjustments that this farm should make to its resource use and production ('good' *and* 'bad') to reach its benchmarks. Dairy studies have shown that DEA is a flexible and holistic tool to suit particular objectives for the benefit of both business management and the public good (Soteriades *et al*., 2015), yet little effort has been placed on moving the method from agricultural research to farm management. This study lays the groundwork for DEA assessments tailored to fulfil the needs of farm management.

**Material and methods** DEA constructs a *best-practice frontier* (Figure 1: a simple, single-input, single output case) consisting of the best performers (i.e. the efficient farms) in the sample (farms A, B, C, D, E, F) and all other farms (G) are benchmarked against it. Farm G is inefficient as it could be producing more output using less input relative to one or more efficient farms. To become efficient, farm G needs to eliminate its input and output inefficiencies to reach a point on the frontier. DEA detects the magnitudes of these inefficiencies in each input and output and then aggregates them into a single overall 'score', between 0 and 100%, which gives the farm advisor a convenient overview of the farm's performance. However, the inefficiencies may also be 'priced' to demonstrate to the farmer the financial benefits of operating efficiently. The single-input single-output case can be easily extended to multiple inputs and outputs (Soteriades *et al*., 2015). Detailed data for 675 UK dairy farms were selected from the database of Kingshay Farming & Conservation Ltd., covering the year 2014–2015. Six inputs and three outputs were considered for the DEA exercise. Inputs: cows in herd (numbers); forage area (ha); replacements (numbers); purchased feed (kg dry matter); somatic cell count (SCC; '000s/mL); and bacterial count (BC; '000s/mL). Outputs: milk yield (L); butterfat yield (kg); and protein yield (kg). DEA-derived inefficiencies for SCC, BC and butterfat and protein yields were used to estimate the financial benefits (better milk price) that a farm could gain by eliminating them. The 'new' milk price was calculated with AHDB Dairy's Milk Price Calculator.

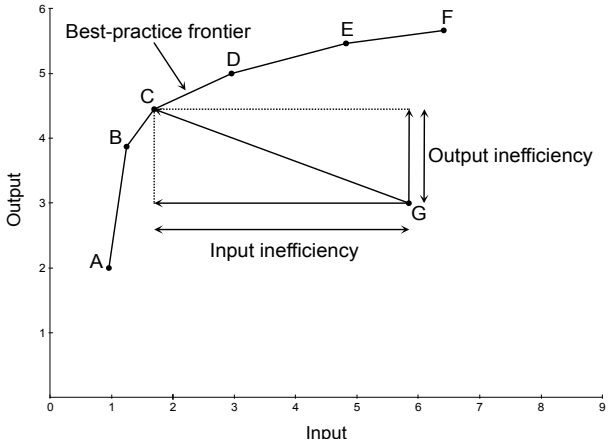


**Figure 1** A single-input single-output case

**Results** The 'new' milk price for the farm exhibiting the highest SCC and BC inefficiencies in the sample could have increased from 20.43ppL to 29.55ppL, had this farmer concentrated on reducing SCC, BC and on increasing butterfat and protein yields. Similarly, another farm in the sample could be improving its margin over purchased feed per cow by £725/year just by using purchased feed more efficiently.

**Conclusion** The examples demonstrated that DEA can help farmers substantially improve economic performance, as well as to monitor overall performance. This exercise can be extended to guide other priorities such as the improvement of farm environmental performance, the monitoring of efficiency over time or the benchmarking of farms between and within specific farm types. As uptake of smart farming technologies expand there is an increasing opportunity to add value by processing data into meaningful decision support using tools such as DEA.

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**References**

Cooper, W. W., Seiford, L. M. and Tone, K. 2007. ISBN: 9780387452838.
Fraser, I. M. and Cordina, D. 1999. Agricultural Systems 59, 267–282.
Soteriades, A. D., Faverdin, P., March, M. and Stott, A. W. 2015. Agricultural and Food Science 24, 235–248.

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006

# A review of the activity data used to estimate national methane emission factors for Irish livestock

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**Application** This review identified new activity data from national sources (e.g., Teagasc national farm survey), which can be used to update the static input variables (e.g., grazing season length) of livestock methane emission factors. This new data, if adopted, should increase the Irish inventories capacity to capture the mitigation efforts of the livestock sector.

**Introduction** The Irish national greenhouse gas (GHG) inventory uses default tier 1 emission factors and country specific methods (tier 2) to estimate livestock's enteric fermentation and manure methane emissions. Tier 2 methods were developed in 2006, for cattle, from an extensive farm facilities survey conducted by Hyde *et al*. (2008) and from the research of O'Mara (2006) on cattle methane emissions. The tier 2 emission factors developed significantly improved the national accuracy of measuring Irish livestock methane emissions, but did increase national inventory data requirements. The tier 1 approach only required collecting data on the national population of animals in a livestock category, but for the tier 2 methods significantly more information is required to characterise the livestock herd. This information can be difficult to update e.g., milk fed to dairy calves. The goals of this research were twofold; first to evaluate the current activity data used to estimate livestock methane emission factors, and second to identify what new data, if any, is available to update these emission factors.

**Material and methods** The activity data used to estimate livestock methane emission factors in 2015 was reviewed. The review considered all input data variables required to compute livestock methane i.e., livestock populations, body weights, birth and mortality rates, parturition dates, farm feeding practices, housing periods, milk and meat production, age at slaughter, and farm facilities (e.g., animal waste management systems). Inventory input variables were evaluated in terms of their age and origin (e.g., national statistics or expert opinion). Following this initial review, we then assessed what livestock data was available to populate the national greenhouse gas (GHG) inventory. This assessment first considered current suppliers of inventory data, namely the Central Statistics Office (CSO) and the Department of Agriculture, Food and the Marine (DAFM). We then evaluated new data source options i.e., the Teagasc national farm survey (NFS), Bord Bia's Origin Green sustainability surveys (SDAS and SBLAS), and the Irish Cattle Breeding Federation (ICBF). Additionally, we compared how current and new data sources collected and verified farm data.

**Results** Our review of the livestock methane section of the 2015 Irish national inventory revealed that the activity data used for several important input variables (e.g., milk composition) have not changed for over 10 years. Generally, these input variables were estimated in 2015 using 2003 national statistics from the CSO and the DAFM, even though this statistical data is available from the same organisations for the year 2015 and prior years. Therefore, we recommended that the inventory should use, where possible, national data from the correct year for key livestock variables (e.g., milk composition and livestock slaughter weights). The inventory review also found that some key livestock inventory inputs (e.g., cow body weight and housing dates) are still based on the expert opinion of O'Mara (2006). This information is largely not collected by current inventory data sources, but is collected in the Teagasc NFS, Bord Bia sustainability survey and by the ICBF. This activity data is verified by the Teagasc NFS and current data sources using farm records, livestock passports and via farm inspections. For other data sources (Bord Bia and ICBF), data from farm diaries are typically used e.g., length of the grazing season. This is consistent with the expert opinion approach the inventory currently uses for such variables.

**Conclusion** New activity data was identified to regularly review and update several additional input variables of the livestock methane section of the national GHG inventory. This new livestock data has the potential to improve the accuracy of methane emission estimates and to increase the capacity of the inventory to capture the mitigation efforts of the livestock sector.

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**References**

Hyde, B., Carton, O. T. and Murphy, W. E. 2008. Farm Facilities Survey – Ireland 2003. Report prepared for the Department of Agriculture by Teagasc, Johnstown Castle, Co. Wexford, Ireland.
O'Mara, F. 2006. Development of emission factors for the Irish cattle herd. Environmental Protection Agency, Johnstown Castle, Co. Wexford, Ireland.

**007**

# Effect of dietary tanninferous *Vachelia karroo* leaf meal on methane emission and productivity in yearling Boer bucks

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**Application** *Vachelia karroo* (*V. karroo*) leaf meal in the diets of goats provide suitable alternative to mitigate methane emission.

**Introduction** A major problem facing the world is the emission of greenhouse gases. It has been identified that animal production is one of the main sources of methane ($CH_4$) emissions because it has 21 times the global warming potential of carbon-dioxide (Broucek, 2014). Tannins, as feed supplements or as tanninferous plants have been hypothesized to reduce $CH_4$ emission in ruminants (Patra *et al*., 2006). However, there is neither extensive nor conclusive data on tanninferous *V. karroo* leaf meal inclusion levels for optimal reduction in $CH_4$ production and emission in Boer bucks. The aim of the present study was to determine the effect of tanninferous *V. karroo* leaf meal inclusion levels on $CH_4$ emission and productivity of yearling male Boer goats.

**Material and methods** The study was conducted at the University of Limpopo Experimental farm in August, 2016. Fresh leaves of *V. karroo* were harvested at the farm in June, 2016. *Avena sativa* grass hay was used as a basal diet. The grass is well grazed during summer and is suitable for hay making. Twenty-four yearling Boer bucks with an average initial live weight of 23 ± 2 kg were allocated, in a completely randomized design, to four dietary treatments containing *V. karroo* leaf meal inclusion levels of 10, 15, 20 or 30% of the total diet. These inclusions include low and high tannin-levels as indicated in the literature. The goats were fed ad libitum, allowing a 15% refusal of each diet. The experimental lasted for 21 days. The live weight of goats was measured using an electronic weighing scale when the experiment commenced. Feed intake was measured throughout the study period. Methane emissions were measured using a hand-held methane detector according to Chagunda *et al*. (2009a). The laser beam of the methane detector was pointed at a distance of 1 m away from the goat and onto the nasal area to detect the $CH_4$ gas. Measurements were taken on individual goats with a minimum radius of 6m away from any other animal. The measurements were taken at 10:00 hrs (2 hours after feeding) when the goats were ruminating. The measurements for each goat were taken within a period of 60 seconds daily and repeated for 5 consecutive days. Methane produced was then read as parts per million-metre (ppm-m). The effects of *V. karroo* leaf meal inclusions levels on diet intake and $CH_4$ emission of yearling Boer bucks were subjected to analysis of covariance (ANCOVA) using SAS (2008). Where the covariates showed no significant effect, the data was analysed with ANOVA in a completely randomized design at 5 % level of probability with diet as a fixed factor (SAS, 2008). Where significant treatment effects were detected, means were separated by Fisher's least significant difference (LSD).

**Results** *Vachelia karroo* leaf meal inclusion level had no effect ($P > 0.05$) on diet intake and body weight of goats. However, *V. karroo* leaf meal reduced ($P < 0.05$) methane production and emission in Boer bucks.

**Table 1** Effect of *Vachelia karroo* inclusion level on dry matter intake, body weight and methane emissions in Boer bucks

| | Diet | | | |
|---|---|---|---|---|
| Variable | $VK_{10}$ | $VK_{15}$ | $VK_{20}$ | $VK_{30}$ |
| DMI (g/d) | 264±40.223 | 337±40.451 | 370±40.602 | 288±40.247 |
| BW (kg) | 23.38±3.534 | 22.88±2.511 | 24.33±2.821 | 23.27±2.576 |
| $CH_4$ emission (ppm-m) | | | | |
| Before | $22.00^{a}$ ±5.686 | $23.00^{a}$ ±5.686 | $21.67^{a}$ ±4.163 | $23.00^{a}$ ±2.603 |
| After | $13.80^{b}$ ±1.963 | $13.07^{b}$ ±1.009 | $13.33^{b}$ ±0.982 | $12.40^{b}$ ±0.757 |

[a, b]: Means with different letters in the same column are significantly different ($P < 0.05$)

**Conclusion** *Vachelia karroo* leaf meal reduced enteric methane emissions in Boer bucks. This indicates that tannins in the leaf meal were enough to inhibit methanogens.

**Acknowledgements** The authors gratefully acknowledge funding from National Research Foundation.

**References**

Broucek, J. 2014. Journal of Environmental Protection 5, 1482 – 1493.
Chagunda, M. G. G., Ross, D. and Roberts, D. J. 2009a. Computers and Electrics in Agriculture 68, 157-160.
Patra, A. K., Kamra, D. N. and Agrawal, N. 2006. Animal Feed Science and Technology 128, 276 – 291.

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**008**

# Improving nitrous oxide emission estimates from cattle excreta

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**Application** Nitrous oxide emission factors from grazing cattle excreta were, on average, lower than the IPCC default. Magnitude of emissions were strongly affected by environmental and soil conditions, but not by manipulation of urine composition.

**Introduction** Cattle excreta deposited at pasture are a large source of a potent greenhouse gas, nitrous oxide ($N_2O$). Currently, Ireland uses the IPCC default emission factor (EF) of 2% to estimate excreta-derived $N_2O$ however there is a large uncertainty around this figure. The aims of this study were to (1) quantify $N_2O$ emissions and EFs from excreta deposited to pasture by grazing animals, (2) assess impact of environmental drivers on emissions and potential of using soil-specific manipulation of grazing timing as a $N_2O$ mitigation tool and (3) assess potential mitigation of $N_2O$ emissions from urine patches by manipulating urine composition.

**Material and methods** Two experiments were conducted. In the first experiment which was a randomised split-plot design with five replicates real ruminant urine and dung were applied to three pasture soils in spring, summer and autumn. Nitrous oxide was measured with a manual static chamber method for 365 days following treatment application. In the second experiment which was a randomised block design with six replicates urine with incremental additions of minor constituents hippuric acid (HA) and/or benzoic acid (BA) was applied to pasture and $N_2O$ measured using the same method for 66 days.

**Results** The average $N_2O$ emission factor was 0.31% and 1.18% for cattle dung and urine, respectively. $N_2O$ loss was driven by rainfall, temperature and soil moisture, with highest $N_2O$ EFs during late grazing and from the imperfectly-drained soil (Table 1). However, manipulation of ruminant urine by adding HA and/or BA was found to have no effect on $N_2O$.

**Table 1** $N_2O$ emission factors as affected by soil type, grazing season and type of excreta

| Soil type | Grazing season | Dung | SEM | Urine | SEM |
|---|---|---|---|---|---|
| Well-drained Sandy Loam | Early | 0.03 | 0.04 | 0.32 | 0.09 |
| | Summer | 0.02 | 0.03 | 0.31 | 0.14 |
| | Late | 0.13 | 0.04 | 0.3 | 0.09 |
| Moderately-drained Sandy Loam | Early | 0.06 | 0.06 | 0.65 | 0.26 |
| | Summer | 0.16 | 0.06 | 0.34 | 0.06 |
| | Late | 0.24 | 0.06 | 1.16 | 0.10 |
| Imperfectly-drained Sandy Loam | Early | 0.15 | 0.18 | 1.12 | 0.26 |
| | Summer | 0.54 | 0.22 | 1.63 | 0.39 |
| | Late | 1.48 | 0.21 | 4.81 | 0.97 |
| Average | | 0.31 | 0.16 | 1.18 | 0.48 |

**Conclusion** The $N_2O$ EFs found in this study were lower than the current default values used in Ireland. Adopting these new, country-specific EFs will lead to reduction in $N_2O$ inventory. Manipulation of minor constituents in urine composition had no effect on $N_2O$ however other urine manipulations might prove successful.

**Acknowledgements** This research was funded by the Department of Agriculture, Food and the Marine (RSF10/RD/SC/716 and 11S138).

**References**

Krol, D. J., Carolan, R., Minet, E., McGeough, K. L., Watson, C. J., Forrestal, P. J., Lanigan, G. J. and Richards, K. G. 2016. Science of the Total Environment 568, 327-338.
Krol, D. J., Forrestal, P. J., Lanigan, G. J. and Richards, K. G. 2015. Science of the Total Environment 511, 362-268.

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009

# Clinical presentation, somatic cell count and cytokine secretion in response to intramammary infection of Holstein Friesian heifers with isolates from two *Staphylococcus aureus* lineages

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**Application** Bovine adapted isolates of *S. aureus* from different lineages caused a different disease outcome in dairy cows. This finding may influence the detection of mastitis (somatic cell count; SCC) as well as treatment decisions.

**Introduction** *S. aureus* is the most frequent cause of clinical and subclinical bovine mastitis in Ireland. Four bovine-adapted lineages (CC71, CC97, CC151 and ST136) were identified among a collection of Irish *S. aureus* mastitis isolates (Budd, *et al*., 2015). *In vitro* studies found differences in a variety of virulence traits between isolates of these lineages, including differences in biofilm formation, host cell internalisation, immune gene induction and cytokine secretion by infected bovine mammary epithelial cells (bMEC) (Budd, *et al*., 2016). The objective of this study was to assess the ability of isolates of *S. aureus* from different lineages to cause infection *in vivo*.

**Material and methods** All procedures were approved by the UCD Animal Ethics Committee and conducted under licence. One isolate each of CC97 and CC151, that differed in ability to cause an immune response in bMEC, were selected on the basis of *in vitro* experiments. Fourteen first-lactation Holstein Friesian cows were purchased from 2 farms. Animals were low SCC (composite SCC < 50 000 cells/ml) and free of intramammary infection. In mid-lactation animals were assigned to treatment group blocked by source farm and inoculated into the left hind quarter of the mammary gland, 7 with 173 c.f.u. of the CC97 isolate (Group 1) and 7 with 583 c.f.u. of the CC151 isolate (Group 2). The contralateral quarter of each cow was inoculated with PBS. Clinical signs of infection (temperature, milk appearance and yield) were monitored for 30 days. Blood and milk samples were taken to determine bacterial counts in milk, somatic cell count, white blood cell populations and cytokines. Data was log transformed when required and analysed with repeated measures ANOVA using SAS PROC MIXED® to compare the mean responses of Group 1 and Group 2. The model fitted for each variable included effects for group, time, source farm, initial SCC, initial value of the variable, group by time and any other significant interaction.

**Results** Differences in disease presentation between groups were observed, with two animals from Group 2 developing clinical mastitis and requiring antibiotic treatment, while one animal from Group 1 did not develop an infection for the duration of the study and another cleared the infection within 10 days. Fever (temperature > 39.5□C) was observed in 3 animals from Group 2 and in none from Group 1. Significant differences in SCC and bacterial load between groups were observed during the infection (Fig. 1). Milk yield was significantly lower in Group 2 infected animals ($P < 0.05$) with a significant group by time interaction which largely reflected a greater difference in milk yields on days 2 to 6. Data is also being collected on cytokines and chemokines secreted locally and systemically during the course of infection.

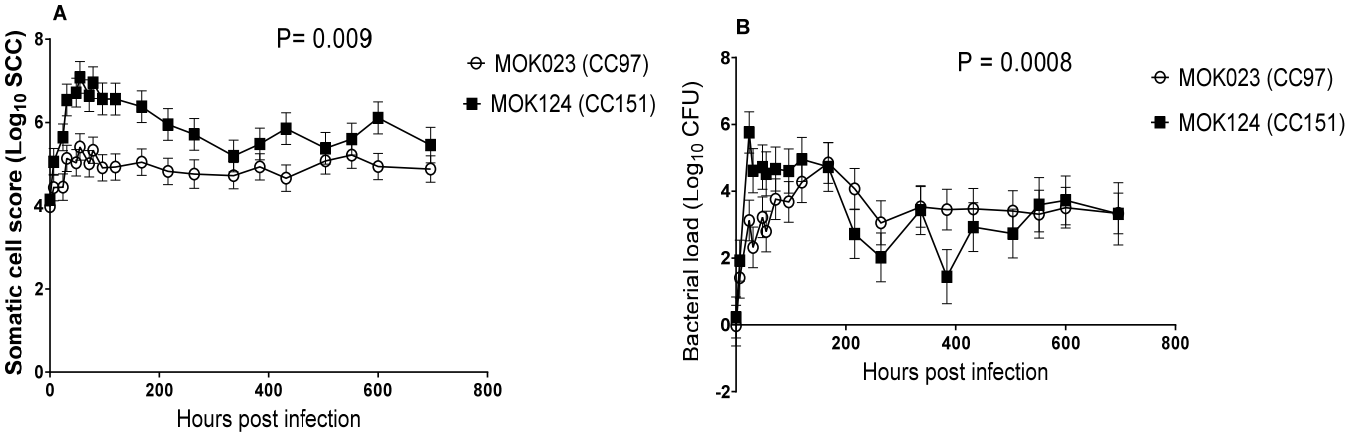


**Figure 1** SCC (A) and bacterial load (B) in Group 1 and Group 2 infected animals.

**Conclusion** The results of this study demonstrate that a *S. aureus* isolate from lineage CC151 caused more severe, clinical mastitis than an isolate from lineage CC97 which resulted in mild, subclinical mastitis. Diversity between isolates of *S. aureus* may therefore influence the clinical presentation of mastitis, which in turn may influence disease detection and treatment needs.

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**References**

Budd, K E, McCoy, F, Monecke, S, Cormican, P, Mitchell, J and Keane, O M 2015. PLoS One, 10**,** e0134592.
Budd, K E, Mitchell, J and Keane, O M 2016. Veterinary Microbiology 189**,** 24–31.

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**010**

# Use of selective dry cow therapy to control mastitis and reduce antimicrobial use

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**Application** Selective dry cow therapy (SDCT) involves administering antimicrobials only to those cows that show signs of infection at dry-off. Results from this study indicate that SDCT is possible in low SCC herds which regularly record individual animal SCC.

**Introduction** Irish dairy herds generally employ blanket dry cow therapy as a preventative measure of mastitis control, administering antimicrobials to all quarters of all cows at drying off. Given recent concerns over antimicrobial resistance, blanket dry cow therapy may come under scrutiny as an unnecessary use of antimicrobials. Selective dry cow therapy offers an alternative, more directed use of antimicrobials to only those cows that require antimicrobials at dry-off to cure existing infections. The aim of this study was to quantify the impact of treating cows with teat seal only at dry off on subsequent SCC and udder infection.

**Material and methods** This research was conducted at 3 Teagasc Animal and Grassland Research & Innovation research herds; at the Clonakilty research herd between 2015 and 2017, inclusive, and repeated at the Moorepark and Curtins research herds in 2017. Prior to dry-off, weekly milk recording data was used to identify cows which had not exceeded 200,000 somatic cells at any point in lactation and which had no clinical case of mastitis during lactation. Cows which met these criteria were eligible for the study. Eligible cows were randomly assigned one of two treatments; 1) antibiotic plus teat seal (AB&TS), or 2) teat seal only (TS). Weekly records of individual animal SCC were available. Somatic cell count was analysed as 1) average across lactation, 2) minimum animal SCC across lactation, 3) maximum animal SCC across lactation, and 4) test-day SCC. Analyses were conducted using records from 1) the first 3 weeks of lactation, 2) the first 120 days of lactation.
Somatic cell count was log-transformed to base 10 prior to analysis. The effect of treatment was quantified using a repeatability model accounting for concurrent experiment treatment level, breed (proportion of Holstein, Jersey, or Norwegian Red), heterosis, recombination, month of calving, parity (n=4; 1, 2, 3, 4+), year (n=3), and previous average daily milk yield. For SCC traits with a single lactation value (ie average, minimum and maximum SCC) animal was repeated across years of the study. Animal was repeated across lactation week when the dependent variable was test-day SCC. The likelihood of having an SCC reading ≥200,000 was quantified using logistic regression adjusted for the same fixed effects as the linear model.

**Results** Cows eligible for the experiment comprised 36%, 46%, and 56% of the Moorepark, Curtins and Clonakilty herds, respectively (n = 364 cow lactations). The SCC of TS cows was greater than those cows that received AB&TS (Table 1). Teat seal only cows were 2.9 times more likely to have an SCC reading >200,000 within the first 120 days of lactation. However, the majority of cows (>80%) in both treatments maintained SCC <200,000. All herds (with the exception of one recording) maintained a bulk tank SCC <200,000 throughout the study indicating that using TS only did not impact at the herd level.
Altering the threshold for selection by assigning cows to TS only which never exceeded 100,000 SCC in the previous lactation did not impact on results. In those analyses TS only cows still had higher SCC across lactation and a higher proportion of quarters infected with bacteria than the cows given both AB&TS.

**Table 1** Minimum, maximum and test-day SCC of cows administered TS only or AB&TS within 3 weeks or 120 DIM

| | Test day SCC | | Minimum SCC | | Maximum SCC | |
|---|---|---|---|---|---|---|
| | 3 weeks | 120 DIM | 3 weeks | 120 DIM | 3 weeks | 120 DIM |
| TS | 46,132 | 36,669 | 29,799 | 10,673 | 69,406 | 160,066 |
| TS&AB | 37,437 | 30,123 | 25,096 | 9,557 | 54,051 | 105,730 |
| p-value | 0.0387 | 0.0003 | 0.0392 | 0.129 | 0.0344 | 0.004 |

**Conclusion** Administering TS only to cows which have not had high SCC throughout lactation may offer a viable method to reduce on-farm anti-microbial use while not impacting on herd-level SCC.

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011

# Elevated inflammatory markers and impaired complement activation in cervical mucus early postpartum are distinguishing features of dairy cows which develop clinical endometritis

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**Application** Cows that develop endometritis have perturbed fertility and spend less time at peak lactation. Thus, identifying these at-risk cows early postpartum is critical.

**Introduction** Endometritis is a major economic problem in the dairy industry which reduces fertility and productivity of dairy cows. The global incidence ranges from 6.25% to 75%, and is particularly in high yielding dairy cows as reported by Foldi *et al*. (2006), Gilbert *et al*. (2005), Salilew-Wondim *et al*. (2016), Crowe and Williams (2012). Postpartum uterine infection and inflammation perturb fertility through lengthening the calving interval, increasing the number of services per conception and reducing calving rate. Moreover, cows with endometritis spend less time at peak lactation. While uterine inflammation is a physiological event, sustained inflammation beyond 21 days postpartum (DPP) reduces profitability. Here, we hypothesize that the inflammatory signature of cervico-vaginal mucus (CVM) reflects endometrial inflammation and may provide a useful source of biomarkers to prognose clinical endometritis in dairy cows.

**Material and methods** CVM was collected from 20 dairy cows (10 with clinical endometritis and 10 healthy) using gloved hand at 7 and by Metricheck at 21 DPP. Cytological smears were prepared from CVM and stained with Diff Quick. Polymorphonuclear (PMN), mononuclear leukocyte and epithelial cells were counted, total protein levels were estimated using BCA assay and levels of IL-1β, IL-6, IL-8, serum amyloid A (SAA), haptoglobin (Hp) and C5b were analysed by ELISA in CVM. Statistical analyses were performed using GraphPad® Prism 5. Assuming non-normality with D'Agostino-Pearson omnibus test, animal groups were compared with Mann-Whitney test and a Wilcoxon paired test was used to compare the results within the group between 7 and 21 DPP. Results are presented as mean ± standard error of mean (SEM) and were considered statistically significant at P-value < 0.05.

**Results** PMN numbers were consistently high 7 and 21 DPP in CVM from all animals, but were greater in cows with clinical endometritis compared with healthy cows 21 DPP. In contrast, epithelial cell percentages were greater in healthy cows than in cows with clinical endometritis 21 DPP. Total protein levels decreased significantly in CVM from healthy cows and were lower than in cows with clinical endometritis 21 DPP. All inflammatory biomarkers except C5b, remained high in cows with clinical endometritis from 7 to 21 DPP, indicating sustained and chronic endometrial inflammation. IL1β, IL-6, IL-8 and Hp levels were higher in CVM from cows with clinical endometritis compared to healthy cows 21 DPP. Interestingly IL-1β levels were raised in affected cows but not in healthy ones 7 DPP suggesting that early measurement of IL-1β levels might provide a useful predictive marker of clinical endometritis. In contrast, SAA and C5b levels were increased in healthy cows 21 DPP, compared to affected cows suggesting that these acute phase proteins might have an anti-inflammatory role.

**Conclusion** This study shows that CVM is informative if used to measure cytokines, chemokines, acute phase proteins and complement components to monitor uterine inflammation postpartum. While elevated IL-1β levels 7 DPP are a key characteristic feature of the trajectory associated with subsequent disease, SAA and C5b may prevent endometritis through mediating tissue remodelling, and resolving inflammation.

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**References**

Crowe, M. A. and Williams, E. J. 2012. Journal of Animal Science 90, 1722-1727.
Foldi, J., Kulcsar, M., Pecsi, A., Huyghe, B., de Sa, C., Lohuis, J. A., Cox. P. and Huszenicza, G. 2006. Animal Reproduction Science 96, 265-281.
Gilbert, R. O., Shin, S. T., Guard, C. L., Erb, H. N. and Frajblat, M. 2005. Theriogenology 64, 1879-1888.
Salilew-Wondim, D., Ibrahim, S., Gebremedhn, S., Tesfaye, D., Heppelmann, M., Bollwein, H., Pfarrer, C., Schellander, K. and Hoelker, M. 2016. BMC Genomics 17, 218.

012

# Schmallenberg virus: pathogenicity and teratogenicity in the embryonated chicken egg model

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**Application** These studies demonstrate for the first time that the embryonated chicken egg (ECE) model is a suitable *in vivo* small animal model to study Schmallenberg virus (SBV). Furthermore, these results are consistent with the clinical-pathological findings of natural SBV and Akabane virus (AKAV) infection in ruminants.

**Introduction** Schmallenberg virus (SBV) is a Simbu serogroup Orthobunyavirus that emerged for the first time in north-western Europe during 2011. The virus is now sero-endemic in Europe (Collins *et al*., 2017a) with recent re-emergence (Collins *et al*., 2017b). Both SBV and Akabane virus (AKAV) have similar epidemiology, pathogenesis and clinical signs. *In vivo* research studies on teratogenic viruses in ruminants are expensive and can require a long time to complete. These challenges can be mitigated by using small animal models such as embryonated chicken eggs (ECE). Hence, the aims of this research were to investigate if chicken embryos are susceptible to experimental SBV infection and, if so, to what extent and to compare the pathogenicity and teratogenicity of SBV and AKAV infection in an ECE model.

**Material and methods** The study design and methodology, including the age at which embryos were inoculated and virus inoculum doses used, were derived from previous studies which investigated the pathogenicity of Simbu serogroup Orthobunyaviruses in ECE models (McPhee *et al*., 1984). Two studies were conducted. In Study A, 0.2ml of undiluted cell culture-grown SBV ($10^{6.4}$ $TCID_{50}$/0.2 ml) was inoculated into the yolk sac of chicken embryos at 6 days (n = 43) and 8 days (n = 41) of incubation.In Study B, groups of approximately 40 embryos were infected with SBV (n = 178) or AKAV (n = 177) at virus doses ranging between $10^{2.0}$ and $10^{6.0}$ $TCID_{50}$/0.2 ml at 7 days of incubation. Control embryos in both studies were inoculated with 0.2ml of sterile phosphate buffered saline (PBS). Chicken embryos were incubated at 37°C until day 19 of incubation. when they were euthanised and submitted for necropsy examination. Embryos that died between day 7 and day 18 of incubation were also necropsied. Sterile plain swabs of brain tissue were collected from a sub-sample of 19 day old ECEs (both virus-inoculated and control embryos) at necropsy and tested for either SBV or AKAV RNA using quantitative real-time reverse transcription PCR (qRT-PCR). Chi-square statistical tests were used to compare the proportion of deaths, stunted growth, congenital defects and PCR-positive results between groups of embryos. Fisher's exact test was used when expected frequency values were below 5.

**Results** Mortality was greater in embryos inoculated with SBV at 8-days (76%) compared to 6-days (47%), ($P < 0.01$). The prevalence of stunted growth (6-days: 37%; 8-days: 51%) and musculoskeletal malformations (6-days: 42%; 8-days: 41%), (arthrogryposis, skeletal muscle atrophy, contracted toes, distorted and twisted legs) did not differ between days ($P > 0.05$), however, the prevalence of these findings was significantly higher in virus infected embryos compared to controls. Mortality was greater in embryos inoculated with SBV (31%) compared to AKAV (19%), ($P < 0.05$), suggesting that SBV was more embryo-lethal. However, embryos infected with AKAV had a significantly higher prevalence of stunted growth (SBV: 46%; AKAV: 76%; $P < 0.05$) and musculoskeletal malformations (SBV: 18%; AKAV: 42%; $P < 0.01$), suggesting that AKAV was more teratogenic in this model.

**Conclusion** These studies demonstrate that SBV chicken embryos infected with SBV demonstrated gross abnormalities consistent with congenital Schmallenberg disease as reported in ruminants. When SBV and AKAV were compared, SBV appeared to be more embryo-lethal. However, a significantly higher proportion of embryos infected with AKAV had stunted growth and congenital defects. There was no statistical difference in the prevalence of stunted growth or congenital malformations between embryos inoculated at 6 days or 8 days of incubation.

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**References**

Collins, Á. B., Barrett, D., Doherty, M. L., McDonnell, M., Mee, J. F. 2017b. Transboundary and Emerging Diseases 1–5, https://doi.org/10.1111/tbed.12685
Collins, Á. B., Grant, J., Barrett, D., Doherty, M. L., Mee, J. F. 2017a. Preventive Veterinary Medicine 143, 68-78.
McPhee, D., Parsonson, I., Della-Porta, A., Jarrett, R. 1984. Teratogenicity of Australian Simbu serogroup and some other Bunyaviridae viruses: the embryonated chicken egg as a model. Infection and Immunity 43, 413-420.

013

# Investigating the occurrence and potential mitigation of fluorosis in ruminant livestock along the East African Rift Valley

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**Application** Fluorosis is prevalent in livestock along the Rift Valley, resulting in reduced productivity and increased levels of fluoride in animal products such as meat and milk. Binding fluoride ions within the GI tract may reduce its absorption and retention in the body.

**Introduction** Fluoride concentrations in water along the East African Rift Valley are in excess of the recommended concentration for regular human consumption (Olaka *et al.,* 2016). Naturally occurring fluoride is found in water, soils, crops and animals leading to dental and skeletal fluorosis. Fluorosis severely affects animal productivity and has implications for people and their consumption of animal products. This study examined the occurrence of fluorosis in ruminants along the Rift Valley and the means by which fluoride absorption from the animal's gut can be mitigated, thus reducing the impact on animal production, welfare and human consumption.

**Material and methods** Locations within Kenya and Tanzania were chosen as the main study sites along the Rift Valley. Four-hundred and eighty-seven questionnaires were conducted by the research team on households in the Nakuru and Arumeru regions in Kenya and Tanzania, respectively. Questionnaires included details on numbers of livestock owned, slaughtered and sold per household and the predominant water source provided for the animals. Additionally, dental fluorosis surveys were performed on ruminants. All animals were handled to the required animal welfare standards according to the Home Office competency licence required for scientific investigations within the U.K. Once restrained, the animal's lower lip was pulled down and the eight front teeth were scored according to Dean's Dental Fluorosis Index. The $Chi^2$ statistical test was used to analyse the data collected for dental fluorosis. Fur and hoof clippings were sampled along with urine, faeces and milk to determine fluoride concentration in order to understand its level of retention within the body and its prevalence in the food chain.

**Results** All animals observed exhibited signs of dental fluorosis (Fig. 2), with scores of 1 (very mild) and 2 (mild) most predominant (Fig. 1). In Tanzania, questionnaire data revealed that river water was the most common water source for livestock, whilst in Kenya boreholes were used more often. *In vitro* trials to screen a number of additives for their ability to complex fluoride in rumen fluid are being carried out.

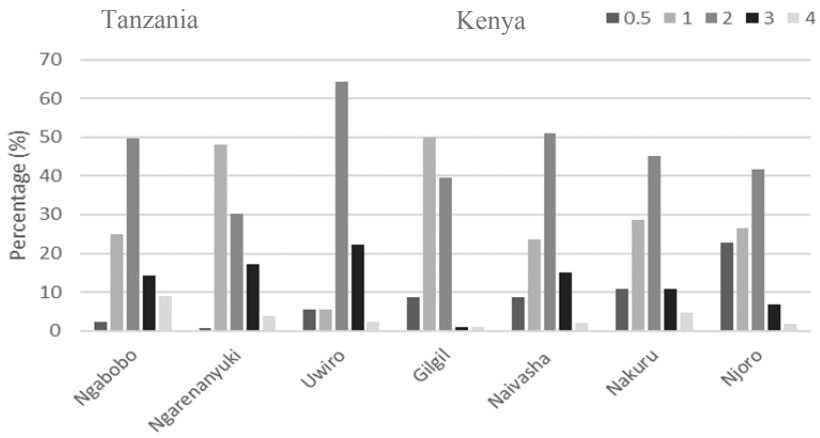


**Figure 1** Percentage of Dean's dental fluorosis scores in each region

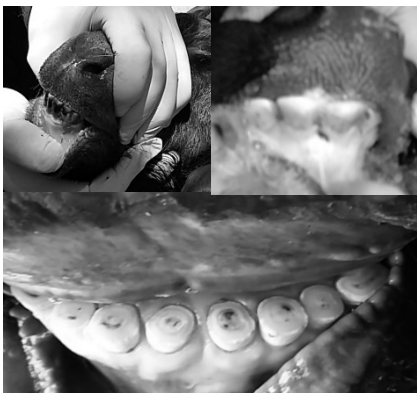

**Figure 2** Photos of dental fluorosis in ruminants

**Conclusion** Significant levels of dental fluorosis are found within ruminant livestock along the East African Rift Valley. Whether the animals are ingesting fluoride predominantly from water, feed or milk is not yet known, but it is likely to be a combination of these. Further analysis of animal tissue samples is being conducted to determine fluoride retention within the body. Solutions to this problem involving additives that form complexes with fluoride within the rumen are being tested.

**Acknowledgements** Special thanks to the EU2020 grant provider for funding the FLOWERED project; to farmers from the study sites and to staff at Eldoret University and NM-AIST.

**References**

Olaka, L.A., Wilke, F.D., Olago, D.O., Odada, E.O., Mulch, A. and Musolff, A. 2016. Groundwater fluoride enrichment in an active rift setting: Central Kenya Rift case study. Science of the Total Environment 545, 641-653

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**014**

# Why the use of preventative treatment for livestock diseases is not always cost-effective: a case-study of an economic game theory model of sheep scab prophylaxis

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**Application** Economic models can help demonstrate whether preventative treatment for a livestock disease is a cost-effective strategy in relation to the treatment choices of a neighbouring farm (Nixon *et al*., 2017).

**Introduction** Infectious diseases in livestock cause losses of millions of pounds per year. Using prophylaxis to target individual diseases can reduce disease incidence (Hosie & Clark 2007). However, this can also lead to unnecessary costs for a farmer (Nixon *et al*., 2017). Therefore, the relative costs and risks of a disease should be examined before making prophylaxis decisions, as demonstrated by Nixon *et al*. (2017) in a game theory model of sheep scab.

**Material and methods** Data from the literature were used (Nixon *et al*. 2017) to estimate cost and risk parameters for a deterministic game theory model, which recommends the optimum sheep scab prophylaxis strategy for a farmer in relation to the strategy of their neighbour. Two prophylaxis treatments were included; injection of long-acting macrocyclic lactones (MLs) and organophosphate dipping. As regional costs and risks differ, the model was run separately for upland farms (northern England, Scotland, Wales) and lowland farms (rest of England). One-at-a-time sensitivity analyses were used on the baseline scab risk parameter (intervals of 0.005, range 0-50%), overall prevention cost and the cost of prophylactic treatment product alone (intervals of £0.05, range £0-£2) to identify when the optimum strategy would change.

**Results** The average cost of sheep scab per year is £35.12 in an upland flock (range £35.01-£35.36) per ewe and her lambs and £40.84 in a lowland flock (range £40.63- £41.02) (Table 3, Nixon *et al*. 2017). Under current costs and risks it is only cost-effective to use prophylaxis in the uplands of Great Britain and only when dipping, regardless of the prophylaxis choices of a neighbour (Figure 2, Nixon *et al*. 2017). However, under different costs and risk of scab, it can become cost-effective to use prophylaxis (Table 1).

**Table 1** Current costs and risk of sheep scab in upland and lowland farms in Great Britain and the values at which it becomes cost-effective to use prophylaxis (either injection of long-acting macrocyclic lactones or an organophosphate dip) (Adapted from Tables 1 & 2 and Figures 3 & 4 in Nixon *et al*.(2017)
**At the range of treatment costs examined (£0-£2), the optimum strategy to not use preventative treatment was unaffected.*

| Model environment | Current risk of sheep scab | Risk of scab at which it becomes cost-effective to use prophylaxis | Current cost of aggregate prevention costs for sheep scab (per ewe and her lambs) | Cost at which it becomes profitable to use prophylaxis | Current cost of treatment product for sheep scab (per ewe and her lambs | Cost at which it becomes profitable to use prophylaxis |
|---|---|---|---|---|---|---|
| Lowland inject | 5.2% | >=16% | £1.82 | <= £0.45 | £1.42 | n/a* |
| Lowland dip | 5.2% | >= 10.5% | £1.36 | <= £0.70 | £0.39 | n/a* |
| Upland inject | 13.9% | >=20.5% | £1.77 | <£1.25 | £1.37 | <= £0.85 |
| Upland dip | 13.9% | >=13% | £1.34 | <£1.45 | £0.39 | <= £0.50 |

**Conclusion** The case study of Nixon *et al. (*2017) suggests that under current costs and risks of sheep scab, treating preventatively is only cost-effective in the uplands of Great Britain and when using an OP dip. However, in areas where scab prevalence is higher than average, preventative treatment could be profitable. In other areas, subsidies and penalties would need to be introduced if eradication of scab is desired. Economic game theory models could be developed to answer similar treatment questions about other livestock diseases.

**Acknowledgements** This work was funded by the Biotechnology and Biological Sciences Research Council and the South West Biosciences Doctoral Training Partnership

**References**

Hosie B, Clark S 2007. In Practice 29, 246-254.
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**015**

# Investigation of the relationship between quarter somatic cell count and udder skin surface temperature of dairy cows measured by infrared thermography

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**Application** Results from the current study could aid in the implementation of non-invasive and user friendly technologies which could be used to automatically detect high somatic cell count (SCC) in dairy cows.

**Introduction** An elevation in SCC in the milk of a cow is a common method of detecting infection in the mammary gland and the impact on farm profit is well recognised (Geary *et al*., 2011). Currently, only parlours which use separate milk lines for each teat cup have the capability to automatically measure the SCC of each quarter on a daily basis. Therefore alternative methods are required to gather udder health data on farms. Infrared thermography (IRT) is a non-invasive technology which can estimate the temperature of an object and has been used in research to detect disease (Polat *et al*., 2010). The objective of the present study was to investigate the use of a plethora of descriptive temperature parameters (DTPs) from udder IRT to detect high SCC in dairy cows on a daily basis.

**Material and methods** To quantify the relationship between udder skin surface temperature (USST) and SCC over time, data was recorded daily on the same 14 Holstein-Friesian cows, at evening (15:00 to 16:00) milking over a two-month period. All udders were shaven before and half way through the experimental period. Thermal images were captured each day by the same operator using a FLIR T430sc thermal camera (FLIR SYSTEMS Inc., Stockholm, Sweden). Thereafter milk samples were taken from each quarter to be analysed for SCC. The distance the animals walked to the parlour every day, the number of days since the animal was shaved, as well as environmental factors (ambient temperature, relative humidity, wind speed and rainfall) was also recorded. Image analysis and temperature extraction was undertaken using the procedures outlined by Byrne *et al*. (2017). Several DTPs were generated including but not limited to; minimum, average and maximum USST. To quantify the association between environmental factors and the DTPs, a stepwise selection of the environmental factors using a multiple regression model in PROC REG (SAS Institute, 2010) was performed on each DTP as the dependent variable. To investigate the usefulness of each DTP as a predictor of SCC, a stepwise selection using a linear regression model in PROC REG (SAS Institute, 2010) was undertaken for each individual DTP on an individual cow level. For all calculations, SCC was transformed using the natural log. As part of further analysis, environmental factors were used to predict USST across time using linear mixed model polynomials in PROC MIXED (SAS Institute, 2010), where quarter nested within cow was included as a random effect. To predict healthy USST on a single day (day 0) for each cow, data from only 5 days previous were used. Similar techniques were used to predict SCC.

**Results** The DTPs with the greatest proportion of total variation accounted for by environmental factors were: average (53%) and maximum USST (50%). None of the DTPs had a significant relationship with the SCC of every animal in the experiment ($P<0.05$). The DTPs accounted for a small proportion of the variation in SCC (<24%), however, every regression co-efficient was not positive. The maximum USST was chosen for further analysis as it was one of the DTPs significantly associated with the SCC of the most cows (10/14). Animal activity and hair on the udder did not have a significant effect ($P<0.01$) on maximum USST. With ambient temperature as the only independent variable, a root mean square error of 0.23°C and a coefficient of determination of 0.72 were achieved when predicting maximum USST. The use of maximum USST did not improve the root mean square error or coefficient of determination value of the prediction of SCC.

**Conclusion** This current study demonstrates that maximum USST can be predicted with a root mean square error of 0.23°C using a single environmental factor (ambient temperature), however, USST could not be used to directly predict SCC in the present study. As some of the variation of SCC can be accounted for by USST, future studies may implement the maximum USST prediction model presented in this current study on a larger longitudinal dataset to attempt to relate deviations in temperature with large increases in SCC.

**Acknowledgements** The authors gratefully acknowledge funding from the Irish Department of Agriculture, Food and Marine Research Stimulus Fund GENCOST (11/S/133).

**References**

Byrne, D. T., Berry, D. P., Esmonde, H. and McHugh, N. 2017. Journal of Animal Science 95, 970-979.
Geary, U., Begley. N., McCoy. F., O'Brien. B., O'Grady. L., Shalloo. L. 2011. Estimating the impact of mastitis on the profitability of Irish dairy farms. In: Hogeveen H., Lam T.J.G.M. (eds) Udder Health and Communication. Wageningen Academic Publishers, Wageningen
Polat, B., Colak, A., Cengiz, M., Yanmaz, L., Oral, H., Bastan, A., Kaya, S., and Hayirli, A. 2010. Journal of Dairy Science 93, 3525-3532.

016

# Evaluation of the communication methods used to disseminate the findings of a dairy research farm for extension purposes

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**Application** This study identified the importance of how research, advisory and education centres in Teagasc need to work together to ensure all stakeholders (students, farmers, industry) are reaping the rewards as such of all the work carried out.

**Introduction** There is a feeling that good research can be often lowered by poor communication of the findings to the clients (Sulaiman, *et al*., 2012). This transferring of research findings to suitable end users is important and should not be underestimated (Department of Agriculture, Food and the Marine, 2011). Extension workers alone are not sufficient tools to disseminate agricultural information (LawalIro, *et al*., 2014). This study sought to evaluate the communication methods used to disseminate the findings of a dairy research farm for extension purposes. The study was carried out from Ballyhaise Agricultural College, Co. Cavan.

**Material and methods** A cross-sectional research design was applied for this study using both quantitative and qualitative approaches. Quantitative research was carried out on a sample of 100 dairy farmers who were suppliers of Lakeland Dairies and LacPatrick at the time, to investigate their access and usage of the Ballyhaise research findings. The farmers were chosen from a list of suppliers provided by both Lakeland Dairies and LacPatrick which included a mixture of scale and production levels. The lists were cross-referenced with a Teagasc client list to ensure that both Teagasc clients and non-clients were included in the study. A cross-sectional survey design was used which involved the collection of data from a number of sources at a single point in time and also allowed the research to look at numerous things at once.
A survey was carried out with the Advanced Certificate in Dairy Herd Management students in Ballyhaise in order to identify if they were benefiting from the research farm and identify what communication methods would be best suited to them. This survey was also administered to the Advanced Certificate in Dairy Herd Management students in all of the other agricultural colleges which provided this course. This included colleges which do not have a research farm in order to establish if they were at a disadvantage compared to the students in a college with a research farm.
Semi-structured interviews of dairy advisors based in counties Cavan and Monaghan were carried out to discuss the findings from the farmer survey. The aim of these interviews was to get an insight into the advisors views and opinions regarding the dissemination of the findings of the Ballyhaise dairy research farm to farmers. It was also hoped that the interviews would help identify the needs of dairy advisors in relation to the dissemination of these findings.
Semi-structured interviews were also carried out on dairy teachers from each of the colleges that participated in the student survey. The aim of these interviews was to get an insight into the teacher's views and opinions regarding the dissemination of dairy research findings to students and how the teachers could be aided in the dissemination of these findings.
Potential dissemination methods which were identified in the Literature Review and Surveys were trialled to assess their effectiveness in disseminating information from the Ballyhaise Dairy Research Farm. These were Twitter and YouTube.

**Results** This study found that current communication methods such as discussion group meetings and open days were being used to disseminate the findings of dairy research farms. The study identified the following methods which could be used on a more frequent basis; email, text message, smartphone app, Twitter and YouTube videos. The study also found that farmers realise the relevance of Ballyhaise to them but are not aware of research being conducted. Results indicate that students on research farms do not have a greater understanding of research results in comparison to those who are not. Results highlight the requirements and support that advisers and teachers will need if they are to successfully disseminate research findings to farmers and students.

**Conclusion** This study encourages the dissemination of research findings in the agricultural industry as a whole, not just within Teagasc.

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**References**

Department of Agriculture, Food and the Marine, 2011. Stimulating Sustainable Agricultural Production through Research and Innovation (SSAPRI), Dublin: DAFM.
LawalIro, S., Boadi, B. Y., Oladokun, O. and Kalusopa, T., 2014. The Generation and Dissemination of Agricultural Information to Farmers in Nigeria: A Review. IOSR Journal of Agriculture and Veterinary Science 7(2), 102-111.
Sulaiman, V. R. *et al*., 2012. Necessary, But Not Sufficient: Critiquing the Role of Information and Communication Technology in Putting Knowledge into Use. The Journal of Agricultural Education and Extension 18(4), 331-346.

**017**

# Effect of toasting time and inclusion rate on rapeseed meal digestibility for broilers

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**Application** The crushing process of rapeseed involves a step of hydrothermal treatment needed to remove solvent from the meal. This treatment can alter the protein and amino acids digestibility, in particular the lysine digestibility.

**Introduction** Rapeseed meal represents a possible alternative for partial substitution of soya meal in monogastric feed (Kozlowski and Jeroch, 2014). In order to enhance the nutritional value of rapeseed meal for poultry, the effect of hydrothermal treatments should be better understood and controlled. The aim of this study was to assess the effect of toasting time and inclusion rate on the digestibility of energy, protein and amino acids of rapeseed meal.

**Material and methods** The rapeseed meal samples were prepared at OLEAD, the French technological platform for protein and oilseed crops. A batch of rapeseed was cold pressed and submitted to solvent extraction to obtain a rapeseed cake. The cake was then submitted to desolventization through continuous indirect steam pressure. Toasting was performed with direct steam addition, at a temperature of 100-110 °C, during 120 minutes and sampling every 20 minutes.
A control diet was formulated with a base containing wheat, corn, soybean meal, soy oil and a premix. Fourteen experimental diets were formulated from seven processed rapeseed meals (toasting time of 0, 20, 40, 60, 80, 100, 120 minutes) incorporated at two inclusion rates (15 and 30 %) with the base and premix. Digestibility was evaluated with growing broilers (Ross PM3) according the method of Bourdillon *et al.* (1990). From hatching to day 17, the broilers were fed a common standard diet. Then they were divided in 15 groups of 8 birds with similar weight and placed in individual cages. An adaptation period with experimental diets lasts from d17 to d22 follow up by a balance period of three days with total collect of excreta. Rapeseed meals and diets were evaluated for dry matter, gross energy and crude protein. Freeze-dried and ground excreta were evaluated for gross energy and crude protein. Amino acids determinations were performed on rapeseed meals, diets and 3 pools of excreta for the control diet and experimental treatment.
Apparent metabolisable energy corrected for nitrogen retention (AMEn) and the digestibility coefficients of protein and amino acids were calculated by the substitution method with the control diet.

**Results** The energy and protein digestibility of rapeseed meal samples are lower when measured with a 30 % inclusion rate. Mean AMEn values are 2254 and 1803 kcal/kg DM at inclusion rates of 15 and 30 % respectively. Mean protein digestibility coefficients are 71.6 and 67.6 % at inclusion rates of 15 and 30 % respectively. The toasting time has a negative effect on AMEn values of rapeseed meal from 40 min for samples included at 15 % and from 20 min of process for samples included at 30 % in experimental diets. Significant and negative correlations are observed between toasting time and protein digestibility coefficients ($R^2 = 0.83$ and $R^2 = 0.77$ for 15 and 30 % inclusion rates respectively).
Toasting decreased the lysine content of rapeseed meal from 2.3 to 2.0 % DM. The digestibility of all amino acids is decreased except for tryptophan. The digestibility of lysine and cysteine are particularly sensitive to toasting, coefficients decreased from 83.5 to 70.9 % for lysine and from 81.1 to 47.9 % for cysteine from 0 to 120 minutes of toasting.

**Table 1** Energy and protein digestibility of rapeseed meal samples

| Inclusion rate (%) | 15 | | | | | | | 30 | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Toasting time (min) | 0 | 20 | 40 | 60 | 80 | 100 | 120 | 0 | 20 | 40 | 60 | 80 | 100 | 120 |
| AMEn (kcal/kg DM) | 2356 | 2468 | 2175 | 2246 | 2236 | 2273 | 2022 | 1964 | 1774 | 1741 | 1864 | 1769 | 1838 | 1673 |
| Protein digestibility (%) | 73.0 | 73.6 | 71.6 | 72.5 | 70.6 | 70.5 | 69.4 | 70.6 | 67.9 | 68.7 | 68.0 | 65.7 | 67.2 | 65.0 |

**Conclusion** The 30 % inclusion rate of rapeseed meal leads to an under estimation of energy and protein digestibility. The duration of toasting has a negative effect on energy, protein and amino acids digestibility and seems to increase the variability of *in vivo* measurements.

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**References**

Bourdillon, A., Carré, B., Conan, L., Francesch, M., Fuentes, M., Huyghebaert, G., Janssen, W.M., Leclercq, B., Lessire, M., McNab, J., Rigoni, M. and Wiseman, J. 1990. British Poultry Science 31, 567-576.
Kozlowski, K. and Jeroch, H. 2014. Annals of Animal Science 14 (2), 245-256.

**018**

# Exogenous feed enzymes as a means of increasing feed efficiency in grower-finisher pigs: a systematic review and meta-analysis

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**Application** The exogenous enzymes that most consistently improved gain to feed (G:F) of grow-finisher pigs were mannanase and multi-enzyme complexes (C). Enzyme supplementation was most likely to be efficacious when used with corn-based diets. The G:F response to enzyme supplementation was influenced by dietary energy level.

**Introduction** Supplementing feed with exogenous enzymes has been suggested as a strategy to improve feed efficiency in grower-finisher pigs. Many studies have tested the effect of dietary enzyme supplementation; however, the ability of exogenous enzymes to improve feed efficiency is not always consistent. A systematic review and a meta-analysis were conducted to determine which exogenous enzymes are most consistent in improving feed efficiency and to investigate the effect of diet formulation on the response to in-feed enzyme supplementation in grower-finisher pigs.

**Material and methods** A systematic literature review was conducted using the on-line database "Web of Science™" using the keywords "enzyme name", "growth" and "pig". The enzymes included in the literature search were xylanase (X), xylanase+β-glucanase (XB), mannanase (M) protease (P), cellulase (CEL) and α-galactosidase (Gal). A database including the G:F of the experimental control group ($G:F_{ctr}$) and the G:F of the enzyme-supplemented group ($G:F_{enzy}$) together with dietary composition from each selected study was prepared. The metafor package in R (R Core Team, 2015) was used to conduct the meta-analysis (Viechtbauer, 2010) and generate forest plots. Mean difference (MD) was the size estimate effect used in the meta-analysis and was calculated by subtracting the mean G:F of the control group from the supplemented group ($MD= G:F_{enzy} - G:F_{con}$) and tested in a mixed model with enzyme type, ingredient composition and dietary energy level following similar methodology to Bougouin *et al*. (2014).

**Results** Gain to feed was improved in 56, remained un-changed in 47 and deteriorated in 12 of the studies, in response to enzyme supplementation. Gain to feed was improved when M (+1.19 g gain/100 g feed, P<0.001) or C (+2.21 g gain/100 g feed, P<0.001) were supplemented in the diet. Overall, enzyme supplementation increased G:F when supplemented to corn (+1.21 g gain/100 g feed, P<0.001) or wheat (+1.22 g gain/100 g feed, P<0.001) based diets but no response to enzyme supplementation was observed when supplemented to barley, rye or sorghum diets (P>0.10). Diets with corn as the main cereal source were more likely to have increased G:F due to X, XB, M and P supplementation whereas G:F increased only in wheat based diets supplemented with M and XB (Figure 1). The most consistent improvement in G:F with multi-enzyme cocktail supplementation was found when M (P<0.01) and/or CEL (P<0.05) were included in the complex. The G:F in response to enzyme supplementation was influenced by dietary energy and protein level (P<0.01). When enzymes were supplemented to diets formulated below energy recommendations (Blas *et al*., 2013), the G:F response was increased compared to diets formulated above these recommendations (+1.57 *vs.* +0.91 g gain/100 g feed, P<0.001).

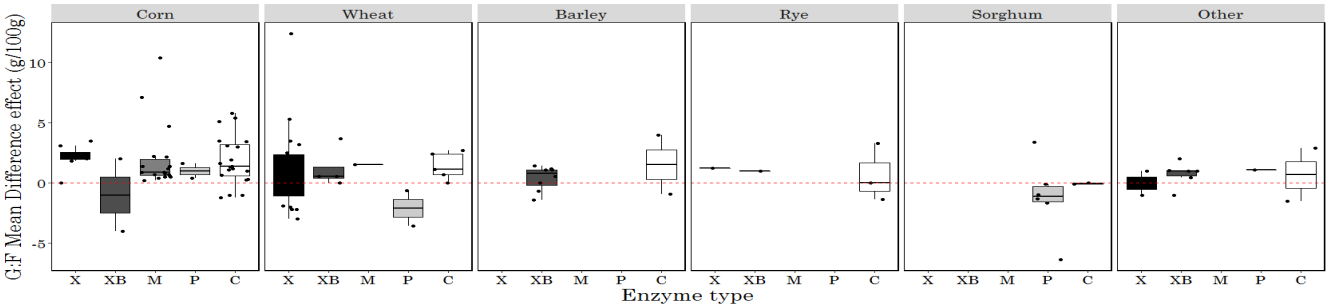


**Figure 1** Effect of cereal source on G:F mean difference when enzymes were supplemented to grow-finisher diets.

**Conclusion** The most consistent improvement in G:F in grower-finisher pigs resulted from M and C supplementation. The response to supplementation was influenced by the ingredient composition and energy density of the diet.

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**References**

Blas, C., Gasa, J. and Mateos, G.C. 2013. FEDNA, Spanish association for the development of animal nutrition, 24-40.
Bougouin, A., Appuhamy, J.A., Kebreab, E., Dijkstra, J., Kwakkel, R.P., 2014. Journal of Poultry Science 93(8), 1981-1992.
R Core Team. 2015. R Fundation for Statistical Computing, Vienna, Austria.
Viechtbauer, W. 2010. Journal of Statitstical Software 36, 3.

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019

# Effects of feeding varying levels of crude protein with or without exogenous phytase on growth and slurry gas emissions of growing pigs

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**Application** There have been growing concerns about greenhouse gas (GHG) emissions arising from intensive swine production. Nutritional manipulation has been explored to reduce such emissions without affecting growth performance.

**Introduction** The growth in the world's population is expected to increase the demand for animal products including pork, consequently increase GHG emissions, including methane ($CH_4$), carbon dioxide ($CO_2$), nitrous oxide ($N_2O$) and hydrogen sulphide ($H_2S$). To combat the issue of global warming, one of the mitigation strategies that can reduce GHG emissions in swine production is feed management. This study was carried out to evaluate the effects of dietary crude protein (CP) levels and phytase inclusion on growth and manure gaseous emissions in growing pigs.

**Material and methods** Six groups of five Large White pigs, individually housed, were randomly allocated to one of six dietary treatments with three dietary CP levels (14, 16 and 18 %) each with or without phytase of 5000u activity in a 3×2 factorial arrangement. Initial body weight averaged 17.80±0.10kg. Crystalline L-lysine, DL-methionine, L-threonine and L-tryptophan were included in the diets to meet amino acid requirements (NRC, 2012). Pigs were fed twice daily (morning and evening) and water was provided *ad libitum* with automatic drinkers for an 8 week growth trial, in which average daily weight gain (DWG), average daily feed intake (DFI) and feed conversion ratio (DFI/DWG) were determined. Fresh faecal and urine samples were taken from 4 random pigs per treatment for three consecutive days using same pigs at the end of week 8. Samples were stored in the freezer and later mixed to make slurry for each pig with a 40:60 faeces:urine ratio (Marszalek *et al*., 2014). Twenty four, 500 ml bottles fitted with one litre Tedlar bags were filled with 250 mls of slurry samples. The samples were incubated at 37°C for 30 days, after which gas emissions ($CH_4$, $CO_2$, residual nitrogen ($RN_2$) and $H_2S$) were measured using Geotech Biogas 5000. Data were analysed in a 3×2 factorial arrangement with n=5 for growth performance and n=4 for gas emissions (GenStat 12.1). Treatment means were separated using Tukeys test at $P < 0.05$.

**Results** Table 1 shows that DWG increased with increased CP level, whilst dietary treatments did not affect DFI. However, FCR was reduced with increased levels of CP and with phytase supplementation. CP and phytase interacted on methane emission ($P<0.001$): methane production increased with increased dietary CP, though at a smaller magnitude in the presence of phytase. Furthermore, phytase reduced slurry $CO_2$, $RN_2$ and $H_2S$ production, whilst increasing dietary protein level increased $RN_2$ and $H_2S$ production without impacting on $CO_2$ production.

**Table 1** Crude protein levels and phytase effects on average daily weight gain (DWG), daily feed intake (DFI) feed conversion ratio (FCR), and slurry emission of $CH_4$, $CO_2$, residual nitrogen ($RN_2$) and $H_2S$

| Phytase (P) | No | | | Yes | | | | P-values | | |
|---|---|---|---|---|---|---|---|---|---|---|
| CP | 14 | 16 | 18 | 14 | 16 | 18 | s.e.d. | CP | P | CP×P |
| DWG (g) | $452^{d}$ | $636^{abc}$ | $698^{ab}$ | $543^{cd}$ | $598^{bc}$ | $738^{a}$ | 2.5 | 0.001 | 0.245 | 0.148 |
| DFI (g) | 1430 | 1623 | 1632 | 1548 | 1562 | 1627 | 106 | 0.175 | 0.779 | 0.491 |
| FCR | $3.19^{a}$ | $2.56^{bc}$ | $2.34^{cd}$ | $2.88^{ab}$ | $2.62^{bc}$ | $2.20^{d}$ | 0.10 | 0.001 | 0.044 | 0.055 |
| $CH_4$ (%) | $0.28^{bc}$ | $1.03^{bc}$ | $4.08^{a}$ | $0.13^{c}$ | $0.23^{bc}$ | $1.43^{b}$ | 0.41 | 0.001 | 0.001 | 0.001 |
| $CO_2$ (%) | 18.9 | 18.8 | 18.2 | 14.5 | 14.7 | 14.3 | 1.82 | 0.927 | 0.001 | 0.972 |
| $RN_2$ (%) | $33.5^{cd}$ | $50.0^{ab}$ | $56.5^{a}$ | $27.3^{d}$ | $35.8^{cd}$ | $42.9^{bc}$ | 3.70 | 0.001 | 0.001 | 0.257 |
| $H_2S$ (ppm) | $50.5^{b}$ | $55.5^{b}$ | $117.4^{a}$ | $35.0^{b}$ | $51.8^{b}$ | $64.3^{b}$ | 16.53 | 0.002 | 0.021 | 0.116 |

**Conclusion** Increasing protein levels increased DWG and reduced FCR, but also increased $CH_4$, $RN_2$ and $H_2S$ emissions. Ongoing modelling activities on total slurry production per unit DWG are expected to inform the impact of increase CP levels on emission intensity. Meanwhile, phytase supplementation improved FCR and reduced gas emissions, supporting the view that the use of phytase in swine production can assist to reduce the environmental footprint of pork production.

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**References**

Marszalek, M., Kowalski, Z. and Makara, A. 2014. Technical Transaction Chemistry 1-Ch, 81-91.
National Research Council. 2012. Eleventh Revised Edition. Washington, DC.

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020

# Effect of β-xylanase and β-glucanase in wheat based diets achieved through different agronomical conditions on nutrient digestibility and growth performance in young pigs

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**Application** The addition of exogenous enzyme (β-xylanase and β-glucanase) to wheat grain naturally contaminated with mycotoxins will improve growth performance and nutrient digestibility in young pigs.

**Introduction** Wheat is one of the major feed ingredients included in swine diets but also one of the most variable in composition. Environmental conditions under which wheat is grown can exacerbate these differences, affecting factors such as starch content, cell wall carbohydrates (Longstaff and McNab, 1986) and mycotoxin levels (Edwards, 2004). A large proportion of the variation in feeding value of cereal is due to the difference in non-starch polysaccharides (NSP) which can lead to increase digesta viscosity and reduced nutrient digestibility. Pigs do not produce sufficient endogenous enzymes that efficiently degrade NSP due to the structure of their gastrointestinal tract. Also, swine are among the most sensitive species to mycotoxins with negative effects on feed intake, weight gain and faecal consistency being reported in pigs. With the ban on antibiotic growth promoters (AGP) in animal diets the need to provide pigs with good quality mycotoxin free grain is of vital importance. As an alternative to AGP the inclusion of exogenous enzyme may help to overcome the negative effect of feeding poor quality grains. The objective of the present study was to examine the effect of feeding a wheat based diet varying in quality, achieved through different agronomical conditions, with or without the supplementation of a β-glucanase and β-xylanase enzyme mix on young pigs.

**Material and methods** The experiment was designed as a 3 × 2 factorial design and approved under University College Dublin Animal Ethics Committee (AREC-13-56-O'Doherty). Ninety-six (11.6 kg SD 0.97) pigs were assigned to one of the six dietary treatments: (T1) low quality wheat diet, (T2) low quality wheat diet and an enzyme supplement, (T3) medium quality wheat diet, (T4) medium quality wheat diet and an enzyme supplement, (T5) high quality wheat diet and (T6) high quality wheat diet and an enzyme supplement. The parameter categories assessed included growth performance, nutrient digestibility and faecal consistency. To obtain wheat (cv. *JB Diego*) grain of different levels of chemical composition (starch, fibre, protein etc.) (low, medium and high), two blocks (A and B) of wheat were established and harvested in the 2014 season. To return wheat grain of high quality (high starch and low fibre), Block A had a relatively early sowing date (25$^{th}$ October 2013) and followed the recommended winter wheat husbandry practices. Block B (low starch and high fibre) had a delayed sowing date (15$^{th}$ November 2013) and subsequently, delayed husbandry practices and harvest date. The medium quality wheat grain was made up of a 50% mixture of high and a 50% mixture of the low quality grain. The inclusion rate of wheat was 500 g/kg. The diets were formulated to contain similar levels of net energy (10.4 MJ/kg) and standardised ileal digestible lysine (12 g/kg). Celite (300 mg/kg) was added to the feed during the milling process in order to measure the coefficient of apparent total tract digestibility (CATTD) of nutrients using the acid insoluble ash technique.

**Results** The low quality wheat grain had a lower hectolitre weight compared to the medium and high quality wheat (66.4 vs. 70.2 vs. 73.8 g/kg hL respectively). The level of zearalenone (0.249 vs. 0.142 vs. 0.036 mg/kg), aflatoxin (0.004 vs. 0.004 vs. 0.003 mg/kg) and ochratoxin (0.30 vs. 0.17 vs. 0.005 mg/kg) present in the low quality wheat grain were greater than that in the medium and high quality wheat grain, respectively. Pigs offered the low quality wheat diet had a lower average daily gain (ADG) (0.54 vs. 0.65 vs. 0.66 kg/day, SEM 0.012, $P<0.001$), a lower gain to feed (G:F) ratio (0.46 vs. 0.57 vs. 0.58 kg/kg, SEM 0.011, $P<0.01$) and a higher faecal score (3.0 vs. 2.8 vs. 2.7, SEM 0.045, $P<0.001$) compared to the medium and high quality wheat, respectively. The low quality wheat diet also had a reduced CATTD ($P<0.05$) of nitrogen (N) (0.72 vs. 0.76 vs. 0.77 SEM 0.008) and gross energy (GE) (0.76 vs. 0.79 vs. 0.79, SEM 0.007) compared with pigs offered the medium and high quality wheat diet, respectively. The addition of an enzyme improved the G:F ratio (0.55 vs 0.53 kg/kg, SEM 0.008) and faecal score (2.8 vs. 2.9, SEM 0.04, $P<0.05$) compared to the diets without enzymes.

**Conclusion** In conclusion, this study reports that the higher level of mycotoxins present in the low quality wheat reduced ADG and the CATTD of nutrients in pigs offered this diet. The inclusion of a β-glucanase and β-xylanase enzyme mixed improved G:F ratio and faecal scores.

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**References**

Longstaff, M. and McNab, J. 1986. British Poultry Science 27, 435-449.
Edwards, S. G. 2004. Toxicology letters 153, 29-35.

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021

# Examination of the effects of exogenous fibrolytic enzymes on the release of xylose from different cereal types *in vitro*

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**Application** It may be optimal to assess the feed components of broiler diets to create a bespoke enzyme supplementation protocol to magnify the feed-enzyme response.

**Introduction** Exogenous fibrolytic enzymes are added to livestock feed to degrade non-starch polysaccharides (NSP) thus improving their digestion and nutrient release. Arabinoxylans (AX) are the primary cell wall polysaccharides in plants composed of pentose sugar xylose along with some arabinose. Major feed ingredients in broiler diets include maize, and wheat which vary in their structures and NSP content (Knudsen, 1997). Fibrolytic enzymes can alleviate anti-nutritive factors in feeds, which may allow farmers to use cereals that are currently unpopular in the industry, which is why barley and oats were also selected. An increased understanding of exogenous enzymes' effects on individual cereals is necessary to maximise the economic benefits and the magnitude and frequency of feed-enzyme responses. The aim of this study was to identify the quantity of xylose released from four individual cereals following 72 hour *in vitro* incubation with a range of commercially available fibrolytic enzymes in a solution and temperature analogous to that found in broiler gut.

**Material and methods** The total xylose contents of the four cereals (barley, maize, oats and wheat) was determined by total hydrolysis of their non-cellulosic polysaccharides with Trifluroacetic acid as outlined in the method by Fry (1988). The concentration of xylose was determined using High-Performance Anion-Exchange Chromatography Coupled with Pulsed Electrochemical Detection (HPAEC-PAD) fitted with a CarboPac PA20 Column (Dionex, Thermo Scientific).
For the second part of the experiment, the same four cereals were ground to a 300 micron powder and were subjected to a *in vitro* incubation with four treatments, control, (no exogenous enzymes added), and three commercial enzyme products. The enzymes were administered at the manufacturer's suggested dosage (40-100g/tonne). Enzyme A contained predominantly xylanase activity (160,000 BXU/g). Enzyme B contained predominantly β-glucanase activity (700,000 BU/g) with endo cellulose (165,000 ECU/g) as well as endo- xylanase (190,000 BXU/g). Enzyme C contained predominantly mannase (1,000,000 MNU/g) with β-glucanase (300,000 BU/g) as well as endo- xylanase (200,000 BXU/g). All the cereals were mixed in 50mM sodium citrate buffer (pH 5.2) at 5mg/ml and one of the four treatments, then incubated at 40.7°C in a shaking incubator for 72 hours, as this has been identified as the maximum transient time, due to feed storage in the crop of poultry (Duke *et al*, 1968), following which xylose concentration was determined as described above. Data was analysed using one and two way ANOVA with $P<0.05$ taken as being statistically significant.

**Results** All of the four cereals analysed had similar amounts of xylose present within the whole grains, Barley had the most at 5.22 ± 0.17g/100g, Maize had 4.46 ± 0.14g/100g, Oats had the lowest level, at 3.54 ± 0.06g/100g and Wheat had the second lowest at 3.75 ± 0.22g/100g. There was a significant Cereal x Enzyme interaction ($P<0.001$) in the gut simulation, indicating that the different enzymes had different substrate specificities (Table 1). Surprisingly, there was no release of xylose from the maize sample. For Oats, enzymes A and C released similar small amounts of xylose, but enzyme B did not release any. For barley, all enzymes released some xylose, but enzyme A worked better than enzyme C and B. The greatest release of xylose was seen with a combination of Wheat and enzyme A, where 37.8% of the available xylose was released. Enzyme C also released a similar amount of xylose from Wheat, but enzyme B released about the same as the Control.

**Table 1** The percentage release of xylose after 72h in-vitro after various enzyme treatments

| | Control | Enzyme A | Enzyme B | Enzyme C | Effect of Cereal x Enzyme |
|---|---|---|---|---|---|
| Barley | 3.1 | 11 | 6.5 | 8.7 | |
| Maize | 0 | 0 | 0 | 0 | |
| Oats | 0 | 6 | 0 | 5.1 | |
| Wheat | 6.8 | 37.8 | 8.3 | 32.3 | |
| | | | | | SED = 1.9 |
| | | | | | P <0.001 |

**Conclusion** Supplementation with fibrolytic enzymes *in vitro* releases varying levels of xylose dependant on the type of cereal and the blend of enzymes used. It can be suggested that the optimum enzymes for releasing xylose from cereals, which impact gut health as well as feed digestibility and animal growth, will differ in composition based on cereal choice.
Enzyme efficacy could also be affected by the species and age of the animal used.

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**References**

Knudsen, K.E.B. 1997. Animal feed science and technology 67, 319-338.
Fry, S.C. 1988. The growing plant cell wall: chemical and metabolic analysis, Harlow, Longman Group Limited.
Duke, G., Petrides, G. and Ringer, R. 1968. Poultry Science 47**,** 1356-1364.

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# Barley diets varying in chemical composition with or without enzymes can affect performance, nutrient digestibility and nutrient transporter gene expression in finisher pigs

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**Application** The inclusion of a β-glucanase and β-xylanase enzyme mix to barley based diets, with different chemical composition, achieved through different agronomic conditions will improve overall pig performance.

**Introduction** Cereal grains such as barley and wheat vary in nutrient composition due to different cultivars and agronomic conditions which may in turn affect nutrient digestibility and growth performance in animals (Balls *et al.,* 2013). Adverse agronomic conditions may influence the composition of barley, resulting in a predicted high quality barley grain turning into a lower quality barley grain with greater fibre content and lower digestible energy and net energy content. Non-starch polysaccharides (NSP) such as beta-glucans are present in barley grains which have the potential to limit animal performance. The addition of an enzyme preparation can effectively degrade viscous NSP in the diet leading to an enhanced nutrient utilisation and are likely to increase the availability of free nutrients such as glucose and amino acids.

**Material and methods** The experiment was designed as a 2 x 2 factorial and approved under University College Dublin Animal Ethics Committee (AREC-13-56-O'Doherty). Ninety-six pigs (44.7 kg (SD 4.88)) were assigned to one of four dietary treatments; (T1) low quality barley diet, (T2) low quality barley diet containing an enzyme supplement, (T3) high quality barley diet and (T4) high quality barley diet containing an enzyme supplement. The pigs were grouped in mixed gender (50:50) groups of 12 in 8 pens and the pens were equipped with single space computerised electronic feeders. To obtain barley (cv. *Sebastian*) grain of different levels of quality, two blocks (A and B) of barley were established and harvested in the 2014 season. To yield a high quality barley grain, Block A had an early sowing date (3rd April 2014) and subsequently followed the recommended barley husbandry practices. Block B had a delayed sowing date (16th April 2014) and subsequently delayed husbandry practices and harvest date. The inclusion of barley was 500 g/kg. The diets were formulated to contain similar levels of net energy (9.25 MJ/kg) and standard ileal digestible lysine (8.5 g/kg). Celite (500 mg/kg) was added to the diet in order to measure the coefficient of apparent total tract digestibility (CATTD) using the acid insoluble ash technique. Following slaughter the entire digestive tract of 8 pigs per treatment were removed by blunt dissection. Tissue section ($1cm^2$) from the duodenum and ileum were collected and stored in RNA*later*. Total RNA extraction for the duodenum and ileum; cDNA synthesis; and quantitative real-time PCR were performed as described by Vigors *et al.,* (2016). The mRNA expression profiles of cluster of differentiation (*CD36*), peptide transporter 1 (*PEPT1/SLC15A1*) and sodium-glucose linked transporter 1 (*SGLT1/SLC5A1*) were analysed. Data was analysed as mixed procedures of SAS (SAS version 9.4).

**Results** There was an interaction between barley type and enzyme supplementation on average daily gain (ADG) and average daily feed intake (ADFI) ($P<0.05$). Pigs offered the low quality barley diet supplemented with enzymes had an increase in both ADG and ADFI compared to the low quality barley diet. However, there was no response to enzyme inclusion in the high quality barley diet. There was a barley × enzyme interaction on the CATTD of gross energy (GE) ($P<0.05$). Pigs offered the low quality barley diet with enzymes had a higher CATTD of GE compared to the low quality barley diet. However, the increase in the high quality barley diet with enzyme supplementation was not as great as with the low quality barley diet. There was a barley × enzyme interaction observed for *CD36* in the duodenum and *PEPT1/SLC15A1* and *SGLT1/SLC5A1* in the ileum ($P<0.05$). Pigs offered the high quality barley diet with enzymes upregulated the gene expression of *CD36*, *PEPT1* and *SGLT1* compared to the high quality diet. However the low quality barley diet with enzymes down regulated the gene expression of *CD36*, *PEPT1*and *SGLT1* compared to the low quality barley diet.

**Conclusion** The feeding of a low quality barley diet supplemented with a β-glucanase and β-xylanase enzyme mix improved ADG, ADFI and nutrient digestibility as well as modifying the gene expression of *CD36*, *PEPT1* and *SGLT1* in finisher pigs.

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**References**

Ball, M., Owens, B and McCracken, K. 2013. Asian-Australian Journal of Animal Science 26, 378.
Vigors, S., O'Doherty, J.V., Kelly, A.K, O'Shea, C.J and Sweeney. T. 2016. PLoS ONE 11:e0148145

**023**

# Neonatal energy supplementation of low birth weight piglets does not enhance their health and survival

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**Application** Providing energy supplementation orally (2 ml, 3 h after birth) did not improve survival, temperature, blood glucose content or weight gain of small piglets (<1.1 kg birthweight).

**Introduction** Neonatal piglets have low energy reserves and high energetic requirements (i.e. for thermoregulation, to establish the teat order), and thus must ingest colostrum soon after birth to acquire energy and immunoglobulins. While a normal birth-weight piglet can sustain 15h of heat production with its body reserves, a small piglet (e.g. <1.1 kg birth weight) can only do so for 3h (Mellor and Cockburn, 1986), which places them at higher risk of hypothermia and mortality. Providing supplemental energy to small piglets at birth should help them to spare their body reserves and improve their survival. We investigated the effects of supplementing with different energetic products on pre-weaning survival, 24h weight, blood glucose concentration and piglet body temperature 1h after supplementation.

**Material and methods** This work was granted ethical approval by Teagasc Animal Ethic Committee (TEAC133-2016) and authorised by the Irish Health Product Regulatory Authority (AE19132/P051). Piglets were weighed at birth and, 3h later, those under 1.1 kg were either orally dosed with 2 ml of coconut oil (O, 74 KJ/2ml, n=107 piglets), 2 ml of energetic commercial product (Energyn®, INVESA; E, 71 KJ/2ml , n=101 piglets), or 2 ml of distilled water (W, 0 KJ/2ml , n=100 piglets), or sham-dosed (Control, C, n=97 piglets). Just before treatment, piglets were weighed, an ear vein was punctured to collect and measure blood glucose concentration (subsample: O=45 piglets, E=38 piglets, W=49 piglets, C=44 piglets) and rectal temperature was measured. Rectal temperature was also measured 1h post-treatment, and weight and blood glucose were measured 24h post-treatment. Survival was recorded for the 24h period post-treatment and until weaning. Data were analysed using Generalised Linear Mixed Models in SAS.

**Results** Survival was similar between treatments at 24h post-supplementation and weaning (Table 1). Piglets had similar absolute values of and gains in weight, rectal temperature and blood glucose concentrations (Table 1). However, O piglets had a greater 24h increase in blood glucose concentration, compared to all other treatments ($F_{1,133}$=4.37, P<0.05). Weights tended to differ between treatments at 24h post-supplementation, with W piglets the heaviest ($F_{1,335}$=4.49, P<0.05).

**Table 1** Treatment effects on piglet (mean±S.E.) survival, bodyweight, rectal temperature and blood glucose concentrations

| Variable | Coconut oil | Energy | Water | Control | F-value | P-value |
|---|---|---|---|---|---|---|
| Survival (%) | | | | | | |
| 24h | 89.2 (± 4.0) | 87.2 (± 5.0) | 84.8 (± 5.0) | 90.9 (± 4.0) | 0.6 | 0.71 |
| Pre-weaning | 73.9 (± 5.0) | 70.5 (± 5.0) | 75.0 (± 5.0) | 78.5 (± 5.0) | 0.53 | 0.66 |
| Body weight (g) | | | | | | |
| 24h | 950 (± 20) | 940 (± 20) | 960 (± 20) | 940 (± 20) | 2.19 | 0.09 |
| Gain 24h | 31 (± 7) | 20 (± 7) | 38 (± 8) | 36 (± 7) | 1.46 | 0.23 |
| Rectal temp. (˚C) | | | | | | |
| 1h | 37.7 (± 0.1) | 37.7 (± 0.1) | 37.7 (± 0.1) | 37.7 (± 0.1) | 0.18 | 0.91 |
| Gain 1h | 0.6 (± 0.1) | 0.4 (± 0.1) | 0.5 (± 0.1) | 0.4 (± 0.1) | 0.63 | 0.60 |
| Glucose conc. (mmol/L) | | | | | | |
| 24h | 3.78 (± 0.26) | 3.49 (± 0.28) | 3.62 (± 0.28) | 3.52 (± 0.28) | 0.34 | 0.80 |
| Gain 24h | 1.16 (± 0.48) | 0.13 (± 0.56) | -0.25 (± 0.47) | 0.21 (± 0.5) | 1.7 | 0.17 |

**Conclusion** Oral supplementation with different energy sources after birth did not improve the growth, survival, rectal temperature or blood glucose concentration of small piglets. However, coconut oil increased blood glucose concentration of supplemented piglets to a larger extent than other treatments. This difference was not detectable when piglets given the commercial product were compared to all other treatments, suggesting that there might be a difference in energy uptake between coconut oil and the commercial product.

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**References**

Mellor, D.J., Cockburn, F. 1986. Quarterly Journal of Experimental Physiology 71, 361–379.

# Nitrogen (N) utilization of weaned rabbits fed diets supplemented with varying levels of baker's yeast (*Saccharomyces cerevisiae*)

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**Application** Rabbits fed diets supplemented with varying levels of *Saccharomyces cerevisiae* (SC) had lower Nitrogen (N) balance than those fed the basal diet. Dietary supplementation with SC improved N absorption and may reduce faecal odour and $NH_3$ emission.

**Introduction** *Saccharomyces cerevisiae* (SC), a probiotic, has demonstrated to be a valuable and qualitative growth promoter for feeding livestock. Han *et al.* (2001) reviewed several studies, evaluating the use of feed additives and found that dietary supplementation with probiotics may indirectly lead to a reduction in environmental pollutants from animal manure, by improving feed efficiency and nutrient absorption. Therefore, the present study was undertaken to investigate Nitrogen (N) utilization of weaned rabbits fed diets supplemented with varying levels of SC.

**Material and methods** Forty-eight weaned heterogeneous breeds of rabbits in equal number of males and females, aged between 5 - 6 weeks with average initial live weight of 614.5 ± 57.72 g were procured from a rabbit farm. They were randomly allocated to four groups according to average body weight and sex, with twelve rabbits per group. Group SC0 received a control diet without supplementation of SC, and groups SC2, SC4 and SC6 received the control diet supplemented with SC at the rate of 20, 40, and 60 g per kg for 8 weeks (corresponding to 0, 2, 4 and 6 $\times 10^9$ colony-forming unit/kg, respectively). A commercial baker's yeast, Vahine® (Avignon, Monteux, France), containing SC was used for the dietary supplementation. Proximate analysis of the basal mixture (control diet; SC0) which contained maize, soybean, maize offal, brewer's dried grain, groundnut cake, blood meal, rice offal and bone meal as main ingredients, showed that it contained 16.0% crude protein, 14.1% crude fibre (CF), 3.9% ether extract, and 10.2% ash/kg feed. The diets were not pelleted and were offered with clean fresh water *ad libitum*. Rabbits were individually housed in metabolism cages, which can separate urine from faeces. Feed and faeces samples were analyzed for dry matter (DM), CF, N, crude fat and energy according to the procedure of Yang (1993). Urine samples were analyzed for N according to the procedure of Yang (1993). Data obtained were analyzed using the General Linear Model of SAS (2006) and Duncan New Multiple Range Test of same package was used to test for significant differences among means.

**Results** Rabbits in SC2 absorbed the least N (1.16 g/day), while those in SC4 and SC6 absorbed the most (1.37 g/day and 1.33 g/day) followed by rabbits in SC0 (1.25 g/day). N balance in the control group was significantly higher than those in the SC supplemented groups (P <0.0001).

**Table 1** Nitrogen (N) utilization of weaned rabbits fed diets supplemented with varying levels of SC

| | Dietary treatments | | | | | |
|---|---|---|---|---|---|---|
| | SC0 | SC2 | SC4 | SC6 | s.e.m | P-value |
| N intake (g/day) | $1.69^{b}$ | $1.61^{b}$ | $1.85^{a}$ | $1.87^{a}$ | 0.03 | <0.001 |
| Faecal N (g/day) | 0.44 | 0.45 | 0.53 | 0.50 | 0.02 | NS |
| Urinary N (g/day) | $0.59^{c}$ | $0.84^{b}$ | $0.97^{a}$ | $0.97^{a}$ | 0.04 | <0.001 |
| N absorbed (g/day) | $1.25^{ab}$ | $1.16^{b}$ | $1.33^{a}$ | $1.37^{a}$ | 0.02 | <0.001 |
| N balance (g/day) | $0.66^{a}$ | $0.32^{b}$ | $0.35^{b}$ | $0.39^{b}$ | 0.04 | <0.001 |
| NB as % of N- intake | $38.95^{a}$ | $19.80^{b}$ | $18.86^{b}$ | $20.90^{b}$ | 2.18 | <0.001 |

**Conclusion** The lower N balance in the SC-supplemented groups could be due to dietary probiotics ability to reduce urease activity in the gut. These results suggests that dietary supplementation of rabbits diets with baker's yeast (SC) may reduce faecal odour and $NH_3$ emission.

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**References**

Han, I. K., Lee, J. H., Piao, X. S. and Li, D. 2001. Asian-Australasian Journal of Animal Sciences 14 (3), 432-444.
SAS. 2006. Statistical Analysis System. SAS Institute Inc., Cary, North Carolina, USA
Yang, S. 1993. Feed analysis and feed quality assay. Beijing Agricultural University Press.

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025

# Negative effects of cross-fostering on survival, growth performance and welfare of pigs

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**Application** Cross-fostering should be kept to a minimum and should only be performed early during lactation to avoid negative effects on pig welfare and performance. Late cross-fostering should be avoided at all costs.

**Introduction** Cross-fostering (CF) is a management technique widely used on pig farms to reduce pre-weaning mortality and within litter body weight (BW) variation. CF can be stressful for sows and piglets and should be kept to a minimum. Although no negative consequences on performance of CF up to 5 days after farrowing are reported (van Erp-Van Der Kooij *et al.,* 2003); piglets should be CF between 12-24 h after farrowing (early CF) as the teat order is not established at this time (Heim *et al.,* 2012). In practice, CF continues through lactation such that piglets may be subjected to late CF (i.e. CF > 7 days after farrowing). In such cases it is usually the smallest piglets that are the most likely animals to be CF in the belief that this will help them to 'catch up' and achieve better weaning weights. The objective of this study was to investigate the possible implications of late CF on pig mortality, growth performance, carcass traits and welfare.

**Material and methods** The study had ethical approval from the Teagasc Animal Ethics Committee (TAEC 40/2013). Pigs (n = 1,016) born within one week were classified as either 1) non-CF (NCF) or according to the week they were CF as 2) CF during the first (CFW1) and 3) second or third (CFW2+) weeks of lactation. Pigs were individually weighed and inspected for the presence of tail, ear and body lesions at weaning (7.03 ± 1.61kg) and at the end of the first (4 weeks post-weaning; 12.9±3.03 kg) and second (8 weeks post-weaning; 31.9 ± 5.50 kg) weaner and grower (12 weeks post-weaning; 66.3 ± 9.12 kg) stages. Average daily gain (ADG) was calculated for each stage. Mortality was recorded through to slaughter (20 weeks post-weaning; c. 115 kg). At slaughter cold carcass weight, muscle depth and fat thickness were recorded. Differences were detected between CF treatment for sow parity, birth weight and number of piglets born alive and therefore, a nested case control design was used whereby pigs were matched for these parameters. All data were analysed in *R* v3.4. Each pig was considered the experimental unit. Growth performance and carcass traits were analysed using linear mixed model equations in the *lme4* package. Welfare traits were analysed using binomial logistic regression using the *glm* function with a binomial distribution from the stats package.

**Results** All pigs (n = 1,016) were used to investigate the effect of treatment on the risk of mortality. 106 pigs died during the lactation period and 24, 3 and 14 pigs died during the nursery, growing and finishing stages, respectively. CF pigs were at higher risk of death regardless of treatment ($P < 0.05$; Table 1) with similar odds for the risk of death in CFW1 and CFW2+ pigs compared with NCF pigs. Using the nested case-control design, growth performance did not differ between treatments ($P > 0.05$); however, CFW2+ carcasses were 4.9 kg lighter and had 3 mm less muscle depth ($P < 0.05$) compared with NCF and CFW1 pigs. There were no differences between treatments in the presence of tail and body lesions ($P > 0.05$) but ear lesions were more likely in CFW1 compared to NCF and more likely in CFW1 compared to CFW2+ ($P < 0.05$; Table 1) pigs.

**Table 1** Odds ratios (OR) ± 95% confidence intervals (CI) for the risk of mortality and ear lesions in 1,016 pigs followed from birth to slaughter in one commercial farm according to the lactation week when they were cross-fostered (CF)

| | Mortality from birth to weaning | | | Mortality from birth to slaughter | | | Ear lesions | | |
|---|---|---|---|---|---|---|---|---|---|
| | | 95% CI | | | 95% CI | | | 95% CI | |
| | OR | Lower | Upper | OR | Lower | Upper | OR | Lower | Upper |
| NCF vs. CFW1 | 2.06[a] | 1.12 | 3.62 | 2.06[a] | 1.23 | 3.35 | 2.19[a] | 1.29 | 3.78 |
| NCF vs. CFW2+ | 2.99[a] | 1.85 | 4.76 | 2.43[a] | 1.57 | 3.70 | 0.90 | 0.52 | 1.56 |
| CFW1 vs. CFW2+ | 1.45 | 0.77 | 2.82 | 1.18 | 0.67 | 2.10 | 0.41[a] | 0.22 | 0.76 |

[a] Different from reference category; $P < 0.05$

**Conclusion** Rather than reducing pre-weaning mortality, CF increased the risk of death at all stages of the production cycle irrespective of when it was performed. Moreover, early CF had a negative impact on ear lesions which are associated with poor welfare and late CF was associated with lower carcass weight. Neither treatment improved pig performance relative to non-CF pigs although the underlying reasons are not yet understood.

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**References**

Heim, G., Mellagi, A.P.G., Bierhals, T., De Souza, L.P., De Fries, H.C.C., Piuco, P., Seidel, E., Bernardi, M.L., Wentz, I. and Bortolozzo, F.P. 2012. Livestock Science 150, 121–127.
Van Erp-Van Der Kooij, E., Kuijpers, A.H., Van Eerdenburg, F.J.C.M. and Tielen, M.J. M 2003. Livestock Production Science 84, 31–38.

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026

# Effect of sugar beet pulp and L-carnitine in gilt gestation diets on gilt weight and lactation feed intake and progeny growth

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**Application** Pigs born to L-carnitine supplemented gilts had increased muscle depth and a heavier cold carcass weight at slaughter (142 days old). Inclusion of 40% sugar beet pulp does not increase gilt lactation feed intake.

**Introduction** Advances in genetic selection for increased sow prolificacy has resulted in a reduction in piglet birthweight and increased piglet mortality. Therefore, strategies to increase piglet weight and robustness at birth are of increasing importance. Gilts tend to give birth to lighter piglets and their progeny have lower pre-weaning growth rates than piglets from multiparous sows (Carney-Hinkle *et al.* 2013). Previous research found that supplementing gestating sows with L-carnitine (L-car) increased piglet birthweight and lifetime growth (Birkenfeld *et al.* 2006 and Ramanau *et al.* 2008); however there is little information regarding L-car supplementation to gilts. Furthermore, due to a lower gut capacity in gilts, lactation feed intakes are low relative to that of multiparous sows. Increasing fibre level with sugar beet pulp (SBP) during gestation may increase gut capacity at parturition, thereby facilitating higher voluntary feed intake during lactation. It was hypothesised that L-car combined with a high SBP diet could benefit piglets in gilt litters by increasing birthweight and consequently improve lifetime piglet growth. Furthermore, feeding high levels of SBP in the diet will increase lactation feed intake. To test this hypothesis, we conducted a 2 x 2 factorial experiment with two factors for fibre (0% SBP and 40% SBP) and two factors for L-car (0g/day and 0.125g/day).

**Material and methods** Eighty four pregnant gilts (4 batches, ~21 gilts each) were used. At d38 of gestation, gilts were blocked within batch by live-weight and back-fat depth (P2), and randomly assigned to one of four dietary treatments until parturition; Control (~90%Barley; ~ 9%Soya; 0g/d L-car), High SBP (~48%Barley; ~ 9%Soya; 40% SBP), Control + L-car (0.125g L-car/d), and High SBP + L-car (0.125g/d L-car). Gilt live-weight and P2 were recorded on d90 and d108 of gestation and at weaning. Daily feed intake of gilts was recorded throughout gestation and lactation. Gilts were saliva sampled every 3 weeks between d90 and d108 of gestation and samples analysed for cortisol concentration. At farrowing, total number of piglets born alive and born dead was recorded. A blood sample was taken from the left ear vein of piglets at 24h post-partum and glucose concentration was determined. Piglets were individually weighed at birth, 24h, d6, d13 and d26 of lactation. Post-weaning, all pigs were grouped by gilt treatment (12-14 pigs/pen), offered the same sequence of diets and managed identically to slaughter (d142). Pigs were weighed on d75, d108 and d142 and daily feed intake of pigs was recorded. Data were analysed using the mixed models procedure in SAS (v.9.4) using a 2 x 2 factorial arrangement. The model included fibre, L-car and interaction terms as main effects, replicate as a fixed effect and block as the random effect.

**Results** There was no fibre by L-car interaction for any variable of interest, therefore only the main effects are presented here. Gilt P2 was unaffected by treatment ($P>0.05$). Gilt weight was unaffected by L-car, but gilts fed high SBP were heavier on days 90 and108 of gestation ($P<0.05$). Lactation feed intakes were similar for all treatments. Cortisol levels were numerically higher for gilts fed high SBP compared to those fed low SBP (0.53 u/dL vs. 0.47 u/dL± 0.04; $P=0.18$). Numbers born were similar for all treatments. Piglet birthweight was unaffected by treatment. Males were born heavier than females (1.2kg vs.1.1kg ± 0.02; $P<0.05$). Glucose concentration was unaffected by treatment. All piglet pre-weaning weights were unaffected by treatment, however, piglets from gilts fed L-car were lighter at weaning compared with piglets from gilts that were not fed L-car (6.8kg vs. 7.3kg ± 0.1; $P<0.05$). Similarly, piglets from L-car supplemented gilts had lower ADG from d6 to weaning than piglets of non L-car supplemented gilts (229g/day vs. 255g/day± 0.007; $P<0.05$). Post-weaning, pigs from L-car fed gilts had higher ADG than pigs of non L-car fed gilts (618.1g/day vs. 594.8g/day± 6.62; $P<0.01$) up to d75. At slaughter, pigs from high SBP fed gilts and L-car supplemented gilts had a heavier carcass cold weight (86.6kg vs. 85.3kg± 0.4; $P<0.05$) and higher muscle depth (51.2mm vs. 50.5mm± 0.2; $P<0.05$) on average, than pigs from control gilts.

**Conclusion** Although our results do not support our original hypothesis, the inclusion of L-car in gilt gestation diets resulted in an increase in the carcass weight of progeny at slaughter. Furthermore, carcass muscle depth was increased in progeny due to L-Car supplementation. Increasing fibre level by including 40% SBP in a gilt gestation diet did not result in increased lactation feed intake and/or increased piglet growth to weaning.

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**References**

Birkenfeld, C., Kluge, H. and Eder, K. 2006. British Journal of Nutrition 96, 334-342
Carney-Hinkle, E., Tran, H., Bundy, J., Moreno, R., Miller, P. and Burkey, T. 2013. Journal of Animal Science 91, 2885-2893
Ramanau, A., Kluge, H., Spilke, J. and Eder, K. 2008. Livestock Science 113, 34-42

# Natural strategy for the replacement of zinc oxide in newly weaned piglets

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**Application** The European commission has banned inclusion of pharmacological levels of zinc in piglet diets by 2022. This study demonstrated that a eubiotic acid and essential oil blend can replace 3g/kg zinc oxide in commercial pig diets.

**Introduction** Zinc in the form of zinc oxide is routinely prescribed at 3g/kg in the weaning diet for two weeks post-weaning to help control scours and enteric disorders at this time. Recently this use has been linked to environmental pollution and antibiotic resistance in some bacteria, (FEEDAP, 2017; Yazdankhah *et al*. 2014). The reduced maximum content (0.15g/kg) of zinc oxide allowed in pig diets will come into legislation before 2022. This is expected to result in high economic losses of up to €1,080 million/year for the EU pig industry. The aim of the study was to evaluate a commercially available eubiotic acid and essential oil blend in replacement for therapeutic zinc oxide in an *E. coli* challenge study.

**Material and methods** Thirty six piglets (genetics; PIC (Yorkshire × Landrace) × Duroc)) were allocated to treatment diets at weaning (21 days) for a duration of 28 days and balanced for litter origin, gender and start weight in a random block design (2 pigs per pen, 6 replicates). All diets met or exceeded the nutrient requirements as recommended by NRC (2012) for weaning pigs and were fed in a mash form. Treatments were; Control (CON), basal diet no additive; Genex Weaner a eubiotic blend (EUB) (Anpario plc, Worksop, UK) included at 4g/kg; Zinc oxide (ZnO) commercially available included at 3g/kg. Eight days post weaning (29 days of age), piglets were orally challenged with 6 mL ($6.1 \times 10^9$ cfu/mL) ETEC (*E. coli*, strain K88+) according to the method outlined in Owusu-Asiedu *et al*. (2003). Animals had access to feed and water *ad libitum*. Body weight and feed intake were measured weekly and all data were evaluated using ANOVA and Tukey's t-test for pairwise differences using JMP Pro software (JMP.inc).

**Results** The EUB group had the highest daily live weight gain over the 28 day period as shown in Figure 1, with an increase ($p<0.05$) of 73g/day compared to the CON group. A significant ($p<0.05$) improvement of 2.11kg in final body weight after 28 days (figure 2) was achieved in the EUB group compared to the CON group. No difference was seen between EUB and ZnO groups in terms of final body weight and daily live weight gain. Feed conversion efficiency did not differ significantly between treatments. The E.coli challenge demonstrated the expected effect and was effective in reducing performance of piglets compared to unchallenged individuals on the same unit.

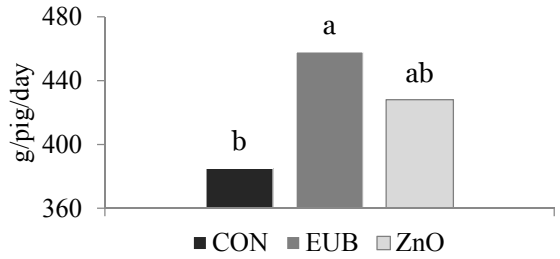


**Figure 1** Overall daily live weight gain from day 0-28

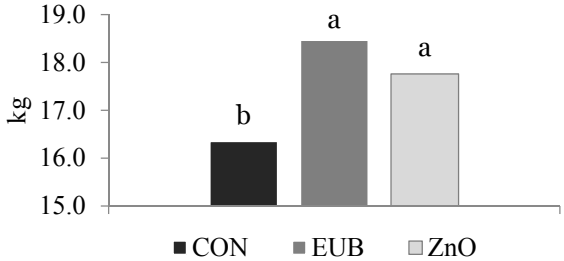


**Figure 2** Final body weight day 28

**Conclusion** The eubiotic, included in feed at 4g/kg, has been shown to be an effective replacement for therapeutic levels of zinc oxide in piglet weaning diets without compromising growth performance or efficiency. Using costs prevailing at the time of the study the eubiotic treatment provided an economical benefit over the control and zinc oxide treatment.

**References**

EFSA Panel on Additives and Products or Substances used in Animal Feed, 2017. EFSA Journal 12(5), 3668.
Owusu-Asiedu, A., Nyachoti, C.M. and R.R. Marquardt. 2003. Journal of Animal Science 81(7), 1790-1798.
Yazdankhah, S., Rudi, K. and Bernhoft, A., 2014. Microbial Ecology in Health and Disease 25(1), 25862.

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**028**

# Replacement of soybean meal with *Nannochloropsis* spp: its effect on *g*rowth performance of weaner rabbit

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**Application** Increasing the quantity of *Nannochloropsis* spp as a replacement to soybean in the diet of weaner rabbit reduced the feed efficiency of both sexes.

**Introduction** Human consumption of rabbit meat has been utilised as a solution to inadequate protein intake in some regions. Rabbit (*Oryctolagus cuniculus*) has great advantages over other livestock because of its prolificacy and short gestation period, ability to attain market size within six months and the ability to utilize forages and fibrous agricultural by-products (Kagya-Agyemang *et al.,* 2013). However due to high cost of feed, there is the need to explore the use of non-conventional feed sources that have the capacity to yield the same output as conventional feedstuffs and perhaps at a lower cost. An example of the non-conventional feedstuff is algal biomass (*Nannochloropsis* spp.).

**Material and methods** The experiment was carried out at Federal University of Agriculture, Abeokuta, Nigeria. Fifty-four weaner rabbits (twenty-seven for each sex) of mixed breeds were purchased from reputable farms and were acclimatized for two weeks before they were distributed to three treatment groups per sex resulting in six groups of nine rabbits each. *Nannochloropsis* spp (algal biomass) was used to replace soybean at 0, 2, or4% to produce three different diets which each sex of the animals was subjected to. Feed was given *ad libitum* and adequate fresh water was supplied daily. The feeding trial lasted for nine weeks. Data on growth performance which included feed intake, weight and feed efficiency were taken on weekly basis and data gathered were subjected to a two-way ANOVA (with initial weight as a covariate) using the Minitab 16 statistical package. Means were also separated using the Tukey test.

**Results** Sex influenced only feed intake and feed efficiency while feeding *Nannochloropsis* influenced all of the other variables measured except daily feed intake (Table 1).The interaction effect of replacing soybean with *Nannochloropsis* spp is presented in the Table2. The results showed that weight gain and resultant effect on final weights were not ($P>0.05$) affected by the replacement level. However, the results on the gender and diet interaction showed that daily feed intake of the males was similar across the groups while the feed intake values recorded for the females increased as the level of replacement increased. The feed efficiency reduced as the inclusion level of *Nannochloropsis* spp increased.

**Table 1** Main effect of sex and *Nannochloropsis* on growth performance of weaner rabbits

| | Sex | | | *Nannochloropsis (%)* | | | |
|---|---|---|---|---|---|---|---|
| Parameter | Male | Female | SEM | 0 | 2 | 4 | SEM |
| Final weight (g) | 1527 | 1445 | 25.39 | 1552$^{a}$ | 1495$^{ab}$ | 1411$^{b}$ | 31.05 |
| Weight gain (g) | 774.9 | 693.2 | 25.39 | 799.9$^{a}$ | 743.2$^{ab}$ | 658.9$^{b}$ | 31.05 |
| Daily Weight gain (g) | 12.30 | 11.02 | 1.06 | 12.69$^{a}$ | 11.80$^{ab}$ | 10.46$^{b}$ | 1.80 |
| Daily Feed intake (g) | 67.92$^{a}$ | 82.59$^{b}$ | 1.75 | 73.95 | 74.16 | 77.65 | 2.14 |
| Feed Efficiency | 0.18$^{a}$ | 0.13$^{b}$ | 0.04 | 0.17$^{a}$ | 0.16$^{ab}$ | 0.13$^{b}$ | 0.07 |

**Table 2** Interaction effect of sex and *Nannochloropsis inclusion* on growth performance of weaner rabbits

| | Male | | | Female | | | |
|---|---|---|---|---|---|---|---|
| Parameter | 0 | 2 | 4 | 0 | 2 | 4 | SEM |
| Final weight (g) | 1616 | 1529 | 1436 | 1487 | 1462 | 1386 | 48.12 |
| Weight gain (g) | 864.2 | 776.7 | 683.7 | 735.6 | 709.8 | 634.2 | 48.12 |
| Daily Weight gain (g) | 13.72 | 12.33 | 10.85 | 11.67 | 11.27 | 10.07 | 2.26 |
| Daily Feed intake (g) | 67.73$^{bc}$ | 66.85$^{c}$ | 69.16$^{bc}$ | 80.18$^{abc}$ | 81.46$^{ab}$ | 86.13$^{a}$ | 3.32 |
| Feed Efficiency | 0.20$^{a}$ | 0.18$^{ab}$ | 0.16$^{bc}$ | 0.14$^{bc}$ | 0.14$^{c}$ | 0.12$^{c}$ | 0.07 |

**Conclusion** The present results do not conclusively recommend *Nannochloropsis* biomass as a viable replacement for soybean in rabbit diets. Further studies are definitely needed to validate these preliminary outcomes.

**References**

Kagya-Agyemang, J. K., Nelson, A. and Kagya-Agyemang, C. 2013. Growth performance of weaner rabbits fed on dried pito mash as a replacement for maize. ARPN Journal of Agricultural and Biological Science 8(4), 267-272

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029

# Galacto-oligosaccharides: an investigation into dietary inclusion levels for rainbow trout (*Oncorhynchus mykiss*) for improved carcass characteristics

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**Application** Galacto-oligosaccharides (GOS) appear to modify rainbow trout body composition by increasing lipid content. These findings require further investigation before applications can be considered for improving production characteristics.

**Introduction** Prebiotic and probiotic supplements play an evolving role in animal feeds as growth and health promoters, particularly as restrictions tighten on routine antibiotic use, as the issue of antimicrobial resistance persists (PEW, 2017). Prebiotics are substrates that are selectively utilised by host microorganisms conferring a health benefit (Gibson *et al*., 2017). Galacto-oligosaccharides, manufactured by the enzyme catalysed transgalactosylation of lactose, are oligosacharides with prebiotic properties that consist of galactose units linked by β(1→n) bonds, where usually n = 3, 4 or 6, ending in a β(1→4) linked terminal glucose. There are currently only a limited number of studies published regarding the use of GOS for dietary inclusion in salmonid fish species (*O.mykiss* and *S.salar*). The aim of the present study was to evaluate the effects of gradient levels of dietary GOS inclusion on body composition of rainbow trout (*O.mykiss*), in balanced feed formulations.

**Material and methods** One thousand and two hundred juvenile rainbow trout (19.53±0.75g) were randomly allocated to one of four iso-nitrogenous and iso-calorific dietary treatments: 0.00 % GOS (Control), 0.88 % GOS, 1.75 % GOS and 3.50 % GOS in a 12-week experimental study. Fish were arranged between 12 tanks (three replicate tanks per treatment group), each containing 100 fish at the start of the trial period. All tanks were weighed on a communal biomass basis each week and numbers were reduced via a uniform cropping regime across all tanks, to relieve the rising stocking density as fish weight increased by week. Body composition and condition factor were measured via sampling of fish at week 12. Six fish per treatment were measured via fork length and weighed for condition factor (Phelps *et al*., 2013). Whole body moisture and dry matter values were achieved via drying for 24 hours at 105°C. Whole body crude protein, crude energy and lipid content values were measured via LECO FP-528 Nitrogen analyser, bomb calorimetry (Parr Instrument Company, USA) and Soxtec (FOSS Ltd) respectively in technical triplicates. Data was analysed as a dose response design by ANOVA with orthogonal polynomial contrasts for linear, quadratic and deviations using GenStat (18th edition).

**Results** At sampling fish were in the body weight range of 50 – 116 g. Condition factor was not significantly influenced by dietary GOS inclusion. Changes in body composition are detailed in Table 1. Increasing dietary GOS supplementation resulted in a linear increase in whole-body lipid ($p<0.001$) and gross energy content ($p<0.001$). There was a linear decrease ($p=0.002$) in moisture content, while whole-body ash content decreased in a linear ($p=0.002$) and quadratic ($p<0.001$) manner.

**Table 1** Effects of increasing dietary GOS on the body composition (as is) and condition factor of rainbow trout (*O. mykiss*)

| Diet | Control | GOS 0.88% | GOS 1.75% | GOS 3.50% | SEM | p-value | CL | CQ | CD |
|---|---|---|---|---|---|---|---|---|---|
| Moisture (%) | 67.11 | 63.17 | 64.18 | 61.10 | 0.0112 | 0.006 | 0.002 | 0.512 | 0.1 |
| Crude Protein (%) | 15.20 | 15.61 | 15.86 | 15.75 | 0.005 | 0.746 | 0.42 | 0.457 | 0.977 |
| Lipid (%) | 11.79 | 12.55 | 13.01 | 13.69 | 0.003 | <.001 | <.001 | 0.33 | 0.84 |
| Ash (%) | 1.98 | 1.89 | 1.84 | 1.88 | 0.0002 | <.001 | 0.002 | <.001 | 0.973 |
| Total | 96.1 | 93.2 | 94.9 | 92.4 | | | | | |
| Gross Energy (MJ/kg) | 8.20 | 8.51 | 8.70 | 9.06 | 0.0989 | <.001 | <.001 | 0.445 | 0.742 |
| Condition factor | 1.52 | 1.48 | 1.45 | 1.47 | 0.0265 | 0.231 | 0.177 | 0.118 | 0.871 |

CL = p-value linear contrast; CQ = p-value quadratic contrast; CD = p-value deviations contrast

**Conclusion** Significant alterations in whole body composition were observed with increasing dietary inclusion of GOS. This was principally increased body lipid percentage, resulting in greater gross energy content. Further investigation is needed to assess the effects of GOS on growth performance and gut health to help explain these results.

**Acknowledgements** The authors gratefully acknowledge funding from Dairy Crest Ltd and the staff at Exmoor Fisheries for helping to run this study.

**References**

Gibson, G.R., Hutkins, R., Sanders, M.E., Prescott, S.L., Reimer, R.A., Salminen, S.J., Scott, K., Stanton, C., Swanson, K.S., Cani, P.D. and Verbeke, K., 2017. Nature Reviews Gastroenterology & Hepatology. doi:10.1038/nrgastro.2017.75.
PEW. 2017. Alternatives to antibiotics in animal agriculture. The Pew Charitable Trusts.
Phelps, M.P., Jaffe, I.M. and Bradley, T.M. 2013. Journal of Experimental Biology 216, 3742-3750.

**030**

# Nephrotoxic effects of copper oxide nanoparticles in *Cyprinus carpio* assessed by histological profiles

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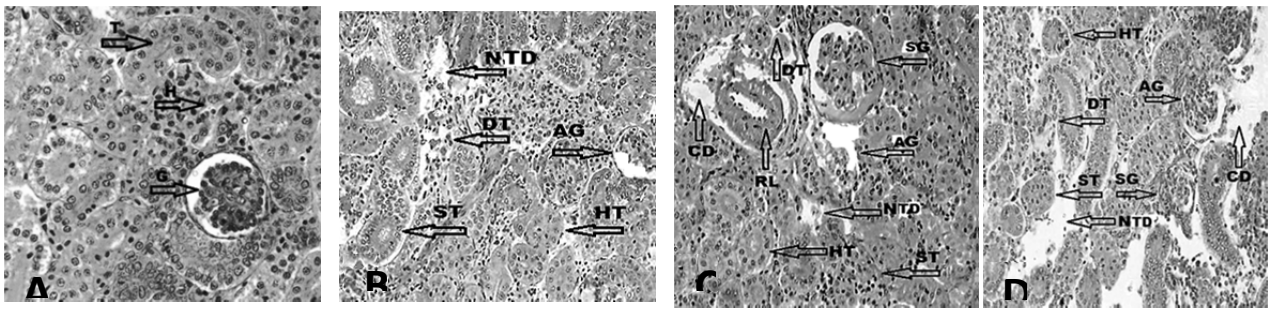
**Application** The results of this study provide an insight into the toxicity of Cu-NPs in aquatic organisms. It may help policy makers to suggest preventive measures against the potential toxicity of nanoparticles (NPs) in aquatic organisms.

**Introduction** Metal oxide NPs such as copper (Cu) have gained significant attention due to their negative ecological effects (Nations *et al*., 2011). A wide range of NPs is being used as biosensor immobilizers for their greater sensitivity and specificity. Due to their low preparation cost and prospective applications, Cu-NPs are intensively used in the industry. Besides, Cu-NPs are also used as one of the main constituents of fungicides, algaecide and herbicides. However, they can cause histological alterations and oxidative DNA damage in the living organisms at the cellular level. The use of engineered NPs can result in their release into aquatic environments causing unexpected hazards for aquatic organisms.Therefore, this study assessed the toxicological impacts of Cu-NPs on the kidneys of *Cprinus* (*C*) *carpio.*

**Material and methods** A fish husbandry system was established prior to the start of experiments to support the health of fish by maintaining the water quality and environment of stock aquariums. *Cyprinus carpio (C. carpio)* of similar weight (40-45g) were procured from a Fish Seed Hatchery, transported in plastic containers with continuous aeration to the laboratory of GC University Faisalabad, Pakistan after approval of the local Ethics Committee of the GC University Faisalabad, on Animal Experimentation. The fish were acclimatized in the tank with 100 L capacity for two weeks prior to the experiment. Un-chlorinated tap water was used and the physicochemical parameters of water were determined by a multi parameter apparatus (HI 9828, HANNA INSTRUMENTS). During the acclimatization period, water temperature was maintained at 25ºC, while dissolved oxygen and pH were 6.6–7.6 mg/l, and 6.9–7.5, respectively. During the acclimatization period, the fish were fed with a commercial fish feed. After acclimatization, *C. carpio* from aquaria were randomly transferred into twelve aerated experimental glass aquaria (10 fish/ aquarium) with the same physicochemical parameters as in the acclimatization period and acclimated for 48 hours prior to the experiment. The fish were exposed in triplicate to 0 (Control) or 0.5 or 1 or 1.5 mg/l Cu as Cu-NPs for 14 days. During this study, water in the aquaria was changed daily and freshly prepared solution was added to maintain the concentration of Cu-NPs at a constant level. At the end of the experiment fish from each aquarium were immediately anesthetized into a 200 ppm solution of clove powder and kidneys were collected for histological analysis. Kidney tissues were fixed in freshly prepared Bouin's fixative (15:5:1, saturated picric acid, formalin, and glacial acetic acid) for 12-14 h at room temperature. The fixed tissues were processed by a standard procedure using hematoxylin-eosin. The slides were observed under a light microscope (Nikon Eclipse E200 POL) fitted with a camera and photographed.

**Results** The results revealed that the Cu-NPs induced alterations in the kidneys of *C.carpio* in a dose dependent manner. The histological (h) alterations were quantified by using a five graded examination scheme: no h alterations (normal histological structure); mild h alterations; moderate h alterations; severe h alterations and very severe h alterations.The alterations in kidneys, including necrosis and tubular degeneration (NTD), hypertrophy of tubules (HT), reduced lumen (RL), abnormal glomerulus (AG), shrinked glomerulus (SG), swollen tubules (ST), complete degeneration (CD) and degenerative tubules (DT) were increased with the increase in the dose of Cu-NP exposure.




A= control (normal histology) B - 0.5 mg/l Cu NPs exposure C - 1 mg/l Cu NPs D - 1.5 mg/l Cu NPs

**Figure 1** Microphotograph of *C. carpio* kidney in control and treated groups showing histological alteration (H&E, X400)

**Conclusion** It is concluded that Cu-NPs were toxic to the fish as indicated by nephrotoxicity in *C. carpio*. Thus, it is suggested that the bioavailability of metal NPs by aquatic pollution and subsequent accumulation in fish tissues might constitute a substantial risk to human health and to the environment.

**References**

Nations, S., Wages, M., Cañas, J. E., Maul, J., Theodorakis, C., Cobb, G.P. 2011. Acute effects of $Fe_2O_3$, $TiO_2$, ZnO and CuO nanomaterials on Xenopus laevis. Chemosphere 83, 1053–1061.

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# The variability in apparent (AID) and standardised ileal digestibility (SID) of rapeseed meal for broilers

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**Application** There is wide variability in ileal digestibility of dry matter (DM) and amino acids. Therefore, using average values for digestibility co-efficients may result in inaccurate diet formulation if this variability is unaccounted for.

**Introduction** There is a lack of information available on the variability and extent of digestible amino acid content of rapeseed meal (RSM) and diet formulations are based on average "book" values for digestible amino acid content. There is often wide variation in these book values and they have been derived under different methodologies including prediction from total amino acid content. Ravindran *et al*. (2005) highlighted the need for a database of apparent ileal amino acid digestibility coefficients to ensure accurate formulation and subsequent optimal performance. This study was part of a large-scale project on RSM use in pig and broiler diets and the aim of this part was to investigate the variability in apparent and standardised ileal digestibility (AID and SID) of RSM for broilers. An additional aim was to ascertain the usefulness of using broiler digestibility co-efficients as a model to predict AID and SID in pigs.

**Material and methods** This work was approved by the AFBI Animal Welfare Ethical Review Body. A total of 92 RSM samples were collected from crushing plants in Hull and Liverpool (Cargill Plc., Weybridge, UK). Each RSM was formulated into separate test diets and was included at 500g/kg, mixed with; (g/kg) maize starch (300), dextrose (100), soya oil (60), titanium dioxide (3) and other ingredients (37). An N-free maize starch (450g/kg)/dextrose (420g/kg) diet was formulated to determine basal endogenous losses. The diets were offered to 15 batches of 64 male broilers (Ross 308) in individual metabolism cages over 15 experimental trials. Each batch contained two controls (the N-free diet and RSM3) in order to compare results between batches. Therefore, there were eight diets offered to eight birds in each batch. This resulted in eight replicates per RSM sample (and 120 replicates for N-free and RSM3 diets). All birds were offered standard starter diet *ad libitum* from 7-18d. At 18d, birds which were allocated to RSM diets were offered standard starter diet and the appropriate RSM treatment diet on a 50:50 mix until 21d. From 21-28d, birds were offered the appropriate RSM treatment diet. Birds allocated to the N-free diet were offered the diet from 25-28d. At 28d, the birds were humanely killed and the contents of the ileum collected to determine AID of DM and amino acids. Basal endogenous losses were determined and used to calculate SID. The minimum, mean and maximum digestibility values were determined and standard deviations for each parameter calculated. Linear regression analysis was conducted to correlate broiler digestibility co-efficients with pig digestibility co-efficients (Magowan *et al*. 2017).

**Results** There were wide ranges in AID and SID values within the sample set for DM and essential amino acids (Table 1). For example, AID and SID for lysine ranged from 0.486 to 0.905 and 0.733 to 1.104 respectively. The standard deviation values for all parameters reflect the wide variability within the sample set. The majority of the relationships between broiler and pig AID and SID values were non-significant. There were significant but poor ($P<0.05$) relationships observed between broiler and pig AID of DM ($R^2=0.06$) and methionine ($R^2=0.40$).

**Table 1** The range and standard deviation in AID and SID of DM and essential amino acids of RSM (n=92) in broilers

| | Apparent ileal digestibility | | | | Standardised ileal digestibility | | | |
|---|---|---|---|---|---|---|---|---|
| | Minimum | Mean | Maximum | Standard deviation | Minimum | Mean | Maximum | Standard deviation |
| Dry matter | 0.387 | 0.654 | 0.873 | 0.076 | * | * | * | * |
| Arginine | 0.395 | 0.794 | 0.946 | 0.076 | 0.604 | 0.942 | 1.098 | 0.070 |
| Histidine | 0.524 | 0.739 | 0.749 | 0.041 | 0.539 | 0.755 | 0.766 | 0.042 |
| Isoleucine | 0.474 | 0.699 | 0.847 | 0.068 | 0.700 | 0.919 | 1.049 | 0.067 |
| Leucine | 0.436 | 0.734 | 0.893 | 0.085 | 0.629 | 0.922 | 1.085 | 0.084 |
| Lysine | 0.486 | 0.677 | 0.905 | 0.072 | 0.733 | 0.930 | 1.104 | 0.062 |
| Methionine | 0.458 | 0.769 | 0.956 | 0.112 | 0.547 | 0.948 | 1.121 | 0.127 |
| Phenylalanine | 0.422 | 0.734 | 0.838 | 0.083 | 0.631 | 0.921 | 1.051 | 0.085 |
| Threonine | 0.415 | 0.624 | 0.792 | 0.075 | 0.685 | 0.922 | 1.086 | 0.074 |
| Valine | 0.447 | 0.678 | 0.823 | 0.070 | 0.684 | 0.923 | 1.063 | 0.072 |

**Conclusion** AID and SID of RSM in broilers is highly variable within a large sample set. This indicates that using average book values for AID and SID to include RSM into broiler diets will result inaccurate formulation. This highlights the need to develop a means to predict AID and SID of individual RSM samples. Additionally, broiler AID of methionine may have potential to be used as a model to predict AID of methionine in pigs but the relationships are quite weak.

**Acknowledgements** The authors acknowledge funding from DAERA and ABVista and also Cargill and Technlogy Crops for sample collection.

**References**

Ravindran, V., Hew, L. I., Ravindran, G. and Bryden, W. L. 2005. Animal Science 81, 85-97.
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# The use of near infra-red reflectance spectroscopy to predict ileal digestibility of essential amino acids in rapeseed meal for pigs and broilers

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**Application** Near infra-red reflectance spectroscopy has potential to predict ileal digestibility of essential amino acids in rapeseed meal for pigs and broilers, resulting in more accurate diet formulation and greater inclusion of rapeseed meal in non-ruminant diets.

**Introduction** The inclusion of rapeseed meal (RSM) in diets for broilers and pigs has received much attention as RSM is viewed as an attractive home-grown partial alternative to soyabean meal for both species. However, there is wide variability in terms of apparent and standardised ileal digestibility (AID and SID) of amino acids (Magowan *et al* 2017; Ball *et al* 2018). To ensure accurate formulation and optimal use of RSM in non-ruminant diets, a means to predict RSM AID and SID of amino acids is necessary. Near infra-red reflectance spectroscopy (NIRS) can predict the nutritive value of wheat for broilers (Ball *et al* 2017) and the aim of this study was to develop NIRS equations to predict AID and SID of amino acids in RSM for pigs and broilers.

**Material and methods** This work was approved by the AFBI Animal Welfare Ethical Review Body. A total of 92 RSM samples were collected, formulated into pig and broiler diets and offered to post-valve T-cannulated pigs and broilers (according to Magowan *et al* 2017 and Ball *et al* 2018). The RSM samples were scanned in duplicate using a NIRSystems 6500 spectrophotometer (Foss, Hillerød, Denmark) over the wavelength range 400-2498nm with readings taken at 2nm gaps. The scans were analysed using the Foss Chemometrics software Win ISI4. The mathematical treatment of standard normal variate and detrend, first derivative, gap of 4 and smooth of 4 was applied. Modified partial least squares regression was performed on the data set on the range 400 nm – 2500nm and NIRS calibration and cross-validation statistics generated to predict digestibility co-efficients and compared with determined values obtained through pig and broiler trials. Definitions: SECV=standard error of cross validation, SECV as % of mean=indication of accuracy of calibration, with values of less than 5% being acceptable and RPD calibration=ratio of prediction to deviation (SD/SECV) with values of >1.5 indicating that calibration has potential to accurately predict.

**Results** The relationship between determined and predicted values for AID of DM and essential amino acids in pigs were reasonably robust with $R^2$>0.6. SEC and SECV were low and SECV as % of mean was below 5% apart from isoleucine, leucine, lysine, threonine and valine. However, RPD values for isoleucine, lysine, methionine and valine AID were greater than 1.5, indicating that while these calibrations are poor, they can distinguish between high and low values in the dataset. In general, the relationship between determined and predicted SID assessed in terms of $R^2$ was quite robust, with all essential amino acid SID $R^2$ values being greater than 0.6. Also, SEC and SECV as % of mean were low for SID of arginine, methionine, phenylalanine and theronine. The calibration predictions for pig SID of isoleucine, lysine and threonine (RPD calibration >1.5) were able to distinguish between high and low values in the dataset. The majority of relationships between determined and predicted broiler AID (as assessed by $R^2$) were reasonably strong ($R^2$>0.6), apart from the relationship between determined and predicted histidine AID ($R^2$=0.26). Contradictorily, SECV as % of the mean was only below 5% for histidine AID (3.1%). The highest RPD calibration values were observed for the prediction of AID of phenylalanine and arginine (1.86 and 1.54 respectively). The relationships between determined and predicted SID of amino acids in broilers were reasonably robust in terms of $R^2$ values. The lowest $R^2$ were observed for histidine and methionine SID ($R^2$=0.30 and 0.35, respectively). Very few SID predictions resulted in SECV as % of mean of less than 5% (arginine, 5.0% and histidine, 3.2%) and only two RPD calibration values were >1.5 (SID of leucine and phenylanine).

**Conclusion** The relationships between determined and predicted digestibility co-efficients were reasonably strong and in many cases the errors associated with prediction for cross-validation (SECV) were low and as a % of the mean, less than 5%. However, a low error value does not necessarily indicate a strong prediction equation, and RPD is regarded as the criterion for judging the strength of prediction. RPD values for several digestibility parameters were above 1.5 which indicates that the prediction equation can distinguish between high and low values in a dataset and may be of some value. Therefore, the digestibility co-efficients with RPD values of above 1.5 should be considered for further development. For pigs, this would include SID of isoleucine, lysine and threonine. For broilers, this would include AID of arginine and phenylalanine and SID of leucine and phenylalanine. These results suggest that NIRS has the potential to predict amino acid digestibility and they are a useful basis to further develop prediction equations.

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**References**

Ball, M. E. E., Magowan, E., Garcia, R. and Graham, H. 2018. British Poultry Abstracts (in press)
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033

# Identification of differentially expressed genes in the *M. longissmus thoracis et lumborum* from full-sibling lambs divergent for fatty acid profile

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**Application** Functional SNPs in the promoter regions of differentially expressed genes involved in the control of the fatty acid profile in lamb muscle will be useful in selective breeding programmes to enhance the benefits of lamb meat for human health.

**Introduction** The fatty acid (FA) profile of lamb muscle influences taste, flavour and quality of cooked and cured meat products (Wood *et al*., 2008). Polyunsaturated fatty acids (PUFAs) are associated with reduced (LDL) cholesterol and blood triacylglycerols, hypertension and diabetes in addition to having positive effects on inflammation and auto-immune function (Galli *et al*., 1994; Zhang *et al,* 2009). Thus, increasing the ratio of PUFA to saturated fatty acids (SFA) in the diet is generally considered beneficial for human health and wellbeing. The objective of this study was to identify differentially expressed (DE) genes in *M. longissimus thoracis et lumborum* (LTL), using a divergent full-sibling model for ovine fatty acid profile. This will help highlight specific genes with the view to performing SNP discovery in the promoter region.

**Material and methods** Twin lambs (64 pairs) were used, were sired by rams from 1 of 3 terminal-sire breeds (Charollais (n = 20), Suffolk (n = 21) and Texel (n = 23)), and were slaughtered at normal commercial finish. Samples (2 g) of LTL muscle were collected within 15 min post-mortem and stored in RNAlater (Ambion Inc., Austin, TX) for subsequent RNA extraction. Quantity and quality of total RNA were assessed using the NanoDrop ND-1000 spectrophotometer (Thermo Fisher Scientific, MA, USA). All RNA samples with $A_{260/280}$ ratios ≥2.0 were used for further analysis. Fatty acids in LTL muscle (1 g) were quantified by gas chromatography (Clarus 580, PerkinElmer, MA, USA) and using a capillary column (Zebron ZB-5MS). The total SFA and PUFA were corrected for both intramuscular fat and slaughter date. Principal component analysis (Proc PRINCOMP; SAS 2012) of within-litter differences for SFA and PUFA was used to identify the most divergent sib pairs, i.e., having high PUFA and low SFA or the converse. Lambs with high PUFA were classified as HIGH while those with low PUFA were classified as LOW. The 8 most divergent pairs were used to evaluate the association between PUFA:SFA status and the level of gene expression. The genes evaluated were chosen from a panel of 63 whose expression levels had been quantified using gene-specific primers and qPCR; the software package qbase$^+$ (Biogazelle, Belgium) was used to calculate normalized relative quantities (NRQ) of each gene. The genes chosen (n= 10) had exhibited significantly increased within-litter variation in expression level, based on the CV, with an experiment-wise error rate of 0.05. The NRQ for the HIGH and LOW groups were compared (Proc GLM; SAS 2012).

**Results** The mean NRQ expression levels differed significantly between HIGH and LOW animals for 5 genes at $P < 0.01$ (LPL, PGC1A, FABP3, FAM134B and TG) and at $P < 0.05$ for LDLR (Figure 1). The NRQ level was greater in HIGH in all cases except LDLR and TG. Four of these 6 genes (LPL, LDLR, FABP3 and FAM134B) are known to be involved in fatty acid metabolism.

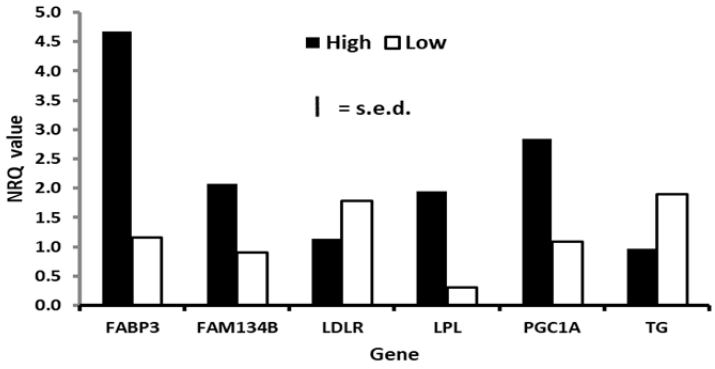


**Figure 1** Mean NRQ values for genes with DE for fatty acid profile (vertical bars represent s.e.d. between groups).

**Conclusion** Of 23 genes known to be involved in FA metabolism, 4 were differentially expressed between sibling divergent for PUFA:SFA. SNPs in the regulatory regions of these genes could potentially be incorporated into the national breeding programme for sheep if significant associations are found.

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**References**

Galli, C., Simopoulos, A.P., Tremoli, E., eds. 1994. Effects of fatty acids and lipids in health and disease. World Review of Nutrition and Dietetics 76, 1–15
Serra, X., Gil, F., Perez-Enciso, M., Oliver, M.A., Vazquez, J.M., *et al.* 1998. Livestock Production Science 56, 215–223.
Wood, J.D., Enser, M., Fisher, A.V., Nute, R., Sheard, R., *et al*. 2008. Meat Science 78, 343– 358.
Zhang, S., Knight, T.J., Stalder, K.J., Goodwin, R.N., Lonergan, S.M., *et al*. 2009. Journal of Animal Breeding and Genetics 126, 259–268.

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034

# RNA-Seq analysis of whole blood from early lactation dairy cows reveals association of anti-viral response pathways with poor fertility

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**Application** We found differences in many pathways associated with viral responses in white blood cells (WBC) obtained soon after calving from cows with subsequent good and poor fertility. Eradicating viral diseases should benefit fertility.

**Introduction** Poor fertility is a major concern to the dairy industry. There are known associations with negative energy balance and/or uterine inflammation (metrits/endometritis) in early lactation. The latter has mainly been attributed to bacterial infections. There are also a number of endemic viral diseases of cattle such as BVDV, BHV and BRSV. These are known to reduce performance but their specific roles in fertility remain uncertain. The aim of this study was to compare the transcriptomic profiles of WBC from Holstein Friesian cows with differing fertility related phenotypes.

**Material and methods** Animals were recruited from 3 experimental farms in UK (AFBI Northern Ireland), Denmark (Aarhus University) and Ireland (University College Dublin). Blood samples were taken at 14 days in milk (DIM) and WBC from 48 animals were processed for total RNA-Seq using Illumina NextSeq 500 platform. Libraries were sequenced at 75 nt length single end reads to reach average 30 million reads/sample. Fastq raw files were quality controlled and mapped on *Bos taurus* UMD3.1.1 (Ensemble87 gene tracks). Phenotype data relevant to fertility included calving ease, body condition score (BCS) at calving, BCS and circulating IGF1 at 14 and 35 DIM, change in BCS and IGF1, total milk yield 0-50 DIM (TMY) and fertility (days to first service (DFS) and conception categorised as in calf by <100, 100-200 or >200 DIM (ICB). Component analysis was performed in R. Residuals were calculated based on REML prediction of phenotypes with herd and diet (nested within)as random effects. K-mean clustering over centre scaled phenotypes produced three significantly discriminant fertility clusters. Differentially expressed genes (DEG) between clusters were identified using DESeq2(1). Gene Set Enrichment Analysis (GSEA) and Mammalian Phenotype Ontology (MPO) were retrieved from MGI using EnrichR package (Chen *et al*., 2013).

**Results** The three clusters contained 17, 22 and 9 cows, respectively. Cluster 1 had a high BCS at calving, a high TMY and intermediate fertility. Cluster 2 calved with a low BCS which remained low, produced a medium TMY and had the worst fertility. Cluster 3 had the best fertility with the lowest DFS and ICB scores. These cows had a medium BCS at calving, high IGF1 at 14 DIM, a high BCS gain but the lowest TMY. GSEA of the WBC collected at 14 DIM identified 18 pathways which were differentially expressed between these three clusters with an adjusted P<0.003. The top pathway (adjusted P=5.4E-09) was MP:0002418, increased susceptibility to viral infection. Other significant pathways included MP:0008576, decreased circulating interferon-beta level; MP:0008563, decreased interferon-alpha secretion; MP:002376, abnormal dendritic cell physiology; MP:0002451, abnormal macrophage physiology and MP:0005348, increased T cell proliferation. Direct comparison between the good and poor fertility clusters produced 64 and 6 DEG which were significantly down- or up-regulated respectively in the less fertile cows. This list included at least 22 down-regulated genes known to be directly or indirectly regulated by interferons (*DDX58, DHX58, GBP5, HERC5, HERC6, IFI44L, IFI6, IFIH1, IFIT1, IFIT2, IFIT3, IFIT5, IRF7, ISG15, MX1, MX2, OAS1X, OAS2, RSAD2, SAMD9, UBA7, USP25*). Seven genes had known function in ubiquitination including ISGylation, a process which modifies proteins and targets them for destruction (*ISG15, DTX3L, ENSBTAG00000016661, HERC5, HERC6, UBA7, USP25*). Other down-regulated genes known to influence immune function included *ADAR, CD274, CMPK2, DCK, DUSP4, EIF2AK2, GBP5, MARCKSL1, MB21D1, PGLYRP1, PTX3* and *TEC*. Amongst these, the encoded proteins act as follows. CD274 is an immune inhibitory receptor ligand. CMPK2 regulates terminal differentiation of monocytes. PTX is induced by inflammatory cytokines and involved in regulating inflammation, complement activation and angiogenesis and TEC is an integral component of T cell signalling with a distinct role in T cell activation. Up-regulated genes included *LTF*, which encodes a major iron-binding protein in milk and body secretions with antimicrobial activity and *MARCO*, a class A scavenger receptor which binds both Gram-negative and Gram-positive bacteria.

**Conclusion** This analysis provides clear evidence that cows which go on to suffer poor fertility later in lactation exhibit major differences in immune function early postpartum. While some of the identified genes play known roles in pathways involved in dealing with bacterial infections, the majority are of greater importance in viral infections. This suggests that in order to improve fertility in dairy cows, greater emphasis needs to be given to reduce the spread of viral disease.

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**Reference**

Chen, E.Y., Tan, C.M., Kou, Y., Duan, Q., Wang, Z., Meirelles, G.V., *et al*. 2013. BMC Bioinformatics 14, 128.

035

# Common differentially expressed genes in muscle, adipose and liver of pigs differing in feed efficiency and intramuscular fat content

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**Application** Identification of molecular mechanisms common to key tissues with relevance to energy partitioning and meat quality would enhance potential to improve efficiency, quality and reduce the ecological footprint from pork production.

**Introduction** Feed efficiency (FE) is a measure of an animal's efficiency in relation to energy conversion from feed into economically important tissue and it is linked to a suite of systemic physiological changes, with muscle, liver and adipose tissue of relevance due to their significance in energy partitioning [1]. The objective of this study was to identify common biological processes occurring in FE-divergent muscle, liver and adipose tissue.

**Material and methods** RNA-sequencing libraries were prepared from muscle, adipose and liver of 16 divergent commercial line Maxgro x (Landrace x Large White) pigs (8 high/low FE). Following RNA sequencing with Illumina HiSeq2500, paired-end reads were mapped to the reference Sscrofa10.2 [2] using TopHat (2.1.0). Read counts were assigned to gene features using HTSeq 0.6.1 [3]. Identification of differentially expressed (DE) genes at the $p<0.05$ level in relation to FE was performed using DESeq2 package (3.4.0). Fixed effects for the analysis of DE genes in liver included RFI groups and slaughter dates, while for muscle and adipose included RFI groups and sows. Common DE genes were identified using Venny [4]. Functional analysis of shared DE genes was carried out in Ingenuity Pathways analysis.

**Results** 811 genes were DE in adipose, 1022 in muscle and 2739 in liver. 24 genes were commonly DE across all three tissues, while 69 were common to adipose and muscle and 194 and 178 were common to liver and muscle/adipose respectively (Figure 1). Most significant molecular and cellular functions in the three-way commonly differentially expressed genes can be seen in Table 2..Commonly differentially regulated genes in all three tissues included *DPP4*, *SLC2A4* which have functions in carbohydrate metabolism and *ABCA8, LMNB2, TYROBP*, with functions in lipid metabolism, while a number of behaviour-related genes including *CHL1, PRKG1, SQSTM1, TNFRSF1B,* were also perturbed in all three tissues.


**Figure 1** Numbers of commonly differentially expressed genes in liver, muscle and adipose in relation to FE

**Table 1** Molecular and cellular functions of commonly differentially expressed genes in liver, muscle and adipose of FE-divergent pigs

| Categories | p-Value | Genes |
|---|---|---|
| Carbohydrate Metabolism | 6.39E-06 | 3 |
| Metabolic Disease | 0.0000119 | 5 |
| Cardiovascular System Development and Function | 0.0000553 | 4 |
| Tissue Morphology | 0.0000553 | 4 |
| Behaviour | 0.000106 | 8 |
| Inflammatory Response | 0.000126 | 7 |

**Conclusion** Metabolic functions, in particular carbohydrate and lipid metabolism, tissue morphology and behaviour were perturbed in a systemic fashion in pigs divergent in feed efficiency.

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**References**

Patience, J.F., Rossoni-Serão, M.C., and Gutiérrez, N.A. 2015. A review of feed efficiency in swine: biology and application. Journal of Animal Science and Biotechnology 6(1), 33.
Kim, D., *et al*., 2013. TopHat2: accurate alignment of transcriptomes in the presence of insertions, deletions and gene fusions. Genome Biology 14(4).
Anders, S., Pyl, P.T. and Huber, W. 2015. HTSeq-a Python framework to work with high-throughput sequencing data. Bioinformatics 31(2), 166-169.
Oliveros, J.C. Venny. An interactive tool for comparing lists with Venn's diagrams. 2007-2015 http://bioinfogp.cnb.csic.es/tools/venny/index.html

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# Differential gene expression profiles of liver and muscle tissues in pigs divergent for feed efficiency

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**Application** Identification of differentially expressed genes/pathways in key metabolic tissues between inefficient and efficient pigs provides a greater understanding of the molecular events influencing feed efficiency. In addition, these genes are targets for the identification of functional SNPs that can assist breeding organisations to select more efficient animals.

**Introduction** By characterizing molecular changes in tissues known to influence feed efficiency, potential functionally important markers of the trait can be identified to select more feed efficient animals. Previously, changes in the expression of intestinal nutrient transporters have been identified in pigs utilized in the current study (Vigors *et al*., 2016). Liver and muscle are key tissues involved in influencing feed efficiency due to their fundamental importance in energy metabolism. The objective was to examine liver and muscle transcriptomic differences using RNA-sequencing to better understand the molecular mechanisms influencing feed efficiency and to identify a list of candidate biomarkers for RFI prediction.

**Material and methods** Two trials were conducted to identify pigs that were divergent in feed efficiency with pigs selected from two separate sources. In trial 1, male pigs (LW x LR x PIC337 boars (PIC Genetics)) (92 d.o., BW 41.35 kg (SD = 4.36)) were fed a standard finishing diet for a 43-day recording period prior to slaughter, to evaluate feed intake and growth and to calculate residual feed intake (RFI). When pigs were 146 d.o., animals designated high RFI (HRFI) (n=12) and low RFI (LRFI) (n=12) (average weight 93.26 kg, SEM 2.37 kg) were slaughtered to collect liver and muscle tissue. In trial 2, male pigs (LW x LR x Maxgro boars (Hermitage Genetics)) (63 d.o., BW 29.4 kg (SD = 2.03)) were evaluated as described above. When pigs were 115 d.o, animals (85 kg, SEM 2.8 kg), HRFI; (n=8) and LRFI; (n=8) were slaughtered and tissue samples were collected. Library preparation and RNA-sequencing were performed based on Illumina protocol. Differential expression analysis was conducted using the DESeq2 package of R. Common differentially expressed genes between liver and muscle were identified by Venny (Venny 2.1) and submitted for functional annotation clustering using DAVID.

**Results** In liver tissue, 484 genes were overexpressed and 477 genes were under-expressed in the HRFI group compared to the LRFI group. In muscle tissue, 6047 genes were overexpressed and 6371 were under-expressed in the HRFI compared to the LRFI group. Following functional annotation clustering, the GO terms *protein targeting to membrane* and *extracellular matrix organization* were identified as being upregulated in HRFI vs LRFI animals (Table1), while the GO terms *type I interferon signalling pathway* and *defence response* were identified as downregulated in the HRFI vs LRFI animals. Genes with the largest fold changes between RFI groups in liver included *PKIB*, *EPB42*, *SLC22A2*, and *TGM7,* while in muscle included *SLC01A2*, *S100G*, *HRG* and *PCK2*. These genes are potential targets for functional SNP identification as genetic markers of feed efficiency.

**Table1** Gene ontology terms overrepresented among differentially expressed genes in liver and muscle

| Term | Count | *P* value |
|---|---|---|
| **GO terms overrepresented among DEGs with higher expression in the HRFI group vs LRFI** | | |
| GO:0006612~protein targeting to membrane | 10 | 0.025945 |
| GO:0030198~extracellular matrix organization | 12 | 0.054506 |
| **GO terms overrepresented among DEGs with lower expression in the HRFI group vs LRFI** | | |
| GO:0060337~type I interferon signalling pathway | 10 | 2.91E-05 |
| GO:0006952~defense response | 32 | 9.10E-04 |

**Conclusion** In conclusion, genes involved in protein metabolism and extracellular matrix organization were upregulated in the HRFI group while the LRFI group had upregulation of terms involved in immunity. This study provides new knowledge on the genes influencing RFI in pigs and identifies genes for functional SNP identification.

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**References**

Vigors, S., Sweeney, T., O'Shea, C.J., Kelly, A.K. and O'Doherty, J.V. 2016. Animal 10, 1848-1855.

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# The effect of exogenous yeast enriched protein concentrate from the bio-ethanol industry (YPC) on pig cytochrome b (CYTB) genomic DNA content in faeces

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**Application** Feeding pigs a diet containing a yeast feed additive resulted in reduced faecal cytochrome b (CYTB) genomic DNA (gDNA) levels. There was a trend for a positive correlation between CYTB gDNA and Feed Conversion Ratio (FCR).

**Introduction** We previously showed xylanase inclusion in weaner pig diets reduced the levels of pig CYTB gDNA in the faeces, but measures of animal performance were not available (Slinger *et al.,* 2017). Dietary yeast affects gut morphology and animal performance (Muthusamy *et al.,* 2011). The present study assessed whether a yeast enriched protein concentrate (YPC) influenced CYTB gDNA content in pig faeces and whether there was a relationship with FCR.

**Material and methods** Newly weaned piglets (n=180) ((Large White x Landrace) x Pietrain) (8.37kg ±1.10) were assigned to one of four dietary treatment groups (n=9 pens per treatment, 5 animals per pen). The four diets differed only in yeast enriched protein concentrate from the bio-ethanol industry (YPC) (AB Agri) content: 0% (control), 2.5%, 5% and 7.5% (w/w). Feed intake and pig weight data was collected and used to calculate FCR. A single faecal sample was collected from each pen on days 14 and 28 of the 36 day trial. DNA was extracted using a modified phenol-chloroform method, where 30mg of faeces was homogenised in 600µl of Nuclei Lysis Solution (Promega) using a hand-held homogeniser, prior to a 2 step phenol-chloroform clean-up, followed by an RNase A (Promega) treatment step and a further phenol-chloroform clean-up step with final ethanol precipitation. SYBR Green (Roche) quantitative PCR (qPCR) was carried out on a LightCycler® 480 (Roche) instrument to assess the DNA composition of the faeces. Pig genomic DNA was detected using primers designed specifically for the pig cytochrome b (CYTB) gene, while bacterial DNA was detected using published 16S primers (Mieszkin *et al.,* 2009). PCR products were verified by Sanger Sequencing (Source BioScience, Nottingham, UK). All data was analysed using Genstat 17th Edition, with significance at P<0.05. Two-way (diet x day of trial) ANOVA was carried out on the DNA data and one-way (diet) ANOVA on the FCR data. The linear relationship between pig faecal CYTB gDNA content and the recorded FCR for both time points was assessed by Pearson's correlation.

**Results** There was no interaction between diet and day of trial on the pig CYTB gDNA content in their faeces, but there was a significant (P<0.05) effect of diet with no effect (P>0.05) of day (Fig 1). There was a significant (P<0.001) effect of diet on FCR at wean-day 14 (data not shown), but no effect (P>0.05) of diet on FCR for days 14-28. There was a trend (P=0.093) for a positive correlation (r=0.2839) between pig CYTB gDNA (at day 14) and FCR (wean-day 14) (Fig 2), but no significant correlation (P=0.393) at the later time point (data not shown).

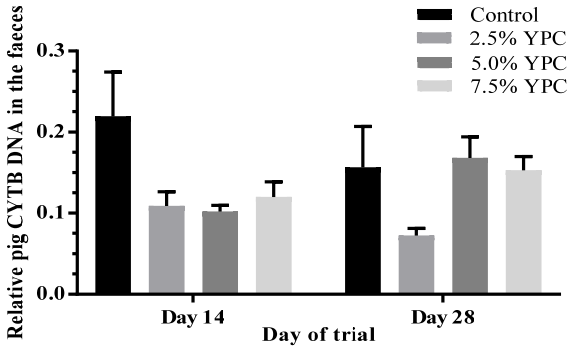


Figure 1: Effect of diet on pig faecal CYTB DNA contents at 14 and 28 days of treatment. Error bars are + SEM, n=9 per diet group.

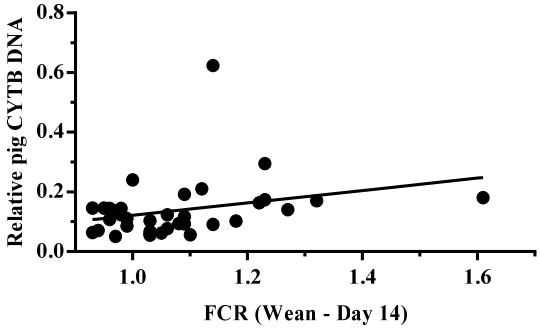


Figure 2: The correlation between animal FCR and the amount of host DNA in their faeces.

**Conclusion** Diet significantly affected the levels of pig CYTB gDNA in the faeces, but there was no effect of day. The effect appears more prominent at day 14, which is when the effect on FCR was also observed. The observed trend for a positive correlation between pig faecal CYTB gDNA content (day 14) and FCR (wean-day 14) indicates that the pig faecal CYTB gDNA content might potentially be a non-invasive marker of gut cell turnover, which reflects FCR.

**Acknowledgements** The work was funded by AB Agri, AB Vista and the University of Nottingham.

**References**

Mieszkin, S., Furet, J-P., Corthier, G. and Gourmelon, M. 2009. Applied and Environmental Microbiology 75(10), 3045-3054.
Muthusany, N., Halder, S., Ghosh, T. K. and Bedford, M. R. 2011. British Poultry Science 52(6), 694-703.
Slinger, K. R., May, K., Chang, A., Bedford, M. R., Parr, T., Brameld, J. M. Advances in Animal Biosciences 8 (1), 47.

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038

# Identification of gene networks contributing to compensatory growth in bovine ruminal epithelium

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**Application** This study provides an insight into the molecular control of compensatory growth (CG) in cattle. This data will contribute to identifying DNA based biomarkers to select for cattle with a greater ability to undergo CG.

**Introduction** Compensatory growth is defined as a physiological process whereby an animal has the potential following a period of restricted feed intake to undergo accelerated growth upon re-alimentation (Hornick *et al*., 2000). This naturally occurring phenomenon is commonly utilised in animal production settings to provide a means of reducing input costs through a reduction in overwintering feed costs (Keogh *et al*., 2015). However, although widely utilised, the biochemical mechanisms controlling this phenomenon are yet to be elucidated fully. The gastrointestinal tract, in particular the reticulo-rumen has been shown to be responsive to both dietary restriction and subsequent CG (Keogh *et al*., 2015). Therefore, the objective of this study was to perform gene network analysis of rumen papillae gene expression data of cattle undergoing CG in order to reveal more information on the interactions between genes governing the expression of CG in this organ.

**Material and methods** This study utilised rumen papillae tissue collected as part of a research programme designed to examine the physiological control of CG in growing beef cattle (Keogh *et al*., 2015). Briefly, 30 Holstein-Friesian bulls (mean live-weight 370±35 kg) were assigned to one of two groups: (i) restricted feed allowance for 125 days (RES; n=15) followed by *ad libitum* access to feed for 55 days or (ii) *ad libitum* access to feed throughout the trial (ADLIB; n=15). The first 125 days of the trial were denoted as Period 1 and the subsequent 55 days, Period 2. Target growth rates for RES and ADLIB were 0.6 kg day$^{-1}$ and in excess of 1.5 kg day$^{-1}$ during Period 1, respectively. Following completion of Period 2 all animals were slaughtered and rumen papillae samples collected from all animals. RNA isolation, cDNA library preparation, RNAsequencing and bioinformatics analysis were performed as per Keogh *et al*. (2017). The weighted gene co-expression network analysis (WGCNA) software package (Langfelder and Horvath, 2008) was used to identify modules of co-expressed genes, which were then correlated with traits associated with the expression of CG. Traits included average daily gain (ADG), CG-index, feed conversion ratio (FCR) and dry matter intake (DMI). RNAseq read count data were filtered for lowly expressed genes, normalised and then Log2 transformed in R. Networks of co-expressed genes were constructed using WGCNA within R. Unsigned, weighted correlation network construction and module detection was performed using the automatic one-step function, blockwise Modules. The resulting modules of co-expressed genes were assigned colour names by the software. Relationships between modules of co-expressed genes and trait data were then calculated by Pearson correlation. Modules with statistically significant ($P < 0.05$) correlations were selected for further analysis as potentially biologically interesting modules associated with the expression of CG. Gene ontology analysis was then performed on genes from each module identified as significantly correlated with trait data. The Database for Annotation, Visualization and Integrated Discovery (DAVID) was used for functional annotation of co-expressed genes within modules. Gene ontology terms were considered significant if the adjusted p-value was less than 0.05. The top hub genes of significant co-expressed modules were then visualized using Cytoscape software.

**Results** Cattle that underwent CG displayed accelerated growth, growing at 1.8 times the rate of their ADLIB counterparts in Period 2. WGCNA identified one module of co-expressed genes which was positively correlated with CG-index ($r = 0.7$, $P = 0.02$). Functional annotation of co-expressed genes in this module revealed biological processes involved in ATP binding, proteasome core complex, receptor binding and mitochondrial inner membrane as well as pathways involved in metabolism, cell adhesion molecules and endocytosis.

**Conclusion** Genes involved in metabolism, cellular interactions and energy production were significantly associated with CG. As the rumen is a highly metabolic organ, co-expression of genes involved in metabolism and cellular function during re-alimentation implies changes in the metabolic rate and cellular turnover in animals undergoing CG. This study provides an insight into the molecular mechanisms regulating the CG phenomenon in cattle. Additionally significantly co-expressed genes may represent potential biomarkers for the selection of CG in cattle.

**Acknowledgements** Authors acknowledge funding from Science Foundation Ireland (13/CDA/2182).

**References**

Hornick, J.L., Van Eenaeme, C., Gerard, O., Dufrasne, I. and Istasse, L. 2000. Domestic Animal Endocrinology 19, 121-132.
Keogh, K., Waters, S.M., Kelly, A.K. and Kenny, D.A. 2015. Journal of Animal Science 93, 3578-3589.
Keogh K, Waters, S.M., Cormican, P., Kelly, A.K., O'Shea, E. and Kenny, D.A. 2017. Plos One. 12(5), e0177852.
Langfelder, P. and Horvath, S. 2008. BMC Bioinformatics 9, 559.

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039

# Dynamic modelling of the core microbiota in the porcine gut

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**Application** Probiotic and prebiotic manipulation of the porcine gut core microbiota could have impacts on microbial short chain fatty acids (SCFAs) fermentation, affecting positively host health and performances.

**Introduction** Porcine core microbiota (CM) is a bacterial community present in the porcine gut microbiota, suggested to be independent of diet or breed (Holman *et al*., 2017). It could therefore represent an optimal and standardised probiotic target allowing modifications of microbiota-derived SCFA patterns, with benefits for host health and performance. Considering both *in vitro* and *in vivo* limitations, we developed a mathematical CM model to uncover, *in silico*, its possible role through comparison with the whole microbiota SCFA pattern found in literature, and to explore, *in silico*, the possibility of using CM genera and/or their substrates in the context of probiotic/prebiotic intervention.

**Material and methods** The porcine CM identified by a meta-analysis study (Holman *et al*., 2017), is composed of up to 20 genera, depending on the CM gut location. To develop our CM mathematical model, we modelled each genus as a bacterial unit (BU), summarising their species and pathways complexity, and modelling each BU as if it were a singular microorganism. The BU models were first validated comparing their output with experimental data from the study on which the BU stoichiometry is based. Where no cultural information was found, the stoichiometry assumption was based either on the theoretical description of the genus or on the phylogenetically closest genus with known data. Therefore, it is assumed that merging the BU models in a CM model, the latter output might be as similar as possible to a culture composed of the same genera. The mathematical formulation of the model consists of a system of ordinary differential equations (ODEs) based on a previous model (Kettle *et al*., 2015), characterising different kinds of substrates related to the different type of bacterial growth. The environment modelled is an *in vitro* fermenter, either batch or continuous culture, since we did not include any biotic factors in the system of ODEs. The model was solved using the microPop package in R (Kettle *et al*., 2017). The major SCFA and lactate concentrations from the model are compared with three different studies, which used faecal, caecal or colonic inoculum in either batch or continuous culture (Lin *et al*., 2011; Tanner *et al*., 2014; Ding *et al*., 2015). In each case, the model simulated the conditions described in the study, through the modification of the starting concentration value of inoculum and substrates. The model was also used to simulate the effect of probiotic and prebiotic approaches targeting each BU and their substrates. In this case, probiotic/prebiotic simulations were compared with the output of a model simulating the non-enhanced CM under the same experimental conditions.

**Results** The modelled SCFA pattern shows a great similarity to experimental data not used for developing the model. This indicates that the CM is largely responsible for the SCFA pattern produced by the whole microbiota. The small differences in acetate and butyrate production may indicate that CM provides acetate to the non-CM bacteria (Figure 1). The probiotic simulation showed that not all the BU enhancements had an appreciable effect on the SCFA pattern, whereas the prebiotic simulations showed, an increasing SCFA concentration, with all the substrates used as a prebiotic (data not shown). Thus, we simulated a therapy combining *Faecalibacterium BU*, *Treponema BU* and resistant starch, in a continuous culture.

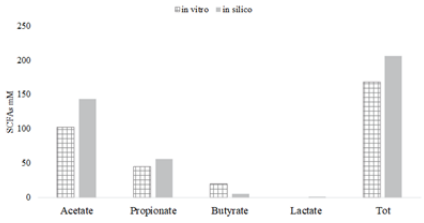


*Figure 1. SCFA production from a mixture of resistant starch 4.32 g/l), non-starch polysaccharides (7 g/l), protein (15 g/l) and sugars (2 g/l)* in vitro *by porcine faecal microbiota (Tanner et al., 2014) and* in silico *by porcine faecal core microbiota*

**Conclusion** Although a validation through *in vitro*/*in vivo* experimentation is necessary, our modelling data showed that the CM forms the basis of microbial SCFA production despite the limited number of CM genera, and it could provide acetate, used by the non-CM genera (e.g. producing butyrate). Furthermore, the model shows that a possible CM genera/substrate enhancing therapy could increase the total SCFA concentration with a five-fold increase in butyrate concentration.

**Acknowledgements** The authors gratefully acknowledge SRUC and BioSS for financially supporting this work.

**References**

Ding, H.H., Cui, S.W., Goff, H.D. and Gong, J. 2015. Bioactive Carbohydrates and Dietary Fibre 6, 62–68.
Holman, D.B., Brunelle, B.W., Trachsel, J. and Allen, H.K. 2017. mSystems 2, 1–14.
Kettle, H., Holtrop, G., Louis, P. and Flint, H.J. 2017. Methods in Ecology and Evolution 00:1–11.
Kettle, H., Louis, P., Holtrop, G., Duncan, S.H. and Flint, H.J. 2015. Environmental Microbiology 17, 1615–1630.
Lin, B., Gong, J., Wang, Q., Cui, S., Yu, H. and Huang, B. 2011. Food Hydrocolloids 25, 180–188.
Tanner, S.A., Berner, A.Z., Rigozzi, E., Grattepanche, F., Chassard, C. and Lacroix, C. 2014. PLoS ONE 9, e94123.

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**040**

# An analysis of the intestinal immune response of pigs divergent in feed efficiency in both an unchallenged state and following an *ex-vivo* LPS challenge using the Nanostring-based multigene assay

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**Application** This study provides new knowledge on the differences in immune function in pigs that are divergent in feed efficiency.

**Introduction** Improving feed efficiency is a key goal of the pig industry. However, our understanding of the ability of more efficient animals to respond to an infection is not fully elucidated. Previously, Vigors *et al*. (2016) identified a differential immune response in colonic tissue between pigs divergent in feed efficiency following an ex-vivo lipopolysaccharide (LPS) challenge. However, that study was limited to small number of inflammatory cytokine genes. Therefore, the objective of this study was to examine the gene expression profile of a range of targets involved in epithelial nutrient transport and immune function such as volatile fatty acid transporters, tight junction proteins, pattern recognition receptors and cytokines in: a) the basal unchallenged colonic tissue; and (b) following an ex-vivo LPS challenge of colonic tissue.

**Material and methods** Two trials were conducted to identify pigs that were divergent in feed efficiency with pigs selected from two separate sources. In trial 1, male pigs (LW x LR x PIC337 boars (PIC Genetics)) (92 d.o., BW 41.35 kg (SD = 4.36)) were fed a standard finishing diet for a 43-day recording period prior to slaughter, to evaluate feed intake and growth to calculate residual feed intake (RFI). When pigs were 146 d.o., animals designated high RFI (HRFI) (n=12) and low RFI (LRFI) (n=12) (average weight 93.26 kg, SEM 2.37 kg) were slaughtered to collect colonic tissue. In trial 2, male pigs (LW x LR x Maxgro boars (Hermitage Genetics)) (63 d.o., BW 29.4 kg (SD = 2.03)) were evaluated as described above. When pigs were 115 d.o, animals (85 kg, SEM 2.8 kg), HRFI; (n=8) and LRFI; (n=8) were slaughtered and two colonic tissue samples were collected. The two tissue sections were placed in 1ml of Dulbecco's modified Eagle's medium, one in the presence of bacterial LPS (*Escherichia coli* strain) at a concentration of 10 µg /ml while the other sample was used as a control. A custom nCounter panel was designed, which was capable of measuring 56 genes of interest as well as 15 internal reference genes. Gene expression analysis was performed on unchallenged tissue samples from both trial 1 (6 HRFI & 6 LRFI) and trial 2 (6 HRFI & 6 LRFI). The tissue samples from trial 1 and 2 were analysed together as a complete randomized design experiment with RFI and trial as main effects. Gene expression analysis was performed on the LPS challenged tissue samples from trial 2 (6 HRFI & 6 LRFI). Tissue samples from trial 2 were analysed as 2 x 2 factorial experiment with the statistical model including the effects of RFI and challenge (Unchallenged or LPS) and their associated two-way interactions.

**Results** In the unchallenged colonic tissue, the LRFI group had increased expression of *TNF-α, IL-8*, *IL-10, AP1* and *AOAH (P<0.05)* compared to the HRFI group. In the LPS challenged tissue the LRFI group had higher expression of *GPR43, JAK2*, *NFAM, TLR1*, *TLR7*, *TLR8* (P<0.05) compared to the HRFI group. An interaction between RFI group and LPS was observed with the HRFI group having a greater increase in expression of both *IL-8* and *IL-17* compared to the LRFI group following the LPS challenge (P<0.05).

**Table 1** Differentially expressed genes in unchallenged colonic tissue

| Gene | High | Low | Std Err | *P* value | Gene | High | Low | Std Err | *P* value |
|---|---|---|---|---|---|---|---|---|---|
| *TNF-α* | 50.2917 | 74.3758 | 5.40145 | 0.0048 | *AOAH* | 294.028 | 402.813 | 34.9091 | 0.0389 |
| *IL8* | 1013.47 | 1584.9 | 105.97 | 0.001 | *AP1* | 3010.02 | 3846.04 | 286.56 | 0.0474 |
| *IL10* | 12.3317 | 16.5708 | 1.41369 | 0.0461 | *STAT2* | 2207.13 | 2471.13 | 94.0913 | 0.0605 |

**Conclusion** Differences in the colonic tissue of HRFI versus LRFI pigs was identified in both the basal state and in colon challenged with LPS. These changes predominantly related to genes regulating cytokine function with changes in TNF-α regulated genes of particular interest due to its role in both the immune response and appetite regulation.

**Acknowledgements** This work is funded by the IdentiFEED project (13/S/519), supported by The Irish Department of Food, Agriculture and the Marine (DAFM) under the National Development Plan 2007-2013.

**References**

Vigors, S., O'Doherty, J. V., Kelly, A. H., O'Shea, C. J. and Sweeney, T. 2016. PLoS One 11: e0148145.

041

# Effects of bedding supplementation on the performance and caecal microbiome of broilers fed a wheat-based diet

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**Application** Supplementing bedding with excreta from mature birds acts as a probiotic and alters the gut microbiome of growing chicks, although this does not affect bird growth.

**Introduction** The gut microbiome of broilers is influenced by numerous factors including bedding and environment: colonisation of the gastrointestinal tract begins at hatch where birds ingest bacteria and the microbiome develops in a successional manner. Variations in gut microbes exist due to differences in nutrient composition, the use of additives; such as antimicrobials, and management factors such as bedding type (Torok *et al.,* 2009). Reusing bedding means that young birds are exposed to a complex 'soup' of microbes at an early age and therefore the development of the gut microbiome may be altered (Cressman *et al*, 2010, Wang *et al.,* 2016). It is hypothesised that reusing bedding can influence the development of a stable microbial population: this study investigated the effects of supplementing bedding with poultry excreta on the growth performance and caecal microbiome composition of broiler chickens.

**Material and methods** 168 male day old Ross 308 broiler chicks were randomly allocated to one of two treatment groups: fresh wood shavings (Fresh) or fresh wood shavings supplemented with excreta from healthy adult birds (Supplemented; 50 g per 1 x 0.5 m pen). Birds were housed in one of two brooding pens according to their treatment for the first 15 days of the experiment, after which they were moved to one of 12 (n=6 per treatment) pens (14 birds/pen) for the remainder of the study. All birds were fed the same wheat-based diet: starter days 1-15, grower/finisher days 15 onwards. At days 1, 15 and 22, birds and feed were weighed for the calculation of FCR. At days 15 and 22, one bird per pen was randomly selected, sacrificed and samples of caecal digesta were taken. DNA was extracted and the microbiome was profiled through 16S rRNA sequencing on the Illumina platform. Data were also collected at days 29 and 35 but are not presented here. The effects of bedding on the relative abundance of bacterial phyla was investigated through Linear Discriminant Analysis Effect Size and effects on alpha diversity and performance were analysed through ANOVA where significance was denoted as $P<0.05$.

**Results** No differences in performance data were identified (Table 1).

**Table 1** Effects of bedding on bird performance.

| | Fresh | Supplemented | SEM | *P*-value |
|---|---|---|---|---|
| Liveweight day 15 (g) | 439 | 427 | 6.0 | 0.351 |
| Liveweight day 22 (g) | 876 | 874 | 10.3 | 0.929 |
| Average daily feed intake/bird 15-22 (g) | 91.1 | 95.5 | 3.56 | 0.544 |
| FCR day 15-22 | 1.49 | 1.52 | 0.03 | 0.699 |

A number of bacterial taxa were differentially represented within each treatment group at day 15 ($P<0.05$), including an increase in *Lachnospiraceae* and decrease in *Clostridiaceae* in birds on supplemented bedding. Alpha diversity was also significantly heightened in samples from birds on supplemented bedding ($P<0.001$). Conversely no differences were identified at day 22 ($P>0.05$). It was observed that day 15 supplemented samples were more similar to fresh samples taken at day 22 with no statistically significant differences in either taxa abundance or alpha diversity.

**Conclusion** These results suggest that the supplementation of bedding with excreta from healthy adult birds accelerates the development of a stable, adult microbial population, however this did not affect bird performance.

**References**

Torok, V.A., Hughes, R.J., Ophel-Keller, K., Ali, M. and Macalpine, R. 2009. Poultry Science 88, 2474-2481.
Cressman, M.D., Yu, Z., Nelson, M.C., Moeller, S.J., Lilburn, M.S. and Zerby, H.N. 2010. Applied Environmental Microbiology 76, e6572.
Wang, L., Lilburn, M. and Yu, Z. 2016. Frontiers in Microbiology 7, e593.

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# Supplementation of the newly weaned piglet diet with a combination of casein hydrolysate and yeast β-glucan checks post-weaning diarrhoea

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**Application** Post-weaning disorders, symptomized by diarrhoea, weight loss and mortality, accounts for huge economic losses to the pig industry. This study identifies a combination of casein hydrolysate and yeast β-glucan that checks post-weaning diarrhoea with no negative effects on growth.

**Introduction** Nutritional strategies have been employed to overcome post-weaning disorders and improve growth and health of pigs, especially after the restricted usage of antibiotics and zinc. In one of our previous studies, post-weaning diarrhoea was reduced by supplementing piglet feed with a combination of a 5 kDa fraction of casein hydrolysate (5K) with yeast β-glucan (BG) (Mukhopadhya *et al*., 2016). However, commercial production of 5K involves higher costs compared to the casein hydrolysate. Hence, the objective for this study was to investigate the effects of supplementing post-weaned piglets diet with two casein hydrolysates; whole hydrolysate (EH) or the previously used 5K fraction alone or in combination with yeast β-glucan (BG) on diarrhoea scores and growth of piglets, immediately post-weaning.

**Material and methods** In a complete randomised 2×3 factorial design, 144 newly weaned piglets [progeny of Meatline boars x (Large White x Landrace) sow)], 28 d old, with BW 7.3 ± 0.2 kg were blocked on the basis of sex, body weight and litter of origin and assigned to one of the following dietary groups: 1) control diet (CD); 2) CD + 0.25 g/kg BG; 3) CD + 0.25 g/kg 5K; 4) CD + 0.25 g/kg BG + 0.25 g/kg 5K; 5) CD + 0.25 g/kg EH and 6) CD + 0.25 g/kg BG + 0.25 g/kg EH . The CD was formulated to contain 210 g/kg of crude protein, 15 MJ/kg of digestible energy (DE) and 15.5 g/kg of total lysine. Piglets (3/pen) were housed on fully slatted floors (1.68 × 1.22 m) at an ambient controlled environmental temperature (30°C) with ad libitum access to feed and water. The animals were weighed at the start of the trial (d 0) and on d 10 and all feed were weighed back on d 10 to calculate feed intake. Faecal consistencies based on visual observations were scored twice daily using a scoring system, which assigns scores ranging from 1 to describe hard, firm faeces to 5 which describes watery, mucous-like faeces (severe diarrhoea) (normal scores between 2 and 3). The faecal scores were checked for normality using PROC Univariate and analysed by repeated-measures analysis using the PROC MIXED procedure of SAS using the pen as an experimental unit. The growth parameters were analysed as a 3×2 factorial design using the PROC GLM procedure of SAS using the pen as an experimental unit. Data presented are expressed as least-squares means with their standard errors of the mean (SEM).

**Results** The inclusion of BG and casein hydrolysates significantly decreased faecal scores (Table 1). Addition of casein hydrolysates in presence of BG significantly lowered faecal scores compared to inclusion of BG or casein hydrolysates alone. Weights on d 10, average daily gain (ADG), average daily feed intake (ADFI) and gain:feed ratio (G:F) are also presented in Table 1. The inclusion of BG or casein hydrolysates had no significant effect of the measured parameters apart from ADFI.

**Table 1** Effect of diet supplementation on overall faecal scores, ADG, ADFI and G:F in post weaning piglets

| BG | No | | | Yes | | | SEM | *Significance* | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Casein hydrolysate | 0 | EH | 5K | 0 | EH | 5K | | BG | casein hydrolysate | BG x casein hydrolysate | Time |
| Faecal score | 3.50 | 3.22 | 2.93 | 2.99 | 2.67 | 2.81 | 0.062 | *0.001* | *0.001* | *0.0008* | *0.001* |
| Weight (kg) | 8.96 | 8.58 | 8.96 | 9.22 | 9.36 | 8.8 | 0.262 | ns | ns | ns | na |
| ADG (g/day) | 169 | 130 | 165 | 191 | 196 | 150 | 16 | ns | ns | ns | na |
| ADFI (g/day) | 299 | 247 | 275 | 306 | 277 | 270 | 27 | ns | *0.001* | ns | na |
| G:F | 0.58 | 0.40 | 0.58 | 0.6 | 0.68 | 0.53 | 0.088 | ns | ns | ns | na |

ns = not significant; na = not analysed

**Conclusion** In conclusion, the low cost EH in combination with BG was associated with reduction of faecal scores and maintenance of growth of the newly weaned piglet throughout the experimental period. These results warrant in-depth analysis of the gastrointestinal tract morphology and the gut microbial composition to understand the observed effects.

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**References**

Mukhopadhya, A., O'Doherty, V.J., Noronha, N., Ryan, M.T., Sweeney, T. 2016. Journal of Animal Science 94 (Suppl5), 453-453.

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# Network analysis to study ruminal microbial community and functional microbial gene interactions associated with methane emissions in beef cattle

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**Application** Network analysis is a promising method to predict species-species interactions and microbial gene abundances in rumen ecosystems occupied mostly by uncharacterized microorganisms. The results improve our understanding of the main methanogens associated with methane emissions and their main functional genes and can be used to improve the prediction of methane emissions to be used for breeding and nutritional intervention.

**Introduction** Previous shotgun metagenomic analyses of ruminal digesta identified some microbial information within the microbial communities and their functional genes that might be useful as biomarkers to select cattle that emit less $CH_4$ (Wallace *et al*., 2015, Roehe *et al.* 2016 and Auffret *et al.* 2017). However, identification and validation of robust biomarkers are needed and the biological mechanisms underlying $CH_4$ emissions should be uncovered.

**Material and methods** This study was performed using data of 50 animals from 3 independent experiments (Rooke *et al.* 2014, Duthie *et al.*, 2016, 2017), balanced for breed (Aberdeen Angus, Charolais, Limousin and Luing) and fed with different diets (forage or concentrate). Methane emissions were measured individually during 48 h in respiration chambers (Rooke *et al.* 2014). Total DNA was extracted from post-mortem rumen digesta samples to perform metagenomics analysis. Functional microbial genes were identified based on the KEGG genes database. For phylogenetic analysis, the genomic reads were aligned to the Kraken database (Wood and Salzberg, 2014). Data were expressed as relative abundances of microbes or genes within individual. A network analysis (Miru, Kajeka Ltd, Edinburgh UK) was performed using 200 microbial genera (MG) and 56 microbial functional genes (MFG), all highly correlated to $CH_4$ emissions. This study focused on *Methanobrevibacter* as main methane producer (Auffret *et al.* 2017). Partial least square analysis (PLS, Version 9.1 for Windows, SAS Institute Inc., Cary, NC, USA) was used (diet as fixed effect) to identify the functional genes most correlated with microbial populations variability.

**Results** A Network analysis (R = 0.75) including 200 MG and 56 MFG grouped *Methanobrevibacter*, *Methanothermus*, *Methanosphaera* and *Methanotorris* in a single cluster with almost all the functional genes considered (52/56). Most of the genes (35/52) were associated with *Methanobrevibacter* explaining 91.4% of the variation in *Methanobrevibacter* abundance. Moreover, the PLS analysis also helped to identify which genes (n=35) are mostly associated (VIP) with *Methanobrevibacter* as indicator of it´s most important metabolic functions. In general, the MGF with high VIP value (Table 1) were genes mostly involved in the hydrogenotrophic methanogenic pathway. More importantly a gene (K01079) associated with amino acid metabolism was identified with the highest VIP value. Finally, a lack of connection with other clusters containing bacteria, fungi, protists and other archaea was observed.

**Table 1** PLS results identifying the main functional genes explaining *Methanobrevibacter* abundance variability.

| Factor | VIP[1] | Coefficient | Microbial gene function |
|---|---|---|---|
| K01079 | 1.21 | 0.09 | phosphoserine phosphatase |
| K00584 | 1.20 | 0.08 | tetrahydromethanopterin S-methyltransferase subunit H |
| K00399 | 1.18 | 0.08 | methyl-coenzyme M reductase alpha subunit |
| K00201 | 1.17 | 0.07 | formylmethanofuran dehydrogenase subunit B |
| K14128 | 1.15 | 0.06 | F420-non-reducing hydrogenase subunit G |
| K00123 | 1.08 | 0.04 | formate dehydrogenase, alpha subunit |

[1]VIP: Variable Importance in Projection.

**Conclusion** This study improves our understanding of microbial community variability and their main metabolic functions associated with ruminal methane production. The knowledge is used to identify robust biomarkers associated with methane emissions from cattle.

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**References**

Auffret, M.D., Stewart, R., Dewhurst, R.J., Duthie, C-A., Rooke, J.A., *et al*. 2017. Frontiers in Microbiology (accepted).
Duthie, C-A., Haskell, M., Hyslop, J.J., Waterhouse, A., Wallace, R.J., *et al*. 2017. Animal 11**,** 1-10.
Duthie, C-A, Rooke, J.A., Troy, S., Hyslop, J.J., Ross, D.W., *et al*. 2016. Animal 10**,** 786-795.
Roehe, R., Dewhurst, R.J., Duthie, C-A., Rooke, J.A., McKain, N., *et al*. 2016. Plos Genetics, 12, e1005846.
Rooke, J.A., Wallace, R.J., Duthie, C-A, McKain, N, de Souza, SM., *et al*. 2014. British Journal of Nutrition 112, 398–407.
Wallace, R.J., Rooke, J.A., McKain, N., Duthie, C-A., Hyslop, J.J., *et al*. 2015. BMC Genomics 16, 839-852.
Wood, D. E. and Salzberg, S. L. 2014. Genome Biology and Evolution 15**,** R46.

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044

# Investigation of rumen bacterial and archaeal community profiles of bulls phenotypically divergent for residual feed intake

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**Application** Selection of animals with improved feed efficiency has the potential to maximise nutrient utilisation from feed and reduce feed costs in tandem with decreasing agricultural GHG emissions.

**Introduction** The rumen microbial community affords ruminants the ability to digest cellulose rich feed stuff and to convert such material to meat and milk. Phenotypic performance is dependent on a combination of its genotype, inherent microbiome and the chemical composition and quantity of feed offered. Residual feed intake (RFI) is widely used and a highly accepted measure of feed efficiency in cattle (Herd and Arthur, 2009). Shabat *et al.* (2016) previously found rumen microbiome genes and species could accurately predict an animals feed efficiency phenotype. The objective in this study was to explore the effect of RFI phenotype on the archaeal and bacterial populations present in the rumen of young Simmental bulls offered a finishing diet.

**Material and methods** Residual feed intake was calculated for each animal as the difference between actual dry matter intake (DMI) and expected DMI for a combination of four cohorts of Simmental bulls (n=87) over four years. All animals were managed similarly from birth and offered *ad libitum* concentrate and 3kg of grass silage daily during the finishing period. Animals were slaughtered at a mean bodyweight ± SD of 580.26 ± 74.09 kg. Liquid and solid rumen digesta were collected immediately after slaughter. The volatile fatty acids (VFA) concentration of rumen fluid was measured using gas chromatography. Rumen solid and liquid samples from cattle for the top (mean RFI= 0.85) and bottom (mean RFI=-0.76) sextiles were subjected to microbiomic analysis (n=60). Six samples were mislaid leaving n=54. DNA was extracted using the repeated bead beating and column purification method (Yu and Morrison, 2004). *16S* DNA libraries were constructed following extraction of DNA from rumen digesta and sequenced on the Illumina MiSeq platform. Four samples were removed from the analysis due to low sequence output (n=49). A combination of *de novo* and reference based operational taxonomic unit (OTU) identification was carried out, clustering sequences at 97% similarity and aligning a representative sequence from each clustered OTU to the Greengenes database (version gg_13_8). A non-parametric statistical analysis between treatment groups was conducted with R. Diversity metrics were calculated using PRIMER7. Spearman's correlation analysis was conducted hmisc package R Studio (v3.1).

**Results** No difference in alpha or beta diversity was observed between low RFI (LRFI) and high RFI (HRFI) animals for either digesta type (P>0.05). At a genus level, Spearman correlation analysis indicated that *Fibrobacter* was negatively correlated (P<0.05) with feed efficiency of bulls in both solid and liquid fractions (Figure 1a). The relative abundance of *Fibrobacter* was approximately two fold higher in LRFI in comparison to HRFI animals in both solid and liquid digesta (Figure 1b). *Fibrobacter* was also positively correlated with relative rumen acetate concentration while negatively correlated with relative propionate concentration (P<0.05). LRFI animals showed an increase in acetate:propionate ratio (P<0.05), with *Fibrobacter* abundance correlated with this ratio (P<0.01). At OTU level, there was a positive correlation observed between the OTUs identified as *Selenomonas ruminatium* and *Fibrobacter succinogenes* in both solid and liquid digesta (P<0.001). OTUs assigned to the phlya of *Tenericutes* and *Cyanobacteria* may also potentially influence feed efficiency phenotype(P<0.05).

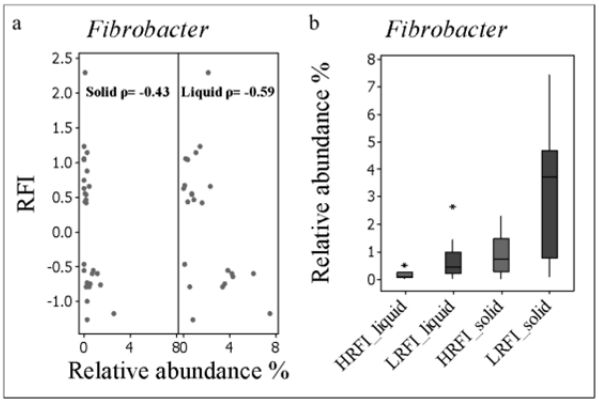


Figure 1. a) Scatterplot of relative abundance of *Fibrobacter* versus RFI in rumen solid and liquid digesta. b) Boxplot illustrating the relative abundance of *Fibrobacter* in rumen solid and liquid digesta of LRFI and HRFI bulls

**Conclusion** This study suggests that RFI phenotype does not affect the overall diversity of the bacterial or archaeal communities present in the rumen. However, we provide evidence that certain bacterial genera and bacterial and archaeal OTUs may influence the phenotypic expression of RFI potentially through their role in ruminal degradation of complex plant polysaccharides.

**References**

Herd, R. and Arthur, P. 2009. Journal of Animal Science 87, E64-E71.
Shabat, S. K. B., Sasson, G., Doron-Faigenboim, A., Durman, T., Yaacoby, S., Miller, M. E. B., White, B. A., Shterzer, N. and Mizrahi, I. 2016. The ISME Journal.
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**045**

# Using systems level multi-omic approaches to compare temporal bacterial colonisation of *Lolium perenne*, *Lotus corniculatus* and *Trifolium pratense* in the rumen

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**Application** Understanding bacterial colonisation of fresh forages in the rumen will allow the development of novel techniques to increase the efficiency of nutrient utilisation in ruminants, increasing animal productivity.

**Introduction** The growing population and increasing demand for meat and milk will require changes to animal production to maintain food security. A key problem is that the breakdown of plant matter in ruminant animals by is not very efficient as much energy is lost as waste, some of this waste being damaging to the environment. An increase in efficiency of digestion is a possible solution both increasing the meat and milk produced while minimising environmental impact. As much of ruminant nutrition comes from rumen microbes, understanding interactions of the rumen microbiome with the forage is paramount. Bacterial colonisation of perennial ryegrass has been previously investigated but other forages which would be present in a grazing situation have not been extensively studied. In this experiment we investigated bacterial colonisation of perennial ryegrass (PRG) red clover (RC) and birds foot trefoil (BFT) and plant chemical changes over a 24 h period.

**Material and methods** *Lolium perenne* cv. AberDart (PRG), *Lotus corniculatus* cv. Maitland (BFT) and *Trifolium pratense* cv. Milvus (RC) were grown from scarified seed in a greenhouse maintained at a 20/ 10$^{o}$C day/ night cycle with a minimum 10h photoperiod. BFT and RC were harvested after 16 weeks by removing the leaves and cutting them in half and PRG was harvested following 6 weeks' growth by cutting into 3cm lengths. The harvested plant matter was incubated in anaerobic incubation buffer (135mL at 39$^{o}$C; Van Soest, 1967) and rumen fluid inoculum (15 mL, strained through two layers of muslin and held under $CO_2$ at 39$^{o}$C; rumen fluid was taken from 3 cannulated cows, and pooled). Samples were taken for each forage at 0, 1, 2, 3, 4, 6, 8, 12, 24 hours and the experiment was repeated 3 times (n=9). Attached bacteria were isolated through overnight incubation in glutaraldehyde (3% v/v in PBS) at 4$^{o}$C followed by centrifugation (10,000 x g, 10 mins) and freeze drying of the pellet. DNA was extracted from the pelleted attached bacteria at each time point using the BIO101 FastDNA® SPIN Kit for soil (Qbiogene, Cambridge, UK). 16S rDNAPCR using 799F2 and 1177R primers, coupled with adaptors, was conducted followed by ion torrent sequencing. Operational taxonomic units at 97% identity were produced by removing the low quality sequences and sequencing errors using the CD-HIT-OUT pipeline and classified against the Greengenes database using MOTHUR. PICRUSt analysis was used to predict the metabolic function of the attached bacteria. Chemical analysis of ground plant matter was performed using Fourier transform infrared spectroscopy (FT-IR). Plant degradation for each plant sample was also evaluated at each time point

**Results** Analysis of bacterial colonisation identified that in all forages there was a dramatic change in genera and abundance at around 4-6 hours. This change was associated with the change from primary colonisation where microbes attach to the surface of plant matter, to secondary colonisation where microbes form stable colonies encompassed in biofilms. On the genus level, the main initial primary colonisers were *Prevotella*, *Pseudobutyrivibrio*, *Ruminococcus*, *Olsenella*, *Butyrivibrio* and *Anaeroplasma* on all forages. *Pseudobutyrivibrio* and *Anaerolasma*on was then more prevalent during secondary colonisation timepoints on all forages. For PRG the change was around 4 h which is in line with previous data from Huws *et al* (2016). However, in BFT and RC this change occurs later at around 6 h. The FT-IR data showed a similar trend in the forages indicating a change in plant chemistry around 4 h in PRG and a change around 6 h in BFT and RC most likely due to a change in polysaccharide content. PICRUSt analysis of the data showed there was a reduction in lipid and amino acid metabolism in the later time points (8 h onwards) compared with the early time points (1-4 h) which is likely due to a reduction in the availability of nutrients.

**Conclusion** This study demonstrates that the attached bacteria on PRG, BFT and RC were similar in composition and abundance but temporal differences were observed at different incubation times between the forages. This indicates that fundamental differences in plant chemistry have a major role in the functional breakdown of plant matter even though the colonising bacteria follow similar successions.

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**References**

Huws, S., Edwards, J., Creevey, C., Rees Stevens, P., Lin, W., Girdwood, S., Kingston-Smith A. 2016. FEMS Microbiology Ecology 92 (1), fiv137, https://doi.org/10.1093/femsec/fiv137

046

# The effect of an isoflavonoid-rich liquorice extract on fermentation and methanogenesis in the Rumen Simulation Technique

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**Application** The inclusion of an isoflavonoid-rich extract from liquorice in the diet could potentially improve the efficiency of the feed utilization by ruminants.

**Introduction** Since the ban of antibiotics as growth promoting feed additives by the European Union in 2006, plant extracts and plant secondary metabolites have been considered as alternatives to manipulate rumen fermentation (Hart *et al*., 2008). Flavonoids have recently gained interest because of their wide range of biological activities, particularly antimicrobial properties (Oskoueian *et al*. 2013). In this study, we tested an extract of liquorice, rich in prenylated isoflavonoids, and particularly glabridin, for its long-term effect on rumen fermentation and methanogenesis.

**Material and methods** The Rumen Simulation Technique (Czerkawski and Breckenridge 1977) was used to study the effect of a control diet alone or supplemented with liquorice extract (Licogen powder, Fitness R Us Ltd, Kiryat Shmona, Israel) at 1 g/L or 2 g/L. A single incubation period using 12 vessels inoculated with rumen fluid from four different sheep (four replicates) was carried out. The experiment lasted for 16 days, using the first 12 days for adaptation and the last 4 for sampling. Fermentation gases were collected in gas-tight bags to measure total gas and methane production. Daily production of ammonia and VFA were determined in the overflow flasks. Data were analysed statistically by randomized block ANOVA, with individual sheep as a blocking term.

**Results** When liquorice was added at 1 g/L no negative effects on fermentation were observed, although ammonia production decreased (-51%; P<0.001). The addition of 2 g/L, however, had a strong effect decreasing total VFA concentration (P=0.014), shifting fermentation towards propionate (P=0.012) at the expense of acetate (P=0.003), as well as dramatically decreasing ammonia production (-77%; P<0.001). Although total gas production was not affected by the inclusion of 2 g/L liquorice in the diet, methane production decreased (P<0.05) by 35% (Table 1).

**Table 1** Effect of supplementing a control diet (C) with liquorice extract (L1 and L2, 1 and 2 g/L, respectively) on fermentation products and methanogenesis in the Rumen Simulation Technique

| Diets | C | L1 | L2 | SED | P |
|---|---|---|---|---|---|
| Fermentation products (mmol/d) | | | | | |
| Total VFA | 33.7$^{b}$ | 34.1$^{b}$ | 28.5$^{a}$ | 1.056 | 0.003 |
| Acetate | 17.2$^{b}$ | 17.0$^{b}$ | 12.8$^{a}$ | 0.795 | 0.003 |
| Propionate | 3.55$^{a}$ | 4.06$^{a}$ | 4.94$^{b}$ | 0.313 | 0.012 |
| Butyrate | 8.08 | 7.95 | 7.56 | 0.333 | 0.332 |
| BCVFA | 3.51$^{b}$ | 3.74$^{b}$ | 0.482$^{a}$ | 0.213 | <0.001 |
| Ammonia | 1.37$^{c}$ | 0.674$^{b}$ | 0.315$^{a}$ | 0.106 | <0.001 |
| Gas emissions | | | | | |
| Total gas (L/d) | 1.2 | 1.23 | 1.3 | 0.069 | 0.384 |
| Methane (mmol/d) | 4.67$^{b}$ | 4.35$^{b}$ | 3.06$^{a}$ | 0.445 | 0.024 |
| Methane (mmol/g DOM) | 0.510$^{b}$ | 0.492$^{b}$ | 0.374$^{a}$ | 0.043 | 0.039 |

$^{a-b}$Means with different superscript differ ($P<0.05$). BCVFA: branched chain volatile fatty acids. DOM: digestible organic matter.

**Conclusion** Liquorice extract could potentially be used in ruminant diets to increase the efficiency of nitrogen utilization. The effects observed on rumen fermentation could be attributed to the high content of isoflavonoids, although the contribution of other phytochemicals cannot be ruled out.

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**References**

Czerkawski, J. W. and Breckenridge, G. 1977. British Journal of Nutrition 38, 371-384.
Hart, K. J., Yanez-Ruiz, D. R., Duval, S. M., McEwan, N. R. and Newbold, C. J. 2008. Animal Feed Science and Technology 147, 8-35.
Oskoueian, E., Abdullah, N. and Oskoueian, A. 2013. BioMed Research International. DOI:10.1155/2013/349129.

047

# Effects of dietary nitrate and lipid supplementation on the rumen microbiota associated with methane production

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**Application** Next generation sequencing (NGS) showed diet differences in microbial populations associated with methane emissions.

**Introduction** Diet manipulation has previously been shown to be an effective methane mitigation strategy, whilst NGS has enabled a more rapid and detailed description of the rumen microbiota. Poulsen *et al.* (2013) showed reductions in methane emissions in response to rapeseed oil supplementation and Thermoplasmata (methylotrophic archaea, such as Thermoplasmata) produced more methane than hydrogenotrophic archaea (Methanobacteriales). The aim of this study was to assess effects of dietary lipid and nitrate supplementation on the rumen microbiota.

**Material and methods** Rumen samples from 79 finishing steers were collected through a naso-gastric tube at the end of a 56 d feed efficiency measurement period and stored frozen prior to analysis. Diets were: control (C; total mixed ration based diet, n=20), and 3 diets in which rapeseed meal was replaced with nitrate (N; 18 g nitrate/kg DM as calcium nitrate, n=20), oil (O; increased by 12 g/kg DM using maize distillers dark grains, n=20), and NO (a combination of nitrate and oil, n=19). Methane emissions were measured in respiration chambers (2d) after the feed efficiency measurement period. Frozen rumen samples were crushed in liquid nitrogen, libraries prepared by amplification of the 16S rRNA gene (V4 region) and sequenced using an Illumina MiSeq. Demultiplexed sequences underwent quality control and taxonomic profiles were generated. Principal component analysis was carried out (Unscrambler X, Camo, Norway) using genus level information. Diet effects on relative abundances (RA) of genera were estimated using ANOVA (Benjamini-Hochberg false discovery rate) in the STAMP statistical package (kiwi.cs.dal.ca/Software/STAMP). RAs for each treatment group were subsequently compared to C, with particular interest in taxa associated with methane emissions.

**Results** In total, 184 taxa were recorded at the genus level. Comparison of PC-1 vs. PC-2 showed a slight grouping of O was observed, PC-1 vs. PC-3 showed two groupings (i) C, N, NO and (ii) O – data not shown here. 80 genera were significantly different (P<0.05) between N and C dietary treatments, 53 between NO and C and 16 between O and C. VadinCA11 (Thermoplasmata) increased in C diet relative to others (N & O P<0.001; NO P=0.015). Relative to C, methane (expressed as g/d or g/kg DMI) was significantly reduced by the addition of nitrate. Whilst a numerical reduction was observed for the addition of oil, these were non significant (Duthie *et al*., 2017). When assessing taxa associated with methane emissions (Table 1), Methanobacteriales (Methanosphera and Methanobrevibacter) were reduced for the N diet (P=0.008; P=0.04) and NO (P=0.351; P=0.164), but increased for the O diet (P=0.054; P=0.18). The increase in Thermoplasmata for the C dietary treatment is probably explained by the presence of trimethylamine precursors in the rapeseed meal of the C dietary treatment.

**Table 1** Differences in mean RA (s.d.) of taxa associated with methane emissions between dietary treatments and C

| Taxonomy | C<br>Mean RA (s.d.) | N<br>Mean RA (s.d.) | NO<br>Mean RA (s.d.) | O<br>Mean RA (s.d.) |
|---|---|---|---|---|
| VadinCA11 | 0.62 (0.16) | 0.30 (0.12) | 0.42 (0.19) | 0.23 (0.14) |
| Methanosphera | 0.09 (0.06) | 0.03 (0.03) | 0.07 (0.05) | 0.16 (0.08) |
| Methanobrevibacter | 6.43 (3.27) | 3.91 (2.09) | 5.14 (2.24) | 9.87 (5.75) |
| VadinCA11 | | P < 0.001 | 0.015 | P < 0.001 |
| Methanosphera | | 0.008 | 0.351 | 0.054 |
| Methanobrevibacter | | 0.04 | 0.164 | 0.18 |

**Conclusion** Taxa associated with methane production within the N dietary treatment were significantly reduced relative to C dietary treatment. It is likely that the addition of oil did not have a large effect on methane emissions due to reductions in Thermoplasmata being counteracted by increases in Methanobacteriales.

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**References**

Poulsen, M., Schwab, C., Jensen, B. B., Engberg, R. M., Spang, A., Canibe, N., Hojberg, O., Milinovich, G., Fragner, L., Schleper, C., Weckwerth, W., Lund, P., Scramm, A., Urich, T. 2013. Nature Communications 4, 1482.
Duthie, C-A, Troy, S. M., Hyslop, J. J., Ross, D., Roehe, R., Rooke, J. A. 2017. Animal 11. doi:10.1017/S175173111700146X.

048

# *Post mortem* observations on reticuloruminal and caecal content, ruminal histology and gene expression of beef finisher cattle in north-eastern Scotland

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**Application** Despite being essentially barley-based, the rations fed to cattle in beef finisher units in Scotland are diverse and there is wide inter-farm variation in parameters relating to acidosis. The proportion of fine particles in the TMR is at least as important a driver of metabolic adaptations as the amount of barley in the ration.

**Introduction** Beef cattle are often grown and finished for slaughter on diets that are rich in cereal grains. Sub-acute ruminal acidosis (SARA) is an inflammatory syndrome arising from high dietary challenge with soluble carbohydrates that is prevalent in dairy and beef cattle fed on high concentrate rations. Some farms are more susceptible to acidosis than others, and within herds some animals are more prone to acidosis than others. The objectives of this study were to 1) characterise the pathological features of the reticulorumen in cattle from divergently managed intensive beef finishing farms, and 2) attempt to relate differences in the severity or nature of these response variables to specific dietary or management practices.

**Material and methods** At the time of slaughter, we obtained samples from 19-20 animals from each of six beef finishing units (119 animals), with diverse feeding practices, which had been classified as being high-risk (three farms) or low-risk (three farms) for SARA on the basis of the proportion of cereal starch and neutral detergent fibre (NDF) in the ration. We measured the concentrations of histamine, lactate and other volatile fatty acids (VFA), lipopolysaccharide (LPS) in ruminal fluid, LPS and VFAs in caecal fluid, and we took samples of the ventral blind sac of the rumen for histopathology, immunohistopathology and gene expression. Subjective assessments were also made of the presence of lesions on the reticuloruminal wall and the colour of the lining of the reticuloruminal wall.

**Results** The farm of origin was the dominant effect on the measured variables, almost all variables differing significantly among farms. The animals on the high risk diets had lower concentrations of short-chain fatty acids (SCFAs) and higher concentrations of lactate and LPS in the reticuloruminal fluid. The diameters of the strata granulosum, corneum and of the vasculature of the papillae were increased on the high-risk farms, as was the expression of the gene *TLR4* in the ruminal epithelium, while the expression of *IFN-γ* and *IL-1β* was lower, as were the counts of CD3+ and MHCII+ cells. As the proportion of barley in the ration increased, so did the concentration of reticuloruminal and caecal lactate, reticuloruminal histamine, LPS and the thickness of the strata corneum and granulosum, while there was a reduction in the total SCFA concentration in the reticulorumen the expression of *IFN-γ* and count of MHCII+ cells in the ruminal epithelium. Increasing the proportion of fines in the ration had mostly similar effects to increasing the proportion of barley, but in several cases, the effect was stronger and there was also an increase in the expression of *NHE3* gene in ruminal epithelium. *NHE3* expression was positively associated with the concentration of LPS in reticuloruminal fluid ($r = 0.30$, $p < 0.05$), and with the level of fines in the ration. *IFN-γ* expression decreased as barley and fines in the ration increased. The levels of expression of the genes in the epithelium correlated moderately or highly with each other. They were not strongly correlated with the thickness of the stratum granulosum, nor with the density of CD3+ nor MHCII+ cells, nor lactate concentration in reticuloruminal or caecal fluid. Of the dependent variables measured in this study, LPS and lactate concentrations are likely the best indicators of pathology, however without any independent marker for performance or health it was not possible to determine de novo thresholds at which LPS or lactate concentrations indicate pathology. A previously proposed threshold of 5 mmol/l of lactate above which an animal is considered to be suffering from acute acidosis seems reasonable and would have been exceeded by 4% of the animals in this study.

**Conclusion** Farm to farm variation in rations and practices was wide and the effects of individual farm factors on animal physiology dominated any other factors that were measured. The proportion of fines in concentrate-based total-mixed rations is a strong driver of the reticuloruminal SCFA, lactate and histamine concentrations. It follows that as much care is required with presentation of the concentrate diet as with its formulation.

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**049**

# The effect of rumen fluid to buffer ratio on *in vitro* fermentation and the stability of the rumen bacterial community composition in cattle

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**Application** Maintaining a bacterial community which closely resembles that of the host animal in *in vitro* models of rumen fermentation is important to ensure meaningful results are obtained where the aim is to manipulate this community.

**Introduction** *In vitro* models of the rumen are commonly used to study the effect of feed, treatments and manipulations on fermentation parameters and more recently the microbial composition. Currently, there is no general consensus on the amount of rumen fluid to add to the model (Yáñez-Ruiz *et al* 2016). Previous work in our lab has shown a rapid decline in alpha diversity with a 1:9 ratio of rumen fluid to buffer. Using *in vitro* rumen models to study treatment effects on the microbiota requires the microbiota to remain as similar to the host animal as possible throughout the fermentation. The aim of this work was to determine the effect of rumen fluid to buffer ratio on fermentation parameters and the stability of the bacterial population over time in an *in vitro* batch culture model of the rumen.

**Material and methods** Rumen fluid was collected from a Charolais cross steer at time of slaughter, raised on the North Wyke Farm Platform (NWFP, Devon, UK) on permanent pasture. Rumen fluid was used to inoculate the *in vitro* model in three different ratios 1:2, 1:4 and 1:9 with Mould's buffer (Mould *et al.* 2005) to a total volume of 50 ml. Fresh grass collected from the animal's permanent pasture immediately prior to slaughter was used as the substrate (0.5 g DM). The *in vitro* fermentation was run for 48 hours with sampling at 0, 6, 12, 18, 24, 36 and 48 hours. Bottles containing only grass and buffer were included to measure fermentation by the grass-associated bacteria and no-substrate bottles were included as blanks. Each treatment at each time point was tested in triplicate (n=15 per time point). *In vitro* dry matter digestibility (IVDMD), gas volume produced (GV, ml) and volatile fatty acid production (VFA, mM) were measured and microbial pellets were collected for bacterial community analysis by 16S rRNA sequencing of the V1-V3 region on the MiSeq V3 platform (Illumina). All measures were standardised per gram DM added and blank corrected where appropriate. General linear models with ratio and time as factors were performed with Tukey's *post-hoc* test (IBM SPSS Statistics 21). Sequencing reads were processed in the software Mothur with PERMANOVA and alpha diversity measures (Chao1, Shannon, Simpson) performed in R.

**Results** IVDMD showed a significant interaction between ratio and time ($P < 0.001$; Table 1). The 1:2 and 1:4 ratios differed prior to 18 hours only. No difference was seen between the three ratios containing rumen fluid from 36 hours onwards. At 48 hours, bottles with rumen fluid added showed no difference to the grass + buffer controls. Gas volume again showed a significant interaction between ratio and time ($P < 0.001$) with the most concentrated ratio (1:2) giving the greatest total gas volume produced compared to the 1:4 and 1:9 ratios (89.7 *vs* 83.1 *vs* 71.5 ml respectively). An interaction was also seen for total VFA production ($P < 0.001$). The 1:9 dilution showed the highest total VFA production at both 36 hours (38.2 *vs* 36.6 *vs* 50.8 mM; as above) and 48 hours (33.3 *vs* 48.3 *vs* 61.9 mM; as above) whilst 1:2 showed higher VFA production at 6 hours. Analysis of the sequencing reads is underway.

**Table 1** The effect of rumen fluid to buffer ratio on IVDMD

| Time (hours) | | | | | | | | p value | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | 6 | 12 | 18 | 24 | 36 | 48 | SEM | Time | Ratio | Time*Ratio |
| 1:2 | 0.5832[a] | 0.6153[a] | 0.6383[a] | 0.6471[a] | 0.6757[a] | 0.6913 | 0.012 | < 0.001 | < 0.001 | < 0.001 |
| 1:4 | 0.4937[b] | 0.5655[b] | 0.6333[a] | 0.6364[a] | 0.6900[a] | 0.6891 | | | | |
| 1:9 | 0.4582[b] | 0.4813[c] | 0.5409[b] | 0.5865[b] | 0.6371[a] | 0.6215 | | | | |
| Grass | 0.3543[c] | 0.3521[d] | 0.3524[c] | 0.3529[c] | 0.4749[b] | 0.6228 | | | | |

[a,b,c,] different subscript denotes significant differences within a column

**Conclusion** The use of the highest ratio (1:2) is not required for *in vitro* fermentation of fresh grass, 1:4 would suffice for fermentations longer than 12 hours resulting in a lower requirement of rumen fluid. Fermentations should not run for longer than 36 hours, as at 48 hours, DM digestibility in bottles containing only grass and buffer was the same as those containing rumen fluid. Sequencing results will determine which ratio, if any, ensures the bacterial community remains stable and similar to that of the original rumen fluid inoculum throughout the fermentation to study rumen microbiota using the model.

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**References**

Mould, F. L., Morgan, R., Kliem, K. E. and Krystallidou, E. 2005. Animal Feed Science and Technology 123, 155–172
Yáñez-Ruiz, D. R., Bannink, A., Dijkstra, J., Morgavi, D. P., O'Kiely, P., Reynolds, C. K., Shwarm, A., Shingfield, K. J., Yu, Z. and Hristov, A. N. 2016. Animal Feed and Science Technology 216, 1-18.

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**050**

# Determination of the bioavailability of trace minerals supplied as nanoparticles in sheep and their effect on performance and health

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**Application** Increased bioavailability of nano copper oxide (CuO) compared to conventional CuO could reduce the dose required to maintain the copper (Cu) status of ruminants and reduce excess Cu excretion into the environment.

**Introduction** In ruminants the apparent absorption of Cu is low, with between 0.02 and 0.07 of Cu from sources such as CuO being absorbed (Parkins *et al*., 1994), resulting in high levels of excretion into the environment. Similarly, the absorption of Zn is low at approximately 0.25, with the fractional rate of absorption increasing at lower dietary Zn concentrations (Suttle, 2010). Improving the bioavailability of trace minerals could reduce the dose required and subsequently decrease the amount excreted into the environment (Swain *et al*., 2016). It is has been shown that supplying trace minerals as nanoparticles improves their bioavailability compared to conventional sources, mainly because of their small particle size and functional coating. For example Rajendran *et al*., (2013) reported a significant increase in plasma Zn concentrations in cows receiving nano-Zn compared to ZnO with an associated improvement in milk yield. However, there is a general lack of information on the bioavailability of nanoparticle trace elements on the minerals status, performance and health in ruminants. The objectives of the study were to establish the bioavailability of nanoparticle trace elements compared to conventional sources in growing lambs, and to determine their effect on performance and health.

**Material and methods** Fifty, six month old, castrated Welsh Mountain lambs were used. For blood metabolites the coefficient of variation is approximately 11% therefore 10 replicates/treatment were required to detect a 10% difference. Lambs were gradually introduced to a pelleted diet over a period of 3 weeks. Lambs were stratified and blocked according to liveweight and allocated to one of four treatments: **O**; 8 mg Cu/kg DM as CuO and 40 mg Zn/kg DM as ZnO, **NH;** 8 mg Cu/kg DM as nano-CuO and 40 mg Zn/kg DM as nano-ZnO, **NL**; 4 mg Cu/kg DM as nano-Cu and 20 mg Zn/kg DM as nano-ZnO or **S**; 8 mg Cu/kg DM as $CuSO_4$ and 40 mg Zn/kg DM $ZnSO_4$, or an initial slaughter group. Lambs were housed in individual, sawdust bedded pens and fed at 90 - 95% of *ad libitum* intake. Feed refusals were collected daily at 7.30 h. Water was continuously available. Blood samples were collected weekly at approximately 11.00 h by jugular venepuncture and stored at -20°C for subsequent analysis by ICP-MS, and every three weeks for determination of ceruloplasmin (Cp), y-glutamyl transferase (GGT), superoxide dismutase (SOD) and alkaline phosphatase (ALP) using a Cobas Mira Plus Autoanalyser (ABX Diagnostics, Bedfordshire, UK). All sheep were slaughtered at a commercial abattoir either at the beginning of the study (initial slaughter group n=10) or at the end of the 9$^{th}$ study week (n=10 per treatment). Liver and kidneys were collected at slaughter for subsequent analysis. Data was analysed by repeated measures analysis of variance using Genstat 17 as a randomised block design using data recorded in week 0 as a covariate where appropriate.

**Results** There was no effect of form or level of Cu and Zn supplementation on feed refusals or live weight gain (P>0.05), with a mean value of 90 g/d. Plasma activity of GGT was high (55-61 U/l) for all lambs at the start of the study and declined through the study except for those supplemented with $CuSO_4$, which remained high. Hepatic Cu concentration was increased (P<0.05) in lambs supplemented with $CuSO_4$ compared to CuO (Figure 1), and there was a trend (P=0.11) for lambs supplemented with nano-Cu to have a higher hepatic Cu concentration than lambs supplemented with CuO. Plasma ALP activity increased (P<0.001) for all lambs from week 0 to week 9 (mean values 157 U/l and 401 U/l respectively) but there was no effect (P >0.05) of form or level of Cu and Zn supplementation.

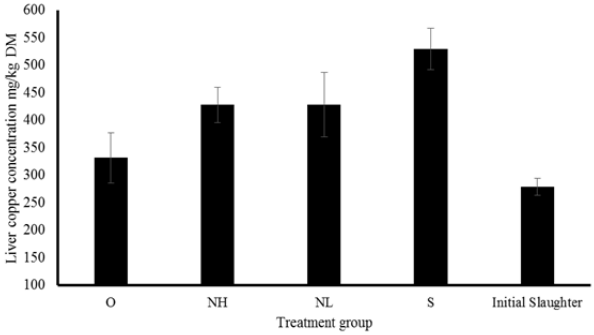


**Figure 1** Effect of form and level of Cu and Zn supplement on liver copper concentration (mg/kg DM) in growing lambs. Error bars indicate the SEM

**Conclusion** Nano CuO may have a greater bioavailability than conventional CuO, but variability between individual lambs was higher than expected therefore a greater number of replicates per treatment may be required to reach full statistical significance. Suppling trace mineral as nanoparticles did not have any effect on performance or health.

**References**

Parkins, J. J., Hemingway, R. G., Lawson, D. C., Ritchie, N. S. 1994. British Veterinary Journal 150, 547-553.
Rajendran, D., Kumar, G., Ramakrishnan, S., Thomas, K. S 2013. Research Journal of Biotechnology 8, 11-17
Suttle, N.F. 2010. The mineral nutrition of livestock (4$^{th}$ edition). CABI publishing
Swain, P. S., Rao, S. B., Rajendran, D., Dominic, G., Selvaraju, S. 2016. Animal Nutrition 2, 134-141.

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**051**

# A preliminary evaluation of the effects of supplementing ewes with Co, including method of administration, on ewe performance and that of their progeny

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**Application** Supplementing ewes with Co from 7 weeks pre joining until mid-pregnancy has no effect on their performance or that of their progeny where pasture meets the cobalt requirement.

**Introduction** The number of lambs reared per ewe joined is a key determinant of profitability on sheep farms. Mean weaning rate on Irish sheep farms is 1.3 lambs per ewe joined (National Farm Survey) and has been relatively constant for the last 30 years. Cobalt deficiency has been identified as a major issue in some sheep flocks in Ireland. A previous study at Athenry reported increased performance of lambs post weaning due to Co supplementation. (Keady *et al*.,2017). Reproductive performance may be restricted when an animal is deficient in a particular mineral (NRC, 2007). As there is a paucity of experimental data, the aim of the current study was to evaluate the effects of supplementation with Co, and method of administration, from pre joining to mid-pregnancy on ewe performance and the performance of their progeny.

**Material and methods** Three hundred and fifty ewes (Belclare, Belclare×Suffolk and >75% Suffolk) (mean initial live weight 74.4kg [SD 9.71]) were allocated at random within genotype to 1 of 3 treatments from 7 weeks pre joining to 6 weeks prior to lambing. The 3 treatments were as follows: no supplementation (control), Co via drench (drench) or Co via bolus (bolus). The concentration of Co in the drench was 2.1 mg/ml as cobalt sulphate heptahydrate ($CoSO_4$ $(H20)_7$); 15 ml were administered to the ewes at 14-day intervals equivalent to 2.25mg/day. The bolus also contained $CoSO_4$ $(H20)_7$ with an expected release rate of 0.6 mg/d and efficacy of 5 to 6 months, and was administered at the start of the study. The ewes were managed as one group in a rotational grazing system until housing in December. Ewes were synchronised using progesterone impregnated sponges and joined with Charollais rams. At housing, ewes were shorn and offered grass silage *ad libitum*. Concentrate (containing standard mineral and vitamin levels) was offered in late pregnancy, the level offered depending on scanned litter size. Post lambing, ewes rearing singles or twins, and their lambs, were turned out to pasture, without concentrate supplementation, and were managed in a rotational-grazing system. Ewes rearing triplets received 0.5 kg concentrate daily for the first 5 weeks post-lambing while their lambs were supplemented with concentrate (up to max of 300 g daily) until weaning (14 weeks of age). Samples of herbage were taken from each paddock pre-grazing and a representative sample of silage offered was collected once weekly for mineral analysis. The data were analysed as a randomised study using Proc MIXED of SAS to fit a model with fixed effects for treatment, breed and ewe age and ewe as a random term for lamb growth traits.

**Results** The mean Co concentration of the herbage from 7 weeks pre joining to housing was 0.10 mg/kg DM. The effects of supplementation with Co and method of Co administration on ewe and lamb performance are presented in Table 1. Neither Co supplementation or method of administration had an effect (P>0.05) on litter size, number of lambs reared/ewe joined, ewe BW, ewe BCS, lamb weight at birth or weaning, or on lamb mortality

**Table 1** The effects of cobalt supplementation and method of administration on ewe and lamb performance

| | | Treatment | | | | Contrast | |
|---|---|---|---|---|---|---|---|
| | | Control (C) | Drench (D) | Bolus (B) | s.e | D v B | C v D+B |
| Ewe BW (kg) | - mid pregnancy | 74.7 | 75.2 | 76.1 | 0.47 | NS | NS |
| | - lambing | 75.3 | 76.5 | 76.5 | 0.55 | NS | NS |
| BCS | - mid pregnancy | 3.3 | 3.4 | 3.4 | 0.04 | NS | NS |
| | - lambing | 3.3 | 3.3 | 3.4 | 0.05 | NS | NS |
| Litter size | | 2.02 | 1.94 | 2.00 | 0.068 | NS | NS |
| Lambs reared/ewe joined | | 1.79 | 1.67 | 1.64 | 0.080 | NS | NS |
| Lamb BW (kg) | - birth | 5.1 | 5.1 | 5.1 | 0.08 | NS | NS |
| | - weaning | 33.0 | 32.5 | 32.7 | 1.23 | NS | NS |
| Lamb mortality (%) | | 10.1 | 11.6 | 14.6 | 0.25 | NS | NS |

**Conclusion** Supplementation with Co, either via drench or bolus, had no effect on ewe reproductive performance or the performance of progeny up to weaning.

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**References**

Keady, T. W. J., Hanrahan, J. P., Fagan, S. P. 2017. Animal Science 95, 379-386
National Research Council, Nutrient requirements of small ruminants, 2007.

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052

# Effect of source and level of vitamin E supplementation on ewe and lamb performance during late pregnancy and early lactation

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**Application** Supra-nutritional levels of vitamin E supplementation may increase lamb birth and weaning weight by increasing placental size and efficiency.

**Introduction** Lamb mortality is over 15% in the UK, which represents a major loss to the sheep production sector. The addition of high level of vitamin E to ewe's diets in late pregnancy has been shown to improve lamb birth weight and neonatal lamb vigour (Capper *et al.,* 2005). However, these responses are inconsistent (Rooke, *et al*., 2009). In the UK, concentrate feed for pregnant and lactating ewes typically contains 100-150 mg/kg vitamin E, which is lower than the (NRC, 2007) requirements. Responses in terms of lamb birth weight and survival have mainly been reported using concentrates containing >500 mg/kg vitamin E (Merrell, 1998). Work with growing lambs suggests that naturally occurring vitamin E in forages may be more readily available that synthetic α tocopherol acetate added to concentrates. (Kasapidou, *et al.,* 2009). The objective of the current experiment was to investigate the effect of source and level of vitamin supplementation on ewe and lamb performance during late pregnancy and early lactation.

**Materials and methods** At day 103 of pregnancy (week -6) 44 twin bearing Suffolk x Mule ewes with a mean live weight (LW) of 77 kg and body condition score (BCS) of 2.9 were allocated by parity, LW and BCS to either a grass silage (GS) or straw (S) based diet supplemented with either a low (L, 2.8 mg/kg LW) or high (H, 9.0 mg/kg DM) vitamin E concentrate to provide 4 treatments; GSL, GSH, SL and SH. Forages were offered *ad-libitum* with concentrates being fed to provide a rising plane of nutrition to meet the metabolisable energy (ME) and metabolisable protein (MP) requirements of twin bearing ewes during late pregnancy and producing 3.0 litres of milk during early lactation (AFRC, 1993). Ewe LW, CS were recorded weekly, with litter weight being recorded 12 h post-lambing. Lamb weights were recorded weekly. Following parturition, the placenta was collected, washed and weighed, and the number of placentomes recorded as described by (Dwyer, *et al.,* 2005). Ewes were group housed between weeks +4 and +8 *post-lambing* and the performance was monitored within each treatment. The data were analysed by ANOVA as a 2 x 2 factorial design using Genstat17.

**Results** *Pre-partum*, ewes offered GS had a higher LW gain, and tended to have a lower BCS loss than those offered S. In addition, they had a higher placental weight. However, forage source had no effect on litter weight. *Post-partum*, lambs from ewes offered S were heavier at week +8. Ewes offered high vitamin E (H) had a higher placental weight, cotyledon weight, litter weight and were heavier at week +8 than those offered low vitamin E (L).

**Table 1** Effect of forage type and vitamin E supplementation on ewe and lamb performance

| | | | | | SED | Probability | | |
|---|---|---|---|---|---|---|---|---|
| | SL | SH | GSL | GSH | | Forage | Vit. E | Int. |
| *Pre-p* Forage DMI (kg) | 0.68 | 0.67 | 1.18 | 1.17 | 0.080 | <.001 | 0.859 | 0.971 |
| *Pre-p.* LW change (kg) | 9.64 | 11.14 | 13.43 | 12.73 | 1.245 | 0.005 | 0.654 | 0.221 |
| *Pre-p.* CS change | 0.05 | -0.16 | 0.13 | 0.11 | 0.131 | 0.066 | 0.225 | 0.344 |
| *Post-p* LW change (kg) | -6.48 | -5.67 | -3.78 | -7.14 | 2.110 | 0.684 | 0.403 | 0.178 |
| *Post-p* CS change | -0.26 | 0.01 | 0.01 | -0.23 | 0.157 | 0.960 | 0.797 | 0.029 |
| Placenta weight (kg) | $0.70^{a}$ | $0.91^{b}$ | $0.90^{b}$ | $0.90^{b}$ | 0.063 | 0.041 | 0.027 | 0.022 |
| Cotyledons weight (kg) | $0.21^{a}$ | $0.30^{b}$ | $0.23^{a}$ | $0.22^{a}$ | 0.024 | 0.190 | 0.027 | 0.007 |
| Litter birth Weight (kg) | 9.74 | 10.99 | 9.81 | 9.90 | 0.473 | 0.135 | 0.056 | 0.095 |
| Week +8 litter Weight (kg) | 42.31 | 43.61 | 38.30 | 40.77 | 1.09 | <.001 | 0.023 | 0.455 |

**Conclusion** Forage effects on ewe performance probably reflect differences in energy and/or energy substrate supply. Supra-nutritional levels of vitamin E supplementation increased litter birth weight and week +8 weight by increasing placental size and efficiency.

**References**

AFRC. 1993. Energy and protein requirements of ruminants. Wallingford, CAB Publishing.
Capper *et al*. 2005. British Journal of Nutrition 93(04), 549-557.
Dwyer et.al. 2005. Theriogenology 63, 1090-1110.
Kasapidou *et al*. 2009. Animal 3, 516-526.
Merrell, B. 1998. Sheep Veterinary Society Proceedings 22, 57-61.
NRC. 2007. Nutrient Requirements of Small Ruminants. The National Academic Press, Washington, D.C.
Rooke *et al*. 2009. Animal 3 (11), 1555-1561.

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053

# Comparison of pre-lambing metabolic profiles of Scottish Blackface and Lleyn twin-bearing ewes farmed together in a Scottish hill environment

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**Application** Pre-lambing metabolic profiles of Lleyn and Scottish Blackface (BF) twin-bearing ewes suggested that Lleyn ewes are as capable of producing twins in a hill environment as BF flockmates, although they probably consume more feed.

**Introduction** Introducing a prolific lowland/upland sheep breed (e.g. Lleyn) into a hill environment, or long-term genetic selection of an established hill sheep breed, e.g. Scottish Blackface, are contrasting options to improve the reproductive capability and productivity of hill sheep enterprises (Zhou *et al*, 2017). However, grazing provides inadequate nutritional supply in hill sheep farming systems (Robinson *et al*, 2002), especially during winter and early spring. Consequently, twin-bearing and multiple- bearing ewes might encounter metabolic disorders, such as twin lamb disease, if energy and protein requirements cannot be met through diet and a safe level of maternal body tissue mobilisation. Pre-lambing metabolic profiling is a recommended method for determining the nutritional status of late-pregnancy ewes (Russel, 1984), as it examines plasma concentrations of β-hydroxybutyrate (BOHB), albumin, urea nitrogen (N), copper (Cu) and magnesium (Mg). The aim of this study was to investigate pre-lambing metabolic status of unimproved Scottish Blackface (UBF), genetically improved Scottish Blackface (IBF; Conington *et al*, 2006) and Lleyn twin-bearing ewes farmed together.

**Material and methods** In this experiment, conducted at SRUC's Hill & Mountain Research Centre (56°N, 4°W), the three genotypes of ewes were managed together in either a predominantly "Hill Grazing" or a "Park Grazing" system, each with different criteria for using grazing resources and feed supplements. The ewes were mated with rams of their own genotype category, and ultrasound-scanned in mid-February. Two supplementary winter feeding levels, high or low, were applied in two periods (1st = early January to scanning; 2nd = scanning to lambing in April). Initial allocation to feeding level was on the basis of actual liveweight change versus a target liveweight change which took account of pre-mating liveweight and body condition score (CS); after scanning, the foetuses carried was also considered. Blood samples were taken from twenty healthy 3- to 5-year-old twin-bearing ewes (based on scanning results) of each genotype on 21st March 2017, to assess pre-lambing metabolic status, using the Dairy Herd Health and Productivity Service. Only data from ewes that gave birth to twins and lambed 25 to 46 days post-sampling (UBF = 19, IBF = 18, Lleyn = 17) were analysed statistically, using a Generalized Linear Model (GenStat 16; VSNi). Terms used in the Maximal Model for each metabolite were genotype, age, CS, 2nd feeding level, system, days from sampling to lambing, and litter weight. Terms used in the final model were based on suggestions from stepwise regressions.

**Results** For all 3 genotypes, assayed metabolites were within recommended ovine reference ranges (Table 1). BOHB and Cu concentrations did not differ among the 3 genotypes. Lleyn twin-bearing ewes had lower albumin concentrations, but higher urea N concentrations than their counterpart UBF and IBF ewes. Mg concentrations differed among the 3 genotypes.

**Table 1** Metabolite concentration (least squares mean ± standard error) values for twin-bearing ewes of 3 genotypes

| Plasma metabolite | Reference range | UBF | IBF | Lleyn | P value |
|---|---|---|---|---|---|
| BOHB (mmol/l) | <1.0 | $0.83 \pm 0.07^{a}$ | $0.78 \pm 0.06^{a}$ | $0.64 \pm 0.07^{a}$ | 0.17 |
| Albumin (g/l) | 25-35 | $32.46 \pm 0.51^{a}$ | $31.93 \pm 0.45^{a}$ | $29.86 \pm 0.50^{b}$ | 0.003 |
| Urea N (mmol/l) | 3-8 | $3.03 \pm 0.14^{a}$ | $3.06 \pm 0.14^{a}$ | $4.05 \pm 0.14^{b}$ | <0.001 |
| Cu (µmol/l) | 9.4-18.8 | $13.45 \pm 0.66^{a}$ | $13.50 \pm 0.59^{a}$ | $13.46 \pm 0.64^{a}$ | 0.998 |
| Mg (mmol/l) | 0.7-1.3 | $0.89 \pm 0.02^{a}$ | $0.96 \pm 0.02^{b}$ | $1.06 \pm 0.02^{c}$ | <0.001 |

**Conclusion** Twin-bearing Lleyn ewes were as competent as BF flockmates in coping with foetal nutrient demands in this hill environment. Lleyn ewes had sufficient energy (BOHB concentration) and protein (albumin and urea N concentrations) supply in late pregnancy and they probably consumed more feed than BF counterparts, as suggested by their higher Mg concentrations. These results are consistent with the findings from springtime 2015 for the same flock (Zhou *et al*, 2016).

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**References**

Conington, J., Bishop, S. C., Lambe, N. R., Bünger, L. and Simm, G. 2006. Animal Science 82, 445-453.
Robinson, J.J., Rooke, J.A. and McEvoy, T.G. 2002. In: M Freer, H Dove (Eds.). Sheep Nutrition Ch.9, 189-211.
Russel, A. J. F. 1984. Livestock Production Science 11, 429-436.
Zhou, P., Lambe, N., Morgan-Davies, C., Sargison, N. and McEvoy, T. 2016. Book of Abstracts of the 67th Annual Meeting of the European Federation of Animal Science, Belfast UK, 29 Aug – 2 Sept 2016. No. 22. 38, 368.
Zhou, P., Sargison, N., Lambe, N., Morgan-Davies, C. and McEvoy, T. 2017. The 9th ISVC, Harrogate (Abstract, in press).

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**054**

# Seasonality of copper and its antagonist elements in grazing pasture

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**Application** Fluctuations in pasture copper and its antagonists can used as a predictor for copper status in grazed sheep. In conjunction with housing patterns, routine pasture analysis can be used to find a targeted approach to trace mineral supplementation strategy.

**Introduction** The presence of antagonists, dietary elements which bind to copper inhibiting its availability; specifically, sulphur in combination with either iron or molybdenum in pasture can contribute to a low copper status and can induce the clinical signs associated with copper deficiency (Gould & Kendall, 2011). Extensively grazed sheep rely primarily on pasture to meet their mineral requirements. Pasture analysis of mineral content is not routine and supplementation for copper is often non-targeted and can lead to imbalance or accumulation. Previous work has shown that finishing lambs are more likely to be of marginal-low copper than high status (Clarkson *et al*., 2017). Low copper status can cause a range of clinical signs including reduced fertility, loss of wool condition, depressed immune function and neonatal ataxia 'swayback'. The aim of this study was to monitor grazed pasture across two grazing seasons to establish seasonal patterns to predict supplementation requirements.

**Material and methods** Grass samples (*n*=240) from 13, predominantly perennial ryegrass, fields on a Leicestershire farm were collected by pinch sampling to mimic grazing each month sheep were at pasture during 2015 and 2016. Samples were freeze dried, ground and mixed prior to microwave wet-acid digestion and elemental analysis using ICP-ms. At monthly intervals, where routine handling allowed, 4 sheep from each of the fields being grazed were sampled through jugular venepuncture into lithium heparin and clot activator vacuum tubes for blood analysis of plasma Cu, caeruloplasmin and superoxide dismutase activity. Sampled sheep received no mineral supplementation during the monitoring period. Microsoft Excel (v15.0) was used to calculate means and standard error and plot graphed data.

**Results** Pasture copper was found to be adequate both years; decreasing during summer and rising late autumn. Iron was lowest during the summer in both seasons with a marked increase during autumn-winter. Molybdenum was consistent year to year; highest during spring. Sulphur rose around May and declined in the autumn. Sheep were found to be of low blood copper status throughout the summer and autumn from the majority of fields, despite adequate copper concentration in the pasture.

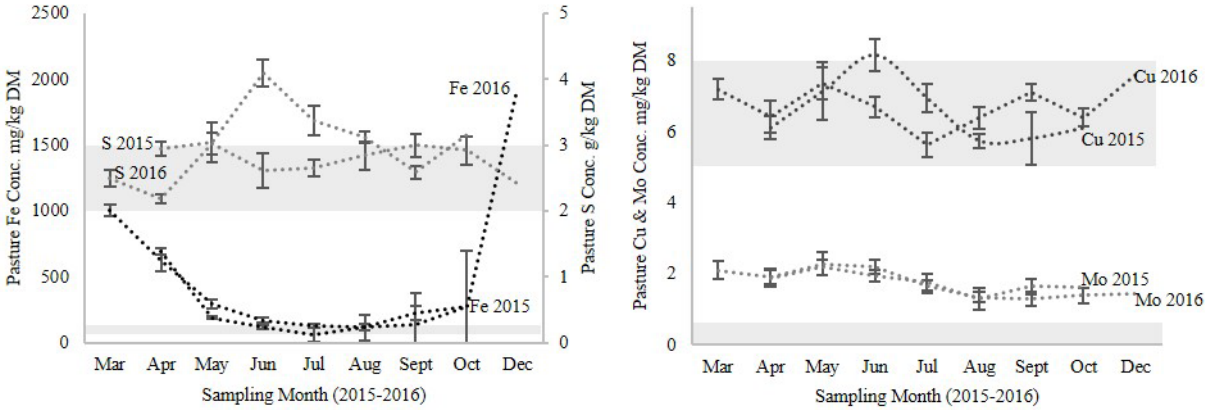


**Figure 1** Mean pasture concentration of copper, iron, molybdenum and sulphur across 2015-16. Bars denote standard error of the mean, grey areas show requirement ranges for sheep (S 2-3 g/kg DM, Cu 5-8 mg/kg DM, Fe 30-50 mg/kg DM, Mo no defined requirement <0.5 mg/kg DM).

**Conclusion** Consistent molybdenum >0.5 mg/kg DM, alongside rising sulphur may contribute to low blood copper during the summer. Through the autumn, blood copper status remains lowered as a result of increasing pasture iron. Intra-ruminal interactions between elements give greater effects than the pasture copper composition, resulting in decreased copper status and increasing the risk of copper responsive problems. To counteract this, copper supplementation was suggested from summer until winter housing. Sheep were set out to pasture in the spring after iron concentrations began to decline.

**References**

Clarkson, A.H., Meades, N., Watters, B. and Kendall, N.R. 2017. In: 9th ISVC. Harrogate, UK, 48.
Gould, L. and Kendall, N.R. 2011. Nutrition Research Reviews 24, 176–182.

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**055**

# Effect of breed and iron supplementation on plasma copper status of growing lambs

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**Application** Breed has an important effect on copper metabolism of sheep and high dietary iron levels reduce the copper status of sheep.

**Introduction** Copper (Cu) is an important trace element required in the structure of over 300 proteins. Copper deficiency impairs the function of Cu containing enzymes, and subsequently affects vital physiological processes in the body (Suttle, 2010). Copper deficiency is common in sheep grazing pastures low in Cu or mainly high in iron (Fe) or molybdenum (Mo) and sulphur (S). Where existence of one or two of these three elements (Fe, Mo, and S) together in a high concentration in sheep diet will inhibit Cu absorption or metabolism. Breed variation can also affect Cu metabolism and may contribute to this issue (Fry *et al*., 2013). Therefore an experiment was conducted to investigate the effects of breed and Fe supplementation on Cu metabolism and performance of Scottish Blackfaces and Texel lambs.

**Material and methods** Thirty-six wethers (18 Texel crossbred (T) and 18 Scottish Blackface (SB)); with live-weight (LW) of 25.6 ± 1.99 kg, were used in a 2 x 2 factorial design experiment. Lambs were blocked by LW and breed and then randomly assigned to one of two dietary treatments (9 lambs per treatment). Dietary treatments were: 1) SB given no Fe supplemental diet (SB-), 2) SB given Fe supplemented diet (SB+), 3) T given no Fe supplemental diet (T-), and 4) T given Fe supplemented diet (T+). Low Fe diets were not supplemented with Fe (basal Fe and Cu was 258 and 8.3 mg/kg DM, respectively) and high Fe diets were supplemented with 800 mg Fe/kg DM (total Fe was 1151 mg/kg DM). Lambs were housed individually throughout the trial period, and fed a diet based on nutritionally improved straw and barley. The formulated basal diet was iso-nitrogenic and iso-energetic fed at restricted level to gain 200 g/day (AFRC, 1993) over 6 weeks period. Live weight was recorded weekly and jugular blood samples were taken fortnightly to monitor plasma Cu levels. Plasma samples collected were analysed for Cu and Fe by ICP-MS. The data was analysed by ANOVA as a 2 × 2 factorial design using GenStat 17$^{th}$ edition.

**Results** There was no effect of diet x breed interaction on LW, daily gain, or feed conversion ratio of lambs. Supplemental Fe or breed of sheep had no effects ($P$>0.05) on performance attributes. Repeated measure analysis showed a significant time × Fe interaction and time × Fe interaction on plasma Cu concentration ($P$<0.05), but there was no Fe × breed interaction or time × Fe × breed interaction on plasma Cu concentration. Due to differences in the plasma Cu concentration between the breeds, week 0 was used as a covariate for plasma mineral ANOVA. There was no Fe × breed interaction on plasma Cu concentration at any weekly time point throughout the study (Table 1). The lambs that received supplemental Fe diet had a lower plasma Cu concentration from week 2 to 6 compared with those receiving no supplemental Fe. Scottish Blackface lambs had a higher plasma Cu compared with T at week 2, and a trend at week 4 and 6. There was no time × Fe, time × breed or time × Fe × breed interaction on plasma Fe concentration. There was also no Fe × breed interaction on plasma Fe concentration at any weekly time points throughout the study. Plasma Fe concentration was higher in the lambs given Fe supplements at week 2, 4, and 6 ($P$<0.05) compared with those given no Fe. Texel lambs had a lower plasma Fe concentrations ($P$<0.05) compared with SB at week 4 but not in the following week.

**Table 1** Effect of iron supplementation and breed on plasma copper concentration of growing lambs (µmol/l)

| Week | Treatments | | | | s.e.d. | P-value | | |
|---|---|---|---|---|---|---|---|---|
| | SB- | SB+ | T- | T+ | | Fe | Breed | Fe x Breed |
| 0 | 17.11 | 17.30 | 14.93 | 15.49 | 1.105 | - | - | - |
| 2 | 17.11 | 15.37 | 15.57 | 13.89 | 0.958 | 0.01 | 0.045 | 0.96 |
| 4 | 14.59 | 12.32 | 12.86 | 11.65 | 0.840 | 0.004 | 0.07 | 0.36 |
| 6 | 16.25 | 13.50 | 14.07 | 13.07 | 0.838 | 0.002 | 0.05 | 0.13 |

SB-: Scottish Blackface given no Fe supplemental diet, SB+: Scottish Blackface given Fe supplemental diet. T-: Texel given no Fe supplemental diet, T+: Texel given Fe supplemental diet.

**Conclusion** High dietary Fe significantly reduces plasma Cu concentrations. Scottish Blackface had numerically higher plasma Cu concentration than Texels throughout the study.

**Acknowledgements** Kurdistan Regional Government is gratefully acknowledged for funding this study.

**References**

AFRC. 1993. Energy and Protein Requirements of Ruminants. CAB Publishing, Wallingford.
Fry, R., Spears, J., Hansen, S., Liu, H. and Ashwell, M. 2010. Journal of Animal Science 93, 499.
Suttle, N. 2010. Mineral Nutrition of Livestock. 4th Edition. CABI, Wallingford, Oxon, UK.

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# A survey of mineral supplementation and delivery strategies on Irish sheep farms

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**Application** Whilst 69% of Irish sheep producers supplement their flocks with minerals and vitamins, most supplementation decisions are undertaken in the absence of veterinary advice or laboratory results.

**Introduction** Mineral supplementation is routinely practiced on many Irish sheep farms. The aim of this paper is to present results of a survey undertaken to establish (a) the mineral supplementation practices on sheep farms and (b) the knowledge and opinions of farmers in relation to supplementation strategies and deficiencies.

**Material and methods** The survey was undertaken with sheep farmers participating in the Teagasc National Farm Survey (NFS). The NFS is operated as part of the Farm Accountancy Data Network of the EU and is the Irish equivalent of the UK Farm Business Survey. The NFS collects data on farm gross output, costs and income from a statistically representative random sample of approximately 1,000 (dairy, beef, sheep, tillage and mixed) farms each year, representing a farming population of approximately 100,000 farms. A supplementary survey on current on-farm practice, farmer knowledge and opinions in relation to mineral supplementation strategies and deficiencies on sheep farms was conducted in conjunction with the regular 2016 NFS. A total of 184 respondents who farmed sheep completed the additional survey. These are representative of 18,284 Irish sheep farms. The surveys were completed on farm by a team of trained NFS recorders. The survey contained 22 questions. The questions were predominantly "closed-ended", some "open-ended" questions were also included. On completion of data collection the dataset was cleaned and screened for anomalies. Any anomalies were checked and corrected. Descriptive statistics were compiled and preliminary analysis conducted using Microsoft Excel.

**Results** Of the 184 survey responses, 177 (96%) were usable. The mean farm size, sheep forage area, average number of ewes, stocking rate and number of lambs reared per ewe joined was 67 (8-1,117) ha, 22 (1-351) ha, 123 (64-1,298) ewes, 7 (0.2-20) ewes/ha and 1.32 (0.2-2.05), respectively. Fifty six percent of respondents offered concentrates to their sheep as a source of mineral supplementation alone, while 69% supplemented with minerals/vitamins in addition to concentrate feed. Thirty one percent of sheep farmers surveyed did not supplement any additional minerals or vitamins to their sheep.

The main reason for not supplementing (among the 67% who provided a reason) was that no mineral or vitamin deficiency problems have been identified (74%). Twenty three percent of farmers who do not currently supplement with minerals and vitamins supplemented previously. The main reason for discontinuing mineral supplementation was no perceived requirement for mineral supplementation. Only 22% of those who do not currently supplement would consider supplementing in the future, and would do so mainly if additional mineral supplementation was required and flock health issues became a problem. Ten percent of respondents would consider supplementing in the future if better information on requirements and timing of supplementation became available. A lack of performance would also cause 10% of respondents to supplement.

Twenty two percent of those who currently supplement their flock with minerals and vitamins supplement based on results from laboratory analysis (soil, herbage, blood or tissue analysis). Blood (10%) and soil (9%) results are the most common form of laboratory analysis on which the decision to supplement is based. Thirteen percent of those who supplement do so based on veterinary advice, with 30% of those saying this veterinary advice was based on the results of samples sent for laboratory analysis.

Sixty five percent said their decision to supplement was based on a reason other than laboratory analysis or veterinary advice, with only 32% specifying the reason. Of those specifying the reason for supplementation, 51% said their decision to supplement was due to tradition/previous experience. Live weight gain (32%) and fertility (14%) were identified as the main health/production problems associated with mineral/vitamin deficiencies. Generic mineral and vitamin products are the most commonly used, followed by cobalt only products. Mineral buckets are the most common method of supplementing ewes while oral drenching is the most common method of supplementing lambs. Ease of use and labour requirements was ranked by 36% of respondents as the most important factor influencing their choice of supplementation method. Ease of use and labour requirements was also identified by 24% of survey respondents as the second most important factor with cost identified as the third most important factor. Selenium was ranked the most important trace mineral for sheep production.

**Conclusion** Sixty nine percent of Irish sheep farmers supplement their flocks with minerals and vitamins. The main reasons for supplementing are to improve lamb performance and ewe fertility. Drenching is the favoured method of supplementation for lambs while buckets are the preferred method for ewes.

**Acknowledgements** The authors gratefully acknowledge the Teagasc Walsh Fellowship scheme and the help of the NFS recorders.

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# Prime lamb production over 12 years of a rotational-grazing system - effect of birth and rearing type on lamb performance

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**Application** Target growth rates from birth to weaning for lambs born and reared as singles, twins and triplets in a grass-based production system are 330, 270 and 280 g/d, respectively.

**Introduction** In mid-season prime lamb production litter size and standard of grassland management are the main factors affecting efficiency (Keady and Hanrahan 2006). Keady *et al*. (2009) reported lamb carcass output of up to 501 kg/ha for a grass based system involving winter housing, prolific ewes (litter size 2.34; lambs reared per ewe joined 1.86) and access to concentrate by lambs from 8 weeks of age. The aim of the current paper is to present the effect of birth and rearing type on the performance, over a 12-year period, of lambs on a grass-based system up to weaning at 14 weeks of age.

**Material and methods** A total of 7964 lambs (989 singles, 4462 twins, 2151 triplets, 312 quads, 50 quintuplets) were produced in a rotational-grazing system, at Athenry, between 2006 and 2017. Their dams (Hill-cross, Lowland-cross and Belclare; overall litter size 1.98) were on various studies, predominantly examining the effects of nutrition during mid and late gestation. Ewes were joined with rams (Charollais, Suffolk, Texel) in October, for lambing in March, and were shorn and housed in mid-December and offered grass silage. Concentrate was offered during late gestation; level varied according to litter size. Ewes were put to pasture within a few days of lambing. Ewes rearing singles and twins, and their lambs received no concentrate at pasture. Ewes rearing triplets were offered 0.5 kg concentrate daily for 5 weeks post lambing and their lambs had access to concentrate (maximum of 300 g daily) until weaning at 14 weeks old. Sward height, measured in each paddock pre- and post-grazing, was used to manage rotational grazing. Analysis of growth employed Proc MIXED (SAS) to fit a linear model with season, dam and dam (season) as random terms; birth and rearing type, dam age and sex were fixed.

**Results** The effect of birth and rearing type on lamb mortality and performance are presented in Table 1. As birth type increased birth weight declined ($P < 0.001$). The proportion of lambs born dead doubled for each increase in birth type from 2 to 4. Lamb daily gain and weaning weight declined as birth type increased. Relative to those born and reared as twins, lambs born and reared as triplets were lighter at birth ($P < 0.001$) but had a higher daily gain (ADG) to weaning ($P < 0.01$), thus having a similar weight at weaning. The annual estimates of mean weaning weight (twin born reared basis) are presented in Figure 1. The horizontal line (overall mean) shows that the annual mean was within 0.7 kg (2.5%) of the overall mean in 9 of 12 years. Results on sward height are in Figure 2; pre grazing height averaged 5.4, 7.4 and 7.9 cm in April, May and June, respectively; post-grazing height increased from 3.5 cm in April to 4.6 in May and 5.1 cm in June.

**Table 1** Effect of birth and rearing type on lamb mortality and performance

| | | Birth-rearing type (S = single, T = twin, Tr = triplet, Q = Quad or quintuplet) | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | S-S | T-S | T-T | Tr-S | Tr-T | Tr-Tr | - | Q-T |
| Birth weight (kg) | | 5.59$^{a}$ | 4.54$^{b}$ | | 3.71$^{c}$ | | | 3.15$^{d}$ | |
| Mortality (%) | - dead born | 6.6 | 4.0 | | 8.4 | | | 16.3 | |
| | - total | 11.8 | 9.9 | | 19.1 | | | 31.1 | |
| ADG (g/d) | 0-5 weeks | 386$^{a}$ | 338$^{b}$ | 302$^{c}$ | 313$^{c}$ | 286$^{d}$ | 275$^{e}$ | - | 273$^{de}$ |
| | 0-14 weeks | 330$^{a}$ | 301$^{b}$ | 271$^{d}$ | 292$^{b}$ | 266$^{cf}$ | 279$^{e}$ | - | 255$^{df}$ |
| Weaning weight (kg) | | 38.0$^{a}$ | 34.0$^{b}$ | 31.2$^{c}$ | 32.5$^{d}$ | 29.9$^{e}$ | 31.0$^{c}$ | - | 28.5$^{ef}$ |

$^{abcdef}$Means (growth traits), within rows, without a superscript in common are significantly different ($P < 0.05$)

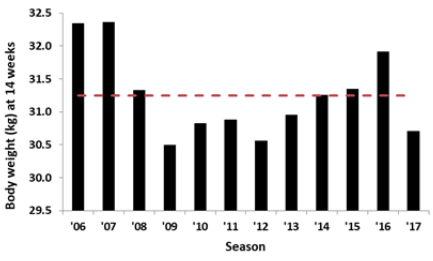


**Figure 1 Weaning weight over 12 seasons**

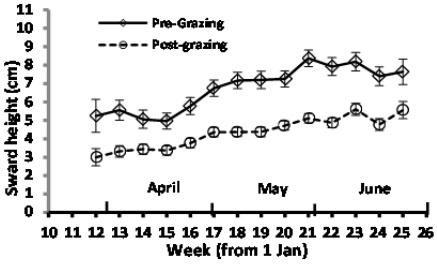


**Figure 2** Mean pre- and post-grazing sward height

**Conclusion** High levels of lamb performance are achievable consistently from rotationally-grazed grass-based systems managed using sward height.

## References

Keady, T.W.J. and Hanrahan, J.P. 2006. Irish Grassland Association Journal 40, 15–27.
Keady, T.W.J., Hanrahan, J.P., Flanagan, S., 2009. Irish Journal of Agriculture and Food Research 48, 87–102.

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# Assessing the effect of stocking rate and prolificacy potential on profitability of grass based sheep production systems using a bio-economic model

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**Application** Variation in stocking rate and prolificacy potential has a significant effect on profit. Increasing the number of lambs weaned per hectare along with increased grass growth and utilization, increased farm net profit.

**Introduction** Stocking rate and ewe prolificacy are key drivers of flock productivity and output across both Irish and international sheep production systems (Keady and Hanrahan, 2006; Ho *et al*., 2014). Stocking rate and ewe prolificacy have a major effect on profit with greater numbers of lambs weaned per hectare resulting in greater profit (Teagasc, 2017). Research to date has assessed the effect of stocking rate and ewe prolificacy on flock performance and lamb output (Earle *et al*., 2016); however, the economic effect was not assessed. The objective of this study was to assess the profitability of Irish grass based sheep production at varying stocking rate and ewe prolificacy levels using a bio-economic model.

**Material and methods** The Teagasc Lamb Production Model (Bohan *et al*., 2016), a bio-economic computer simulation model that simulates sheep production systems, was used to simulate six stocking rate and prolificacy scenarios. The six scenarios simulated included three different stocking rates (10, 12 and 14 ewes/ha) at two different prolificacy levels (1.5 and 1.8 lambs weaned per ewe joined). All input data was obtained from the Sheep Research Demonstration Flock, Athenry, Co Galway, Ireland (Earle *et al*., 2016). Each scenario was simulated on a 20ha farm with a self-replacing March lambing flock. Flock size ranged from 213 to 299 ewes joined to the ram. Grass growth was increased in line with stocking rate and prolificacy ranging from 10,071 kg DM/ha to 14,374 kg DM/ha. Grass utilisation was 80%, 85% and 90% for 10, 12 and 14 ewes/ha, respectively. A final scenario was modelled to investigate the effect of grass growth on stocking rate and prolificacy potential whereby grass growth was maintained at the level achieved by the lowest output system (i.e. 10,071 kg DM/ha) while stocking rate and prolificacy were increased, with the additional energy requirements of the flock being supplied in the form of concentrate supplementation. Risk analysis was conducted using the @Risk programme to assess the effect of variation in key input variables (lamb and ewe mortality, grass growth, fertiliser and concentrate costs, lamb and mutton price) on the profitability of each scenario investigated.

**Results** Results from the bio-economic model showed that the number of lambs weaned per hectare increased as stocking rate and ewe prolificacy increased, and ranged from 16 to 27 lambs/ha. Increasing the number of lambs weaned per hectare reduced the individual lamb growth rate, however, total carcass produced per hectare increased from 272 to 474 kg/ha. Lamb sales increased from €1,299/ha to €2,219/ha, with variable costs rising from €774/ha to €1,224/ha. The average cost of producing a lamb at low prolificacy was €75 but decreased to €65 per lamb at high prolificacy. This translated into an average net profit of €22/lamb and €31/lamb at the low and high prolificacy potentials, respectively. As the number of lambs weaned per hectare increased from 16 lambs/ha to 27 lambs/ha net profit increased from €361/ha to €802/ha. The greatest net profit was achieved when weaning 1.8 lambs per ewe at 14 ewes/ha, with €2,219/ha in lamb sales, a gross margin of €1,210/ha and a net profit of €802/ha. Increasing prolificacy increased net profit on average by €336/ha, while stocking rate increased net profit on average by €84/ha and €19/ha for an increase from 10 to 12 ewes/ha and from 12 to 14 ewes/ha, receptively. In general the bio economic model showed that increasing the number of lambs weaned per hectare increased net profit per hectare but the greatest increase in profitability per hectare was achieved at the higher prolificacy level. Increasing the number of lambs weaned without increasing grass growth and utilization was economically counterproductive. Across all stocking rates, the high prolificacy scenarios were more profitable and had a greater capability to cope with fluctuations in key variables.

**Conclusion** The bio-economic model demonstrated that the number of lambs weaned per hectare, along with increased grass production and utilization, increases net profit. Increasing the number of lambs weaned per hectare reduces the cost of production per lamb and in-turn increases net profit per lamb. Increasing the number of lambs weaned per hectare without increasing grass growth and utilization is economically counterproductive.

**References**

Bohan, A., Shalloo, L., Malcolm, B., Ho, C. K. M., Creighton, P., Boland, T. M. and McHugh, N. 2016. Agricultural Systems 148, 124-134.
Earle, E., McHugh, N., Boland, T. M. and Creighton, P. 2016. Small Ruminant Research 143, 53-60.
Ho, C. K. M., Jackson, T., Harrison, M. T. and Eckard, R. J. 2014. Animal Production Science 54, 1248-1253.
Keady, T. W. J. and Hanrahan, J. P. 2006. Irish Grassland and Animal Production Association Journal 40, 15-27.
Teagasc. 2017. e-Profit Monitor Analysis Dry Stock Farms 2016.

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# More diverse swards improve lamb performance and reduce anthelmintic requirement compared to perennial ryegrass only swards

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**Application** Incorporating herbs into the swards of grazing sheep will reduce the requirement for chemical anthelmintic administration and reduce days to slaughter

**Introduction** Some of the main challenges facing sheep farmers in the future are profitability and sustainable control of internal parasites (Sykes, 1994). Grazing multispecies swards (containing herbs and legumes; MSS) has been shown to improve lamb performance (Kemp *et al*., 2010) and MSS have the potential for increased biomass production at lower inputs of nitrogen (Jing *et al*., 2017), thus increasing system profitability. In addition, herb containing mixtures have shown potential to reduce intestinal parasites (Marley *et al*., 2003). The objective of this study was to investigate the impact of PRG only versus MSS swards and non-herb containing versus herb containing swards on lamb performance from birth to slaughter in terms of average daily gain (ADG) and faecal egg count (FEC).

**Material and methods** Four farmlets were established; PRG receiving 163 kg N per hectare per year (N/ha/y) (PRG); PRG and white clover sward at 90 kg N/ha/y (PRGWC); six species sward (two grasses, two legumes, two herbs) at 90 kg N/ha/y (6S) and a nine species sward (three grasses, three legumes, three herbs) at 90 kg N/ha/y (9S). Each farmlet was stocked with 30 twin-rearing ewes at a stocking rate of 12.5 ewes/ha from turnout after lambing until lambs were slaughtered under rotational grazing management. Lambs were weighed and FEC was measured fortnightly. Lambs were weaned at 14 weeks and were drafted for slaughter at 45 kg live weight. Lambs received anthelmintics as group parasite burden reached a predetermined threshold for FEC and the days between each anthelmintic treatment and the number of treatments administered were recorded. Orthogonal contrasts were analysed between the non-herb containing swards and herb containing swards and between PRG only and MSS (more than one species) using SAS 9.4.

**Results** Treatment comparisons were previously presented (Grace *et al*., 2016) and are not discussed here. Orthogonal contrasts to investigate the impact of PRG only versus MSS and non-herb versus herb containing swards are presented in Table 1. Lamb weaning weight was higher in the MSS than the PRG only swards ($P<0.001$) and was higher in higher in herb containing swards compared to the non-herb containing swards. The MSS swards had reduced days to slaughter compared to the PRG only sward ($P<0.001$) due to differences in the ADG from birth to slaughter ($P<0.01$). In addition, the lambs grazing the PRG required more anthelmintic treatments than the lambs grazing the MSS ($P<0.0001$) and the lambs grazing the non-herb containing swards compared to the herb containing swards required more anthelmintic treatments ($P<0.0001$).

**Table 1** The effect of sward treatment on the average daily gain (ADG; g/day), weaning weight (kg), days to slaughter, days between anthelmintic treatment and no of anthelmintic treatments administered

| Parameter | PRG | PRGWC | 6S | 9S | SEM | PRG v MSS | Non-herb v herb |
|---|---|---|---|---|---|---|---|
| ADG 0 to 6 weeks | 292 | 305 | 339 | 321 | 0.7 | <0.0001 | <0.0001 |
| ADG 0 to weaning | 264 | 272 | 273 | 265 | 0.7 | <0.0001 | <0.0001 |
| Weaning weight | 30.6 | 31.9 | 32.6 | 31.7 | 0.57 | 0.0009 | 0.0237 |
| ADG from birth to slaughter | 236 | 249 | 254 | 251 | 0.6 | 0.001 | 0.0164 |
| Days to slaughter | 175 | 162 | 164 | 164 | 3.9 | 0.0002 | 0.0827 |
| Days from treatment 1 to 2 | 37.9 | 42.9 | 51.1 | 61 | 2.4 | <0.0001 | <0.0001 |
| No. of doses | 2.6 | 2.1 | 1.4 | 1.5 | 0.07 | <0.0001 | <0.0001 |

**Conclusion** Lambs grazing more diverse swards compared to PRG only had improved performance and a reduced requirement for chemical anthelmintics.

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**References**

Grace, C., Boland, T.M., Fritch, R., Sheridan, H. and Lynch, M.B. 2016. NIBIO, 397-399.
Jing, J., Soegaard, K., Cong, W.F. and Eriksen, J. 2017. Plos one 12**,** 169-208.
Kemp, P.D., Kenyon, P.R. and Morris, S.T. 2010. Revista Brasileira de Zootecnia 39**,** 169-174.
Marley, C., Cook, R., Keatinge, R., Barrett, J. and Lampkin, N. 2003. Veterinary parasitology 112**,** 147-155.
Sykes, A. 1994. Animal Science 59**,** 155-172.

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# Effect of birth type and weight on mortality and performance of artificially reared lambs

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**Application** The regression of weaning weight on birth weight is 2.9 kg/kg and this factor can be used to predict the impact on weaning weight of any management intervention that changes lamb birth weight.

**Introduction** In mid-season prime lamb production Keady and Hanrahan (2006) concluded that increasing litter size, and thus the number of lambs reared per ewe joined, is the most important determinant of production efficiency. As mean flock litter size increases, the incidence of triplets and higher multiples increases. For example, in flocks with mean litter sizes of 1.8, 2.0 and 2.2 the expected incidence of triplets is 8%, 15% and 25%, respectively. Consequently, an issue that faces producers with prolific flocks is how to cope with litters with ≥ 3 lambs. In some flocks one lamb from each set of triplets is either cross-fostered to single-bearing ewes, sold for cross-fostering, or artificially reared. Results from previous studies have shown that each 1 kg increase in the birth weight of lambs reared in a rotationally-grazed grass-based system increased weaning weight by 3.35 (Keady *et al* 2007), 3.16 (Keady and Hanrahan 2009a) and 3.41 kg (Keady and Hanrahan 2009b), respectively. The aim of the work reported here was to evaluate the effect of birth type and birth weight on the performance of lambs that were artificially reared.

**Material and methods** A total of 467 crossbred lambs (sired by Suffolk, Texel or Charollais rams; 5 singles, 114 twins, 277 triplets, 59 quadruplets, 12 quintuplets), which were born to ewes managed on a grass based system (rotational grazing) of prime lamb production at the Athenry Research Farm between 2006 and 2017, were removed from their dams, usually in their first days of life, and placed in an artificial rearing unit. Lambs had access to milk replacer offered *ad libitum* until approximately 7 weeks of age. Concentrate was introduced within 2 weeks of birth and offered *ad libitum* until lambs were drafted for slaughter. Lambs remained indoors, in slatted pens. Mean carcass weight and age at slaughter were19.5 kg and 151days, respectively. Data on growth were analysed using Proc MIXED (SAS) with year, dam and dam(season) as random terms and sex, birth type and dam age fixed.

**Results** The effect of birth type on lamb mortality and performance to 14 weeks of age are presented in Table 1. Lamb birth weight declined ($P < 0.001$) as birth type increased. As birth type increased from 2 to ≥4 mortality at 7 and 14 weeks of age increased ($P < 0.001$) and live weight at weaning ($P < 0.001$) declined. Birth weight of lambs that survived to weaning did not differ ($P > 0.05$) from those that failed to survive (+0.02 and +0.02 kg for twins and triplets, respectively). An analysis of the relationship between birth weight and weaning weight, adjusting for birth type, sex and dam age, yielded a significant linear relationship ($P<0.001$); the regression of weaning weight on birth weight was 2.94 (s.e. 0.446) kg/kg. Corresponding estimates based on the differences between means for twins and triplets, and between triplets and quadruplets were 2.65 (s.e. 0.68) and 3.99 (s.e. 1.82), respectively, yielding a weighted average of 2.81 (s.e. 0.638) kg/kg.

**Table 1** Effect of birth and rearing type on mortality and performance of artificially-reared lambs

| | | Litter size | | | |
|---|---|---|---|---|---|
| | | Single | Twin | Triplet | Quad/Quintuplet |
| Birth weight (kg) | | 5.88$^{a}$ (0.359) | 4.55$^{b}$(0.082) | 3.45$^{c}$ (0.055) | 2.79$^{d}$ (0.121) |
| Mortality (%) | -at 7 weeks | 20.0 | 9.6 | 12.6 | 16.9 |
| | -at 14 weeks | 20.0 | 14.0 | 17.0 | 29.6 |
| ADG (g/d) | 0-7 weeks | 170 (37.7) | 219 (10.4) | 232 (8.4) | 211 (12.6) |
| | 0-14 weeks | 229$^{ab}$ (43.6) | 305$^{a}$ (10.4) | 274$^{b}$ (8.8) | 252$^{b}$ (13.5) |
| Weaning weight (kg) | | 28.5$^{abc}$ (3.13) | 32.5$^{a}$ (0.70) | 29.5$^{b}$ (0.48) | 27.8$^{c}$ (1.03) |

$^{abcd}$Means (growth traits), within rows, without a common superscript are significantly different ($P < 0.05$)

**Conclusion** In artificial rearing systems, lamb mortality increases as birth type increases. Each 1 kg increase in birth weight increases weaning weight by 2.9 kg and the response is similar regardless of birth type. The consistency between the present estimates for the regression of weaning weight on birth weight and those from previous studies indicates that this factor can be used to predict the effect on weaning weight of any management intervention that changes birth weight.

**References**

Keady, T.W.J. and Hanrahan, J.P. 2006. Irish Grassland Association Journal 40, 15–27.
Keady, T.W.J. and Hanrahan, J.P. 2009a. Animal 3, 143–151.
Keady, T.W.J. and Hanrahan, J.P. 2009b. Animal 2009, 3, 879–890
Keady, T.W.J., Hanrahan, J.P., Flanagan, S. 2007. Irish Journal of Agricultural Research 46, 169–180.

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**061**

# A study of husbandry practices and abattoir data for 649 sheep farms in Great Britain and their association with flock productivity

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**Application** This study provides a quantitative estimate of the impact of key management practices such as weighing lambs between birth and weaning, vaccinating against Clostridial diseases and abortive agents, and worming ewes during quarantine on number of lambs sold per ewe per year.

**Introduction** Despite the size of the UK sheep breeding flock and the importance of sheep farming for the development of rural areas of the country, there is currently little quantitative evidence on the impact of different husbandry practices on flock productivity. Research in the past decades has shown the impact of single flock management decisions on flock production (Guyer & Dyer, 1954; Penning *et al.*, 1995), but no study has so far assessed the combined effect of several flock management practices on flock outputs. This study aimed at identifying flock husbandry practices associated with flock productivity using abattoir data as a proxy for flock production.

**Material and methods** A survey was developed with questions on husbandry practices (culling, vaccination, quarantine, worming, body condition scoring, lambing management, contact with a veterinarian and medicines usage), flock size and farm characteristics. From June to August 2016, sheep farms that supply lamb deadweight to a major retailer via two meat processors, were visited by independent assessors who completed the survey by interviewing the farmer about management practices applied during the previous year. Abattoir data (number of lambs sold per month per farm) relative to those farms was provided by the processors. Data was inserted in an Excel spreadsheet and analysed using STATA software. The outcome variables of interest were number of lambs sold per ewe per year, and number of lambs sold per grassland hectare per year. After checking data for errors, a descriptive analysis of results was performed, followed by univariable and multivariable regression modelling.

**Results** In total, data from 746 farms was collected. Farms purchasing store lambs were excluded from the analysis (n=78). Industry reference values were used to identify and exclude any extreme outliers or erroneous data (n=19). Final sample size used for analysis was 649 farms, located in Wales (n=395), England (n=186) and Scotland (n=68). Husbandry practices significantly associated with number of lambs sold per ewe in multivariable analysis ($p<0.05$) included vaccinating against agents causing abortion and clostridial disease, worming ewes with derquantel + abamectin (*Startect*) or monepantel (*Zolvix*) during quarantine, weighing lambs between birth and weaning, selecting ewes for culling based on body condition criteria, keeping records of ewes to be culled, and treating lame ewes with best practice (n= 565, r2 = 31%). Lowland farms and flocks managed by farmers who indicated that they had a good knowledge on the financial performance of their flock, had higher flock productivity. While no significant effect of lower ages at weaning (less than 17 weeks) was seen in terms of number of lambs sold per ewe, a significant association was observed with regards to number of lambs sold per hectare.

**Conclusion** This is the first study in the UK linking abattoir derived data to flock husbandry practices in order to identify management decisions associated with higher number of lambs sold per hectare per year. Results suggest association between biosecurity and disease control practices with greater flock efficiencies. Further research is needed to explore other factors influencing flock productivity, possibly related with grassland and genetics management, and to farmer managerial attitudes.

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**References**

Guyer, P., and Dyer, A. J. 1954. Research Bulletin. Missouri Agricultural Experiment Station.
Penning, P. D., Parsons, A. J., Orr, R. J., Harvey, A., and Champion, R. A. 1995. Applied Animal Behaviour Science 45 (1–2), 63–78.

062

# The impact of ewe prolificacy potential and stocking rate on lamb average daily gain and live weight output per hectare within a grass based system

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**Application** Increasing ewe prolificacy potential in combination with stocking rate greatly improves the efficiency of lamb output on a per hectare basis from grass based production systems

**Introduction** Ewe prolificacy potential (PP; predicted number of lambs born ewe$^{-1}$ year$^{-1}$) and stocking rate (SR; ewes ha$^{-1}$) are two of the most influential factors affecting lamb output (Keady *et al*., 2009) and the efficiency at which feed resources are utilised in grass-based lamb production systems (Young *et al*., 2010). Previous studies have tended to focus on efficiency at an individual animal level rather than on overall system efficiency. The objective of this study was to investigate the effect of ewe PP, SR, and their interaction on lamb live weight gain and lamb output at the system level.

**Material and methods** The study was a 2 x 3 factorial design, consisting of two differing ewe PP (medium prolificacy (MP) – Suffolk-sired crossbred ewes -weaning rate 1.5 lambs ewe$^{-1}$ and high prolificacy (HP) – Belclare-sired crossbred ewes- weaning rate 1.8 lambs ewe$^{-1}$) and three SR: low (LSR; 10 ewes ha$^{-1}$), medium (MSR; 12 ewes ha$^{-1}$) and high (HSR: 14 ewes ha$^{-1}$). The study was carried out over three production years. Each treatment was managed in a 5-paddock rotational system for the duration of the study. Lamb average daily gains (ADG; g day-1) were recorded from birth to finishing. The effect of ewe PP, SR, and their interaction on lamb ADG ha$^{-1}$ and lamb output were analysed using a linear mixed model in PROC GLM (SAS, 2012) with ewe PP, SR, year, and the interaction between ewe PP and SR included as fixed effects.

**Results** High PP ewes produced a higher number of lambs born ewe$^{-1}$ (+0.20 lambs; P<0.001) and hectare$^{-1}$ (+ 1.5 lambs; P<0.05) and weaned an extra 0.18 lambs ewe$^{-1}$ (P<0.01) and 2.01 lambs hectare$^{-1}$ (P<0.01). Stocking rate had no effect on the number of lambs born or weaned per ewe (P>0.05). However, the number of lambs born and weaned differed on a hectare$^{-1}$ basis with the lowest number reported at the LSR, intermediate at the MSR, and highest at the HSR (P<0.001). . The HP treatment achieved a 0.45 kg higher lifetime (birth to slaughter) lamb ADG ha$^{-1}$ compared to the MP treatment (P<0.001; Fig. 1). Total lifetime lamb ADG ha$^{-1}$ was lowest at the LSR, intermediate at the MSR, and highest at the HSR (P<0.001). Total lamb live weight output per ha$^{-1}$ also increased with increased prolificacy potential and stocking rate as shown in Figure 2.

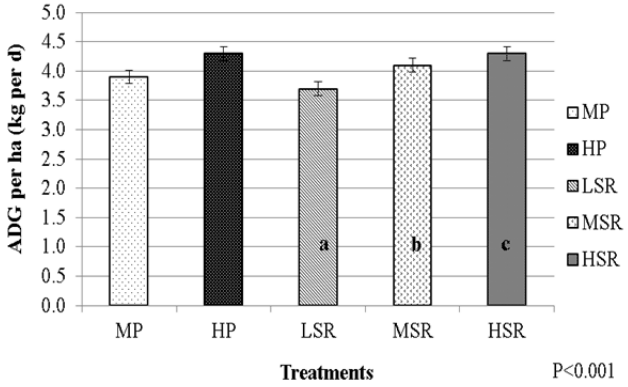


**Figure 1** Effect of ewe prolificacy potential and stocking rate on lifetime daily lamb live weight (kg) gain ha$^{-1}$

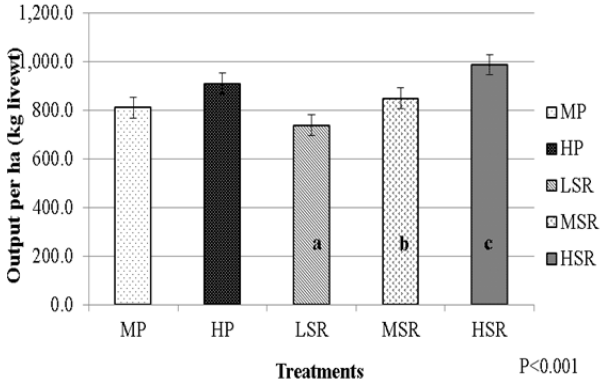


**Figure 2** Effect of ewe prolificacy potential and stocking rate on total lamb live weight (kg) output ha$^{-1}$

**Conclusion** Results from this study demonstrate to potential to increase the level of lamb production and output ha$^{-1}$ within grass based systems by increasing ewe PP and SR.

**Reference**

Keady, T.W.J., Hanrahan, J.P., Flanagan, S., 2009. An evaluation of two grassland-based systems of mid-season prime lamb production using prolific ewes of two genotypes. Irish Journal of Agricltural & Food Research 48, 87–101.
Young, J. M., Thompson, A.N. and Kennedy, A J. 2010. Bioeconomic modelling to identify the relative importance of a range of critical control points for prime lamb production systems in south-west Victoria. Animal Production Science 50, 748–756.

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# Effect of forage to concentrate ratio and fat supplementation on milk composition in dairy sheep: A meta-analysis

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**Application** The determination of adequate levels of inclusion of rumen protected fat (RPF) and Forage:Concentrate ratio (F:C) that improves the milk chemical composition and fatty acid (FA) profile is useful in the nutritional management of dairy sheep at farm level.

**Introduction** Sheep products have received attention due to the possibility of being enriched with fatty acids potentially beneficial to health, especially vaccenic acid (VA), and α-linolenic acid (ALA, C18: 3, n3). The improvement of the nutritional characteristics of sheep milk, mainly in relation to the profile of FA, has led to research towards the study of factors that affect its chemical composition, which also determines the yield and quality of the dairy products derived from it. Nutritional management has the greatest impact in determining FA profile in milk. The objectives of the present study were (1) to analyze the effect of F:C in sheep rations on milk yield production, fat content and conjugated linoleic acid (CLA); and (2) to quantify the overall effect of rumen protected fat supplementation in dairy sheep on milk production, fat, protein and CLA content in milk.

**Material and methods** Two separate meta-analyses were performed to analyze the main nutritional factors (F:C and fat supplementation) that affect milk production and composition in dairy sheep. A total of 55 trials that focused on addition of protected fats and 20 trials focused in F:C rations were analyzed. The method proposed by Hedges (1981) was used to calculate the effect size. The estimation of fixed and random effects was calculated as Dersimonian and Laird (1986). Between studies heterogeneity was quantified using the $I^2$ statistic: for response variables that showed substantial heterogeneity ($I^2$ > 50 %) were constructed mixed-effect models through the inclusion of one or more explanatory variables that could be sources of heterogeneity, follow the methodology previously described Appuhamy *et al.* (2013). A meta-regression analysis was used to explore the sources of heterogeneity of responses such as fat content and composition. All analysis were conducted in R statistical software (R Core Team, 2016) using the 'meta' package version 4.6-0 (Schwarzer, 2016).

**Results** Compared to sheep fed high-concentrate diets (HC; concentrate content > 55% of DM), those fed high-forage diets (HF; forage content > 60% of DM) had 0.087 L/d lower milk yield and 0.8 g/100 g milk protein content. However, fat content and CLA (Fig. 1) concentrations were greater in dairy sheep fed HF (0.32 g/100g and 2.28 mg/g, respectively). The trials that tested RFP averaged an inclusion level of 96.5 ± 60.8 g of DMI. The addition of RPF in sheep diets had a positive effect size on fat (0.22 g/100g), and CLA concentrations (3.98 mg/g) in milk, but protein concentration was reduced (P <0.001). The heterogeneity was higher to 50% ($I^2$ > 88%) in all the analysed variables; the inclusion level of RPF was significant (P <0.001) in the meta-regression with an increase in fat and CLA content as it was increased the RPF inclusion level.

**Conclusion** We conclude that greater availability of unsaturated fatty acids in forage based rations can explain the greater level of fat and CLA content in milk; also, the addition to RPF showed significant effects on milk production and composition, where milk yield, fat and CLA content increased and protein content decreased.

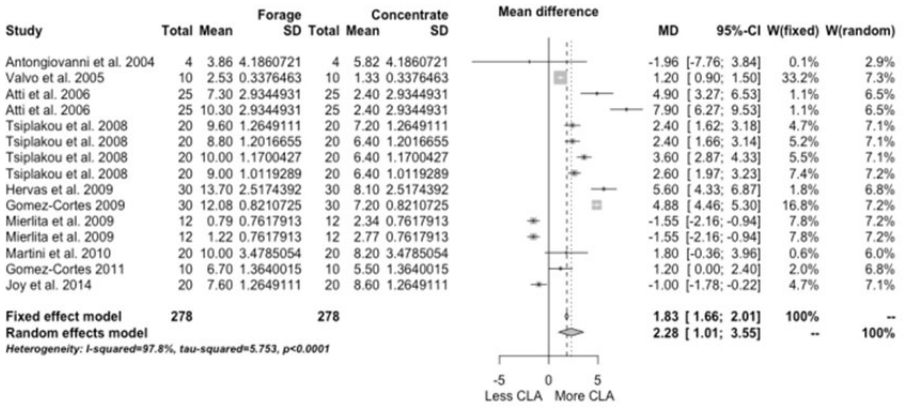


**Figure 1** Forest plot of the milk CLA content (mg/g methylated fatty acid) for the studies focused on comparing forage-based vs. concentrate-based rations in dairy sheep.

## References

Appuhamy J.A.D.R.N., Strathe, A.B., Jayasundara, S., Dijkstra, J., France, J., Kebreab, E. 2013. Journal of Dairy Science 96, 5161–5173
Dersimonian, R. and Laird, N. 1986. Control Clinical Trails 7, 177–188.
Hedges, L. V. 1981. J. Educ. Behav. Stat. 6, 107–128.
R, Core Team 2016. R: A language and environment for statistical computing.
Schwarzer, G. 2016. meta: General Package for Meta-Analysis.

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# Causes and time of neonatal mortality in grass-based systems of lamb production

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**Application** To reduce neonatal mortality, preventive recommendations should focus on best practices during pregnancy and at birth, in particular on the two potentially preventable causes of death (infection and dystocia).

**Introduction** Causes of lamb mortality may be defined as ultimate (initiating) or proximate (final), the former (e.g. traumatic lambing assistance) meaning the precipitating cause and the latter the terminal cause (e.g. major complex polytrauma). Only by identifying the preventable and non-preventable ultimate causes of mortality can farmers, vets and advisors reduce lamb mortality**.** The objective of the current study was to determine the ultimate causes of neonatal lamb mortality in mid-season, grass-based systems of prime lamb production.

**Material and methods** The population at risk for this observational study was 3 flocks (1,100 ewes and 1,855 full term lambs) lambing at the Teagasc Research Centre, Athenry in spring 2017. Flock 1 consisted of three ewe genotypes; Belclare, Suffolk x Belclare and >75% Suffolk. Flock 2 consisted of 4 ewe genotypes; Belclare x Suffolk, Belclare x Texel, Suffolk x Cheviot and Suffolk x Texel. The ewes in these two flocks were joined with Charolais rams. Flock 3 consisted of purebred Belclare, Suffolk or Texel ewes from New Zealand or Ireland. The three flocks were housed in early December and lambed indoors. Flocks 1 and 3 were shorn in December and Flock 2 was shorn in May. All ewes were offered grass silage ad-libitum during the housing period. Concentrates were offered according to silage feed value and expected litter size during the final 6 weeks of gestation. Ewes were turned out to pasture within 3 days of lambing. All full-term foetuses and lambs which died within a week of birth (n=172) were submitted for necropsy examination to the National Veterinary Laboratory Service. All lambs (dead or alive) were tagged and weighed at birth. A recording form containing the dam, lamb, lambing and mortality details was submitted with each carcass. A time-of-death was assigned to each lamb based on the submission form details and the results of the necropsy; 0h (stillborn; did not breath), Day 1 (breathed and died within 24h) and Day 2 to 7. Where a cause of death could be assigned these were aggregated into 5 main groups (accident, congenital defect, dystocia, infection and other) and the remaining cases were categorised as diagnosis not reached (DNR). Cause of death was analysed as a multinomial variable using logistic regression with time-of-death category as the explanatory variable. The analysis was fitted using the Logistic procedure in SAS 9.4.

**Results** The mean neonatal lamb mortality rate was 9.3% (8.8-9.6% by flock). As the mortality rate did not differ between flocks, the overall dataset was analysed. In total, 113, 22 and 37 lambs died at 0h, Day1 and Days 2-7, respectively. Infection was the main cause of mortality, accounting for 37.7% of deaths. Lambs dying between Days 2-7 were more likely to die due to infection ($P < 0.05$) than lambs that were stillborn or lambs that died in Day 1 (Table 1). Dystocia was the second most common cause of mortality. Lambs dying at birth and in Day 1 were more likely ($P<0.05$) to die due to dystocia than those dying between Days 2-7. Infection and dystocia combined accounted for 52.8% of all neonatal lamb mortality. Accidents were the third most common cause of mortality and they occurred equally in all three time-of-death groups. The cause of mortality was not diagnosed for 28.5% of lambs; these were predominantly stillborn.

**Table 1** Ultimate causes of neonatal (0-7days) mortality in 172 lambs (% of lambs that died)

| Cause of death | Time of death | | | |
|---|---|---|---|---|
| | 0h | Day 1 | Day 2-7 | Total |
| Accident | 1.7[a] | 2.9[a] | 3.5[a] | 8.1 |
| Congenital defect | 1.2[a] | 0.6[a] | 1.2[a] | 3 |
| Dystocia | 11[a] | 3.5[a] | 0.6[b] | 15.1 |
| Infection | 16.8[a] | 5.2[a] | 15.7[b] | 37.7 |
| Other | 1.7[a] | 4.1[b] | 1.7[a,b] | 7.5 |
| Diagnosis not reached (DNR) | 19.2[a] | 5.2[a,b] | 4.1[b] | 28.5 |
| Total | 51.6 | 21.5 | 26.8 | 100 |

**Conclusion** The majority of neonatal losses occurred at birth. Infection and dystocia were the most common causes of death overall. These causes of death are potentially preventable. The high DNR rate, particularly in stillbirths, requires further research.

**Acknowledgements** The authors acknowledge the Teagasc Walsh Fellowship scheme, the Regional Veterinary Laboratories and the technical and farm staff at Athenry for their assistance.

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# Production system and gender effects on intramuscular fat deposition and indicators of tenderness of beef from late maturing breeds

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**Application** In late maturing bulls and steers finished at pasture, shear force and collagen profile, which are indicators of tenderness, are unaffected by concentrate supplementation but shear force is lower and fat content is higher in steers.

**Introduction** Current beef production systems in Ireland mainly involve steers slaughtered at 24 months or more. Higher feed efficiency and growth rate of bulls compared to steers has led to a shift towards production of beef from bulls O'Riordan *et al*. (2011). Furthermore, reduction of production costs is being explored through the inclusion of a grazing period before slaughter Mezgebo *et al*. (2017). There is little information comparing steer and bull beef when animals are raised to a similar age at pasture. Therefore, the objective of the study was to compare indicators of tenderness such as Warmer-Bratzler shear force (WBSF), collagen profile (i.e. soluble collagen, insoluble collagen, total collagen), as well as the intramuscular fat (IMF) content of beef from bulls and steers on pasture based production systems and slaughtered at the same age.

**Material and methods** Late maturing (Charolais and Limousin crossbred) bulls (n=30) and steers (n=30) were finished for 120 days on either pasture only (P) or pasture with 40% barley-based concentrate on a dry matter basis (PC). Treatments were balanced for breed. The animals were slaughtered at 19 months of age and *Longissimus thoracis et lumborum* was harvested 48h post slaughter. A sample for collagen and IMF analysis was removed before aging while the sample for WBSF measurement was collected after a 14-day aging period; both were stored at -20 °C until required for analysis. Collagen was analysed using the method of Kolar (1990) and WBSF was measured according to the procedure of Shackelford *et al*. (1994). The IMF content was measured using a bench-top nuclear magnetic resonance instrument (AOAC, 2000). Data was subjected to analysis of variance using the Proc mixed procedure of SAS 9.4 with production system, gender and interaction term as fixed effects.

**Results** The main effects of production system (P vs PC) and gender (bulls vs steers) are shown in Table 1. Collagen characteristics were not affected by production system or gender while WBSF and IMF were affected by gender and there was an interaction between production system and gender for WBSF.

**Table1** Production system and gender effects on collagen, WBSF and IMF

| | Pasture + concentrate | | Pasture | | | P-Values | | |
|---|---|---|---|---|---|---|---|---|
| | Bulls | Steers | Bulls | Steers | SEM | PS | Gender | PS*Gende |
| Soluble collagen (mg/g) | 0.48 | 0.52 | 0.50 | 0.43 | 0.02 | n.s | n.s | n.s |
| Insoluble collagen (mg/g) | 4.37 | 4.42 | 4.61 | 4.43 | 0.12 | n.s | n.s | n.s |
| Total collagen (mg/g) | 4.85 | 4.93 | 5.12 | 4.86 | 0.13 | n.s | n.s | n.s |
| Soluble collagen (%) | 9.79 | 10.42 | 9.79 | 8.95 | 0.26 | n.s | n.s | n.s |
| WBSF (N) | 46.0 | 31.6 | 39.5 | 33.9 | 1.28 | n.s | <.0001 | 0.03 |
| IMF (g/kg) | 2.10 | 12.9 | 1.90 | 8.2 | 0.90 | n.s | <.0001 | n.s |

**Conclusion** Finishing bulls on grass results in beef that is leaner with higher shear force values compared to steer beef from similar production systems. The supplementation of bulls at pasture with 40% barley based concentrate does not decrease shear force or increase fat content in bulls slaughtered at 19 months of age.

**Acknowledgements** The financial support of Food Institutional Research Measure of the Irish Department of Agriculture, Food and the Marine (11/SF/322) is gratefully acknowledged

**References**

Association of Official Analytical Chemists 2000. Official methods of analysis, Method no. 985.14
Kolar, K. 1990. Journal-Association of Official Analytical Chemists 73, 54–57.
O'Riordan, E. G., Crosson, P. and McGee, M. 2011. Irish Grassland Association Journal 45, 131–146.
Mezgebo, G. B., Monahan, F. J., McGee, M., O'Riordan, E. G., Picard, B., Richardson, R. I. and Moloney, A. P. 2017. Animal 11, 1636–1644.
Shackelford, S. D., Koohmaraie, M., Cundiff, L. V., Gregory, K. E., Rohrer, G. A. and Savell, J. W. 1994. Journal of Animal Science 72, 857-863.

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# Prediction of beef *M. longissimus thoracis et lumborum* sensory quality using visible and near infrared spectroscopy

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**Application** Visible – Near Infrared Spectroscopy (VisNIRS) is a rapid and non-destructive technology that has potential to predict beef quality in the factory setting. This technology may potentially be utilized by breeding societies to facilitate the improvement of beef eating quality by genetic selection.

**Introduction** Eating satisfaction, sensory characteristics and nutritional content are key factors that influence consumers in regard to the purchase of fresh meat (Grunert, *et al.,* 2004). Analysis of beef sensory traits by way of a trained sensory panel is time consuming, expensive and destructive of the sample (Prevolnic, *et al.,* 2004). VisNIRS has been proposed as a rapid, non-destructive technique with the potential to predict sensory performance of beef. The aim of this study was to assess the relevance of VisNIR spectra for the prediction of beef sensory traits using chemometric modelling approaches.

**Material and methods** Crossbred beef bulls and steers (18±4 month old, n= 209) finished under controlled feeding and environmental conditions were slaughtered in 12 batches in a commercial plant by electrical stunning followed by exsanguination. ASD Labspec 5000 (ASD Inc., Boulder Colorado, USA) VisNIR spectrometer fitted with a high-intensity contact probe with a 10 mm spot size was used to collect spectra between 350-2500 nm with 1 nm intervals, using the Indico Pro program (ASD Inc.). Spectra were collected in triplicate, for each given scan 20 spectra were collected consecutively and averaged to reduce the noise effect, then saved in reflectance mode - log (1/*R*). VisNIR spectra were recorded on the cut surface of the *LTL* muscle at 24 h *post mortem*, immediately after cutting and after 1h blooming (25 h PM). This was repeated at 48 h and 49 h, respectively. Steaks with a thickness of 2.54cm were removed from the *LTL* at 48 h *post-mortem* (PM), vacuum packaged, aged for 14 days at 4°C and frozen at -20°C. Sensory analysis of beef steaks was assessed using twelve trained panellists at Teagasc Ashtown following a modified version of the American Meat Science Association Research Guidelines for Cookery, Sensory Evaluation and Instrumental Tenderness Measurements of Meat (2nd Edition, Version 1, March 2015). Samples were cooked on a clamshell grill to an internal temperature of 70ºC then served to a twelve member trained sensory panel to assess eleven sensory traits, on a 0-100 line scale. The Unscrambler X version 10.3 (CAMO ASA, Oslo, Norway) was used for PLSR model calibration. Chemometric models were constructed with spectra from four different time points and five different wavelength ranges (full, 350-2500nm; clipped, 450-2300nm; visible, 450-779nm; near-NIR, 780-1099nm; NIR, 1100-2300nm) and using three different mathematical pre-treatments: no treatment, Savitzky-Golay smoothing (SG) and Standard Normal Variate (SNV).

**Results** $R^2$Cal predictions for prediction of eleven beef sensory traits are shown in Table 1. Coefficients of determination obtained for tenderness and juiciness are moderate ($R^2$Cal <0.25). However, predictions for textural traits such as chewy, stringy, crumbly and fatty/greasy mouthfeel are somewhat higher ($R^2$Cal 0.37-0.54). $R^2$Cal values for flavour and after effect traits such as blood/metallic/iron flavour, beefy after effect, blood/metallic after effect and fatty/ greasy after effect ranged from 0.36-0.46.

**Table 1** Prediction of trained panel sensory scores on beef samples using VisNIRS measurements

| | Tender | Juicy | Chewy | Stringy | Crumbly | F/G mf | Beefy flav | B/M/I flav | Beefy a.e | B/M a.e | F/G a.e |
|---|---|---|---|---|---|---|---|---|---|---|---|
| n | 160 | 205 | 166 | 166 | 209 | 209 | 205 | 205 | 160 | 205 | 205 |
| $R^2$Cal | 0.25 | 0.15 | 0.38 | 0.37 | 0.54 | 0.53 | 0.25 | 0.36 | 0.38 | 0.36 | 0.46 |
| Time (PM) | 49 | 24 | 48 | 48 | 25 | 25 | 24 | 24 | 49 | 24 | 24 |
| Math treatment | None | SNV | None | None | None | SNV | SNV | None | SNV | None | None |
| Range (nm) | Full | Full | Vis | Vis | Full | Full | Vis | NN | NN | NN | Full |

n, number of samples; $R^2$Cal, coefficient of determination of calibration; F/G mf, fatty/greasy mouthfeel; B/M/I flav, blood/metallic/iron flavour; Beefy a.e, beefy after effect; B/M a.e, blood/metallic after effect; F/G a.e, fatty/greasy after effect; SNV, standard normal variate.

**Conclusion** The results of this experiment indicate that VisNIRS has potential to predict beef sensory scores, with many traits related to texture well-predicted.

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**References**

Grunert, K., Bredahl, L., and Brunso, K. 2004. Meat Science 66, 259-272
Prevolnic, M., Candek-Potakar, M. and Skorjanc, D. 2004. Czech Journal of Animal Science 49, 500-510.

# The use of cholecalciferol and ergocalciferol sources, to enhance beef vitamin D concentration and its subsequent effect on beef tenderness

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**Application** The biofortification of vitamin $D_3$ is a more effective strategy to produce vitamin D enriched beef, and has potential of being a food based strategy for increasing human vitamin D dietary intake.

**Introduction** Vitamin D deficiency and associated health risks are very much to the forefront of public health policy, particularly in Europe and northern latitudes (Cashman *et al*., 2015). Consequently, there is a definite need for innovatively designed natural food based vitamin D-enhancement strategies which cover a range of stable food sources. Beef is among the few dietary staple foods that contains, naturally occurring vitamin D. Recently, there has been a growing interest in the bio-addition approach as a means for increasing vitamin D intake through biofortification of livestock feeds. Although, there is limited research investigating the biofortification of beef animal diets with vitamin $D_3$ and vitamin $D_2$ sources at allowable EU vitamin D levels. The use of supplemental vitamin D prior to slaughter in beef animals may also have additional benefits of improving meat tenderness. Therefore, the objective of this study was to assess the effects of addition of synthetic vitamin $D_3$ and vitamin $D_2$ as well as UVB-exposed mushroom derived vitamin $D_2$ to the diets fed to beef heifers on beef total vitamin D activity as well as on meat quality.

**Material and methods** Thirty continental (Charolais + Limousin cross) heifers were blocked (on the basis of live weight and age) and randomly allocated to 3 dietary treatments (1) basal diet + 4000 IU vitamin $D_3$/kg of feed (Vit D3); (2) basal diet + 4000 IU vitamin $D_2$/kg of feed (Vit D2); (3) basal diet + 4000 IU vitamin $D_2$-enriched mushrooms (Mushroom $D_2$). Experimental design was a randomised complete block. Dietary treatments were offered for the final 28 days of an 80 day finishing period. The basal diet consisted of a standard *ad-libitum* finishing regime of concentrates and forage (straw) offered at a ratio of 90:10. Animal growth was recorded weekly and individual dry matter intakes were recorded using Calan, Broadbent controlled feeding system. *Longissimus thoracis* (LT) muscles were excised 10 days post slaughter for vitamin $D_3$ analysis carried out by High Performance Liquid Chromatography. LT warner bratzler shear force (WBSF) analysis was carried out according to Wheeler *et al*. (1996) with minor modifications. Sensory analysis of cooked LT steaks was performed in duplicate by a total of 40 naïve assessors over two analysis days as described by O'Sullivan *et al*. (2003). Data were analysed using the MIXED procedure of SAS (SAS, version 9.4). The experimental model included fixed effects of treatment and pen, with block included as a random effect.

**Results** Heifers offered the Vit $D_3$ enriched diet exhibited the highest serum 25-OH-D ($P < 0.001$), compared to Vit $D_2$ and Mushroom $D_2$, with no significant ($P > 0.05$) between these latter two groups. Consequently, heifers offered the Vit $D_3$ had the highest ($P < 0.05$) LT total vitamin D activity compared to either of the two vitamin $D_2$ supplementation sources (Figure 1). Additionally, LT total vitamin D activity did not differ ($P > 0.05$) between the Vit $D_2$ and the Mushroom $D_2$ treatment groups. Dietary treatment did not alter ($P > 0.05$) WBSF values. Mean sensory scores of parameters including appearance, odour, texture, flavour, overall accept and off flavour were not affected ($P>0.05$) by dietary treatments offered. Similarly, dietary treatment had no effect ($P > 0.10$) on animal performance parameters or any carcass related variables including, carcass weight, kill out %, carcass conformation and fat score.

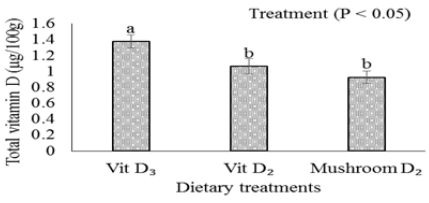


**Figure 1** Effect of dietary treatment on total LT vitamin D activity.

**Conclusion** Vitamin $D_3$ supplementation of cattle diets is a more effective source to increase total beef vitamin D content compared to natural or synthetic vitamin $D_2$ sources. Irrespective of vitamin D source no negative alterations to any sensory or meat quality parameters is an important finding from a consumer acceptance viewpoint.

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**References**

Cashman, K.D. and Hayes, A. 2017. Meat Science 132, 196-203.
O'Sullivan, M., Byrne, D. and Martens, M. 2003. Meat Science 63(1), 119-129.
Wheeler, T., Shackelford, S. and Koohmaraie, M. 1996. Journal of Animal Science 74, 1553-1562.

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# Animal performance and the appearance and stability during retail display of *M. longissimus* muscle from steers fed increasing amounts of barley or fodder beet-based supplements to grass silage

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**Application** If fodder beet is cheaper, it can replace barley in cattle rations without deleterious effects on growth, meat colour or shelf-life.

**Introduction** Grass silage is the main winter forage fed on Irish cattle farms, and is most commonly supplemented with cereal grain to achieve growth targets for finishing cattle. Fodder beet roots can be an economically viable alternative to cereal grains on some farms (Finneran, 2012). If there are differences in the appearance or quality characteristics of beef due to the use of fodder beet, the ability of beef producers to service existing markets could be compromised. The objective of this experiment was to establish selected beef quality responses to fortifying grass silage with increasing amounts of barley or fodder beet.

**Material and methods** Continental crossbred steers (48) were blocked by liveweight (515 (19.4) kg mean (s.d.)) and randomly allocated from within block to eight dietary treatments. The treatments were: grass silage *ad libitum* fortified with barley (830 g rolled barley, 100 g soyabean meal, 50 g molasses and 20 g mineral + vitamin premix per kg) at 2.5, 5.0 or 7.5 kg per head daily or with fodder beet (930 g fodder beet roots, 64 g soyabean meal and 6 g mineral + vitamin premix per kg) at equivalent dry matter (DM) inputs as for barley. Barley and fodder beet were also offered *ad libitum* with 1.5 kg grass silage DM. Animals were individually fed the experimental rations for 112 days after which selected carcass and meat variables were measured. Muscle shelf-life was determined by aerobic display in a refrigerated, illuminated cabinet for 16 days. The data were subjected to regression analysis with a model that had block, type and level of supplement and their interaction as effects. Linear and quadratic response patterns were tested.

**Results** Grass silage had a DM digestibility of 676g/kg and was well preserved (pH 3.8; lactic, acetic, propionic and butyric acids of 124, 24, 2 and 1 g/kg DM; ethanol of 9 g/kg DM; ammonia-N of 76 g/kg N). The crude protein concentration of the barley and fodder beet-based supplement was 139 and 127 g/kg DM, respectively. While barley permitted a higher intake of silage DM than fodder beet, type of supplement did not affect animal performance or meat quality (Table 1). Increasing the supplement allowance linearly affected most variables examined (Table 1). Fodder beet tended ($P<0.1$) to cause greater lipid oxidation in muscle than barley at higher levels of inclusion (data not shown) but the level of oxidation within the length of aerobic display is unlikely to be of commercial importance.

**Table 1** Intake, growth and meat quality attributes

| | Type of supplement | | Level of supplement[1] | | | | | Significance | |
|---|---|---|---|---|---|---|---|---|---|
| | Barley | Fodder | 2.03 | 4.05 | 6.06 | AL | sed | Type | Level |
| Silage DM intake (kg/d) | 4.07 | 3.60 | 5.76 | 4.72 | 3.40 | 1.48 | 0.380 | * | L* |
| Total DM intake (kg/d) | 9.14 | 8.93 | 7.79 | 8.77 | 9.48 | 10.10 | 0.407 | ns | L* |
| Liveweight gain (g/d) | 1005 | 1047 | 802 | 949 | 1043 | 1312 | 157.5 | ns | L* |
| Carcass gain (g/d) | 704 | 699 | 559 | 657 | 668 | 922 | 122.3 | ns | L* |
| Fat yellowness | 16.2 | 15.9 | 15.9 | 16.4 | 16.4 | 15.6 | 0.56 | ns | L*,Q* |
| Muscle pH | 5.49 | 5.48 | 5.50 | 5.48 | 5.47 | 5.50 | 0.019 | ns | ns |
| Muscle lightness | 35.5 | 35.6 | 34.5 | 36.2 | 35.4 | 36.0 | 1.18 | ns | L* |
| Muscle redness (day 16)[2] | 6.9 | 6.0 | 5.8 | 7.1 | 6.7 | 6.2 | 1.39 | ns | ns |
| Lipid oxidation (day 16)[2,3] | 0.55 | 0.58 | 0.56 | 0.53 | 0.49 | 0.69 | 0.125 | ns | L*, Q* |

[1]kg DM/d or ad libitum, [2]Of aerobic display, [3]Thiobarbituric acid reactive substances (mg/kg)

**Conclusion** When balanced for protein and minerals both barley grain and fodder beet roots supported similar performance by finishing steers across a range of rates of input, with little effect on carcass fat colour or meat shelf-life..

**Acknowledgements Teagasc,** Oakpark colleagues for producing the fodder beet and providing the processing machinery.

**References**

Finneran, E., Crosson, P. and O'Kiely, P. 2012. Irish Grassland Association Journal 46, 157-167.

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# Physiological markers of stress and carcass pH of Holstein-Friesian bulls and steers managed under varying production systems

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**Application** The results of this study showed that physiological markers of stress reflect differences in *post mortem* carcass pH decline and ultimate pH, which is a contributor to meat quality at commercial level.

**Introduction** Environmental factors such as gender and age at slaughter for beef cattle finishing systems are predetermined by the producer. Finishing male dairy calves at a younger age as bulls compared to the traditional 24 month old steer production system has the potential to increase beef output per hectare. Although bulls are typically slaughtered at a younger age than steers, many studies have reported that bulls were more susceptible to pre-slaughter stress. Relationships have been observed between animal temperament and stress (King *et al*., 2006). Additionally, cattle with a nervous disposition have been reported to have a greater incidence of dark cutting beef and greater Warner-Bratzler shear force values than their contemporaries (Falkenberg *et al*., 2005). Consequently, the objective of this study was to investigate whether differences in physiological markers of stress and post-mortem carcass pH exist among Holstein-Friesian (HF) bulls and steers slaughtered under varying production systems.

**Material and methods** Data were available from 46 HF animals; 17 bulls slaughtered at 15 months (15MO), 15 bulls slaughtered at 19 months (19MO) and 14 steers slaughtered at 24 months (24MO). On the morning of slaughter, animals were loaded at 0430 and transported a distance of 63 km to the abattoir. Blood samples were collected from the animals in the lairage 1 hour pre-slaughter which was stored in ice for approximately 2 hours and were subsequently stored at 4°C for 8 hours. Samples were collected for plasma concentrations of cortisol, creatine kinase, lactate, haptoglobin and glucose. pH decline was monitored for six consecutive hours post-slaughter on the *M. longissimus thoracic* at the 10$^{th}$ rib. Prior to deboning, ultimate pH was recorded again at the 10$^{th}$ rib. The effect of production system on physiological markers of stress, carcass pH decline and ultimate pH were analysed using mixed models ANOVA (Mixed procedure of SAS, v9.4).

**Results** Plasma concentrations of cortisol were greater (P<0.001) for 15MO than 19MO or 24MO (Table 1). Creatine kinase concentrations were similar for all treatment groups. Haptoglobin concentrations were greatest (P<0.001) for 19MO, lowest for 24MO and intermediate for 15MO. Glucose concentrations were greatest (P<0.001) for 24MO, intermediate for 15MO and lowest for 19MO. The concentration of lactate was greatest for 15MO, lowest for 24MO and intermediate for 19MO (P<0.001). Ultimate pH was greater (P<0.001) for 15MO than either 19MO or 24MO. Decline in pH was greatest for 24MO, lowest for 15MO and intermediate for 19MO (P<0.001).

**Table 1** Effect of production system on physiological markers of stress and carcass pH

| | 15MO | 19MO | 24MO | s.e. | P-value |
|---|---|---|---|---|---|
| Cortisol (ng) | 26.2$^{a}$ | 13.8$^{b}$ | 13.5$^{b}$ | 4.2 | <0.05 |
| Creatine kinase (U/L) | 422 | 392 | 295 | 52.4 | 0.2677 |
| Haptoglobin (Hgb binding capacity/L) | 0.67$^{a}$ | 0.84$^{b}$ | 0.36$^{c}$ | 0.034 | <0.001 |
| Glucose (mmol/L) | 5.41$^{a}$ | 5.02$^{b}$ | 5.53$^{c}$ | 0.122 | <0.05 |
| Lactate (U/L) | 4.15$^{a}$ | 2.52$^{b}$ | 1.12$^{c}$ | 0.38 | <0.001 |
| pH decline | 0.47$^{a}$ | 0.56$^{b}$ | 0.82$^{c}$ | 0.04 | <0.001 |
| Ultimate pH (0-14) | 5.73$^{a}$ | 5.53$^{b}$ | 5.51$^{b}$ | 0.046 | <0.001 |

**Conclusion** Results from this study show that bulls slaughtered at 15 months of age, i.e. the specified bull production system for male dairy calves for the UK market, had higher levels of cortisol concentrations at slaughter and had a slower pH decline than 19MO and 24MO. Although bulls slaughtered at 15 months had a greater ultimate pH, results from this study showed that ultimate pH was within normal ranges (5.4 to 5.8) for all production systems.

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**References**

Falkenberg, S. M., Miller, R. K., Holloway, J. W., Rouquette, F. M., Jr., Randel, R. D., & Carstens, G. E. 2005. In Proceedings 51st International Congress of Meat Science and Technology, p. 29.
King, D. A., Pfeiffer, C. E. S., Randel, R. D., Jr., T. H. W., Oliphint, R. A., Baird, B. E., Jr., K. O. C., Vann, R. C., Hale, D. S., Savell, J. W. 2006. Meat Science 74, 546-556.

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# Effect of duration of concentrate feeding on the n-3 PUFA content of lamb

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**Application** For lambs grazed on pasture, a shorter pre-slaughter period on a barley/maize cereal-based concentrate results in lower intramuscular fat (IMF) and a fatty acid composition that is more favourable from a human health perspective.

**Introduction** Nutritionally favourable polyunsaturated fatty acid:saturated fatty acid (PUFA:SFA) and n-6:n-3 PUFA ratios in lamb arise from feeding pasture in comparison to cereal concentrates (Scerra *et al.,* 2011). Where pasture is not available throughout the year, pre-slaughter supplementation with cereal concentrate can eliminate the effects of pasture on the intramuscular fatty acids (IMFA) (Scerra *et al.,* 2011; Aurousseau *et al.,* 2007). The objective of this study was to determine the effect, on the IMFA composition, of duration of finishing lambs on a concentrate ration following grazing at pasture.

**Material and methods** Thirty-three male lambs grazing at pasture from birth were allocated to a barley/maize-based concentrate and individually fed for one of three finishing durations prior to slaughter (36, 54 or 72 days). At 24 h after slaughter the *M. longissimus thoracis et lumborum* (LTL) was aged for 8 days under vacuum and stored at -20°C for the quantification of total IMFA (Brunton *et al.,* 2015). All data, except for the PUFA:SFA ratio, were normalized using a Box-Cox transformation prior to analysis using the PROC MIXED procedure of SAS (v9.4), where feeding duration was considered as the main effect. Multiple comparisons of the means were performed following Bonferroni adjustment. Back-transformed data are presented in the results.

**Results** A short pre-slaughter period (36 days) on the concentrate diet, or a longer grazing period on pasture, resulted in a lower IMF content (indicated by total fatty acid content in Table 1) and a higher content of n-3 PUFA and PUFA:SFA ratio, and a lower n-6 PUFA:n-3 PUFA ratio.

**Table 1** Fatty acid composition (least square mean ± standard error of the mean (SEM)) of LTL of lambs fed concentrate diet for different pre-slaughter durations

| Fatty acids | Duration (Days) | | | SEM | P-values |
|---|---|---|---|---|---|
| (mg/100g muscle) | 36 | 54 | 72 | | |
| C18:2n6cis | 69.9[b] | 86.1[ab] | 92.6[a] | 5.13 | 0.014 |
| C18:2c9t11 | 11.0 | 10.2 | 7.5 | 1.77 | 0.362 |
| C18:3n3 | 20.4[a] | 17.7[a] | 12.6[b] | 1.52 | 0.002 |
| C20:4n6 | 28.1[b] | 29.1[b] | 34.5[a] | 1.06 | 0.001 |
| C20:5n3 | 15.1[a] | 13.0[a] | 8.9[b] | 1.10 | <0.001 |
| C22:5n3 | 17.0[a] | 15.2[a] | 13.1[b] | 0.70 | <0.001 |
| C22:6n3 | 5.16[a] | 4.56[a] | 3.43[b] | 0.333 | 0.002 |
| Total SFA | 796 | 1082 | 1069 | 100.8 | 0.078 |
| Total MUFA | 806[a] | 1084[b] | 1147[b] | 84.3 | 0.004 |
| Total PUFA | 171 | 181 | 178.4 | 8.90 | 0.790 |
| PUFA:SFA ratio | 0.23[a] | 0.18[ab] | 0.17[b] | 0.014 | 0.020 |
| n-6 fatty acids | 112[b] | 129[ab] | 139[a] | 6.6 | 0.024 |
| n-3 fatty acids | 57.7[a] | 50.5[a] | 38.1[b] | 2.88 | <0.001 |
| n-6:n-3 ratio | 1.97[c] | 2.59[b] | 3.68[c] | 0.119 | <0.001 |
| Total fatty acids | 1728[b] | 2301[a] | 2348[a] | 187.6 | 0.009 |

[a,b,c] Mean values bearing different superscripts are significantly different ($P < 0.05$).

**Conclusion** An IMFA composition more favourable to human nutrition can be achieved by a combined long grazing time at pasture and a short pre-slaughter duration on a cereal-concentrate.

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**References**

Aurousseau, B., Bauchart, D., Faure, X., Galot, A.L., Prache, S., Micol, D. and Priolo, A. 2007. Meat Science 76, 241-252.
Brunton, N.P., Mason, C. and Collins, M. J. Jr. 2015. Journal of Analytical Chemistry 70(10), 1218-1224.
Scerra, M., Luciano, G., Caparra, P., Foti, F., Cilione, C., Giorgi, A. and Scerra, V. 2011. Meat Science 89, 238-242.

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# Transcriptional regulation of *FYN* may orchestrate increases in the n3/n6 fatty acid ratio in the muscle of grass fed cattle

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**Application** This study has increased our knowledge on the genes that drive the increases in the n3/n6 fatty acid (FA) ratio in the *M. longissimus thoracis et lumborum* of grass-fed cattle compared to concentrate fed animals.

**Introduction** Human health advantages associated with the consumption of beef reared on pasture have been proposed, particularly relating to the supply of essential Fatty Acids (FA) (Daley, 2010). A comparison of the muscle transcriptome and lipid profile of outdoor/pasture-fed and indoor/concentrate-fed cattle resulted in the identification of 26 differentially expressed (DE) genes, the expression of which correlated most significantly with the n3/n6 FA ratios in the muscle (Sweeney *et al.*, 2016). The aim of this study was to further explore the panel of DE genes in the context of putative regulators identified from in-silico analysis and literature searches and core enzymes essential for the synthesis of n/3 and n/6 FA.

**Material and methods** The 26 DE genes were evaluated in Cytoscape™ (Shannon, 2003) in the context of putative regulators (PPAR, C/EBP, SCREB and GATA) and also enzymes essential in the synthesis of n/3 and n/6 FA (FADS1 and 2 and ELOV2 and 5). Protein-Protein interaction data was imported from the Intact database (www.ebi.ac.uk/intact). The application Network analyser ™ was used to generate topographical data which was then used to generate Figure 1.

**Results** Of the 26 DE genes, seven up-regulated (*ABCA1, ACSL3, CCL14, EIF4EBP1*, *FABP5, FYN, KLF11) and* six down-regulated *(EML1, MAP7D1, MSMO1, STK40, TULP1, TPCN1)* genes were connected to the network. The enzyme Fatty acid desaturase 2 (FADS2) was also present in this network indirectly associated with the up-regulated gene *ACSL3*. Of the DE genes *FYN* (up-regulated in grass-fed animals) was highlighted within Cytoscape™ as a hub gene with extensive connectivity (Degree=216). FYN was also connected via HSP 90-beta (a molecular chaperone with an ATP binding ability) to important regulators of fatty acid metabolism; PPARγ, SREBF1 and AMPK subunit (PRKAA2) (Fig 1).

**Conclusion** This analysis broadens the perspective on the transcriptional changes observed in muscle from cattle reared on grass and highlights *FYN* as a hub gene connected to central regulators of FA metabolism.

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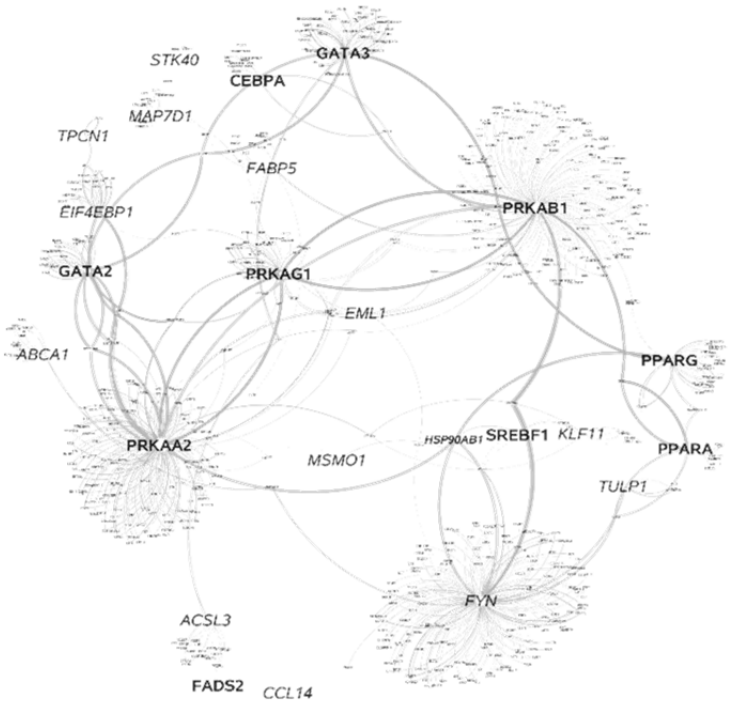


**Figure 1** PPI network, edge weighted spring embedded layout with edge thickness mapped to edge. Putative regulators and enzymes are highlighted in bold. DE genes are in plain text. *unconnected genes are excluded.

**References**

Daley, C.A., Abbott, A., Doyle, P.S., Nader, G.A. and Larson, S. 2010. Nutrition Journal 9, 10.
Sweeney, T., Lejeune, A., Moloney, A. P., Monahan, F. J., Gettigan, P. M., Downey, G., Ryan, M.T. 2016. BMC Genomics 17, 746.
Shannon, P., Markiel, A., Ozier, O., Baliga, N.S., Wang, J.T., Ramage, D., Amin, N., Schwikowski, B., Ideker, T. 2003. Genome Research 13(11), 2498-2504.

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# Effects of farm-scale grassland management strategies on nutritional quality of pasture-reared beef

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**Application** Through better understanding of the causal relationship between whole-farm grassland management strategies and meat quality, this study contributes to the formation of price premia for beef produced under grazing livestock systems.

**Introduction** Although it is widely acknowledged that the nutritional quality beef is affected by the amount of cereals used in the feed ration, impacts of different pasture types are less understood. Using a whole-farm dataset from the North Wyke Farm Platform (NWFP) linking grassland management, animal performance and meat quality, this study compares fatty acid composition and vitamin E content of beef produced on three common pasture systems in the UK: permanent pasture (PP); semi-organic white clover/ perennial ryegrass sward (WC) and high sugar perennial ryegrass monoculture (HS).

**Material and methods** At the NWFP, 30 Charolais x Hereford-Friesian steers are allocated annually to each of the three NWFP systems under the repeated measures design (Orr *et al*., 2016). Following finishing, muscle tissue (*M. longissimus)* was collected from a subsample ($n = 5$ per system per year) of steers over two seasons (2016 and 2017). Fatty acid profiles of meat were assessed using a simplified bimethylation extraction method (Lee *et al*., 2012), while forage fatty acid content was determined using direct saponification (Demirel *et al*., 2004). Both sets of extracts were measured using GC. Vitamin E content of meat and forage was measured using HPLC following hexane extraction. Two-way ANOVA was carried out using GenStat to investigate the effects of both the year and the treatment (system) on meat quality parameters.

**Results** In 2016, average daily gains for PP, WC and HS steers were 0.84, 0.72 and 0.70 kg/day, respectively. In 2017, WC cattle grew faster (0.82) than PP (0.74) and HS (0.79). These rankings were consistent with the corresponding rankings for cold carcass weights at slaughter. While no treatment effect was detected on vitamin E contents of pasture ($p = 0.541$), a significant year effect was observed ($p < 0.001$), with PP recording the largest vitamin E values in 2016 and WC in 2017. These results, however, did not completely translate into vitamin E content of meat (Table 1). Amongst key parameters derived from fatty acid profiles of beef samples, long-chain PUFA (EPA and DHA) were more abundant in PP cattle than WC and HS counterparts. In addition, n-6:n-3 ratios showed significant differences with regards to treatments, years and their interactions, with the value for PP the lowest across two years. Finally, significantly higher levels of total fatty acid contents were detected in the 2017 pasture samples compared to the 2016 samples ($p = 0.018$); this shift was also evident from total fatty acid contents of beef under all systems, although the latter differences were not statistically significant.

**Table 1** Vitamin E and fatty acid profiles of beef for each year and treatment. All values are reported as mg/kg fresh meat

| | 2016 | | | 2017 | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | PP | WC | HS | PP | WC | HS | SED | P (trt) | P (yr) | P (trt.yr) |
| Vitamin E | 5.72 | 4.67 | 4.92 | 4.88 | 4.26 | 5.54 | 0.283 | <0.001 | 0.21 | 0.004 |
| Total FA | 2680 | 2385 | 2458 | 2781 | 2707 | 2666 | 446.5 | 0.812 | 0.423 | 0.940 |
| C16:0 | 625 | 592 | 573 | 617 | 594 | 577 | 115.0 | 0.850 | 0.992 | 0.996 |
| C18:0 | 366 | 305 | 382 | 359 | 332 | 370 | 55.5 | 0.328 | 0.942 | 0.864 |
| C18:1 trans | 58.5 | 40.8 | 55.1 | 68.5 | 48.7 | 65.6 | 14.43 | 0.166 | 0.267 | 0.990 |
| C18:1 cis-9 | 977 | 817 | 821 | 988 | 1025 | 960 | 180.3 | 0.766 | 0.264 | 0.737 |
| CLA c9t11 | 13.9 | 11.3 | 11.2 | 20.4 | 15.2 | 17.3 | 4.14 | 0.396 | 0.030 | 0.901 |
| EPA[a] + DHA[b] | 20.4 | 18.5 | 19.8 | 19.7 | 17.2 | 18.2 | 1.16 | 0.043 | 0.093 | 0.847 |
| PUFA:SFA | 0.075 | 0.085 | 0.075 | 0.094 | 0.102 | 0.086 | 0.0100 | 0.202 | 0.012 | 0.834 |
| n-6:n-3 | 0.910 | 1.10 | 0.930 | 1.10 | 1.15 | 1.18 | 0.0330 | <.001 | <.001 | <.001 |

[a]Eicosapentaenoic acid, [b]Docosahexaenoic acid

**Conclusion** Significant treatment effects on n-6:n-3 ratios found in this study are likely to have human health implications, particularly when further comparisons are made with concentrate finished beef with higher levels of omega-6. As WC and HS pastures were reseeded between 2013-2015, the present results may suggest that pasture establishment affects meat quality in the first year post-reseeding.

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**References**

Demirel G., Wachira, A.M., Sinclair, L.A. *et al*. 2004. British Journal of Nutrition 91, 551-565
Lee, M.R.F., Tweed, J.K.S., Kim, E.J. and Scollan, N.D. 2012. Meat Science 92, 863-866
Orr, R.J., Murray, P.J., Eyles, C.J., Blackwell, M.S.A. *et al*. 2016. European Journal of Soil Science 67, 374-385.

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# Effect of breed and sex on meat quality attributes of Irish male lamb

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**Application** Minor differences in meat composition, tenderness and colour appearance lead to the conclusion that castration of lambs in order to improve meat acceptability and marketability is of little advantage.

**Introduction** Castration of lambs has become less frequent in Ireland in recent years given the superior production performance of ram lambs compared to wethers. There is, however, a perception regarding meat quality attributes of ram lambs. This study was one of the first to examine instrumental meat quality attributes under Irish conditions and aimed to assess the effect of sex on these instrumental attributes.

**Material and methods** A total of 200 spring born male lambs were assigned to a 2 × 2 factorial arrangement with two breeds Scottish Blackface (SB) (n=100) and Texel cross Scottish Blackface (TXSB) (n=100) and two sexes (wether (n=100) and ram (n=100)). Lambs were identified on commercial farms at birth and each alternate male lamb born alive was castrated using a scrotal rubber ring within 48 h of birth. Lambs were purchased for the experiment at approximately 5 months of age and transported to the research centre. Lambs were individually housed on expanded metal feeding pens (182 cm × 122 cm) for a 36 day indoor finishing period. Lambs were allowed a 12 day acclimatization period to adapt to a 95% concentrate diet. For the duration of the finishing period lambs were offered 100 g/day DM of silage and had access to *ad libitum* concentrate. The concentrate used was a 60% cereal-based lamb ration with 15% crude protein and an energy value of 1UFL/kg fresh weight. Following completion of the finishing period lambs were slaughtered and muscle pH and temperature measurements were taken at 1, 3, 5 and 25h post slaughter. After 25h the *M. Longissimus Thoracis et Llumborum* (LTL) was dissected form the carcass and cut into 2.5 cm steaks for further analysis. Samples were analysed for meat colour, chemical composition, cooking loss and tenderness. Tenderness was determined using the Warner Bratzler Shear Force (WBSF) method. Data diagnostics were carried out using the PROC UNIVARIATE of (SAS, version 9.4). Data analysis was carried using the MIXED procedure. The model included fixed effects of breed, sex as well as appropriate interactions with lamb considered as the random effect.

**Results** There were no interactions between breed and sex, therefore the main effects of breed and sex are presented. Wether lambs carcasses had higher conformation score ($P < 0.05$) and higher fat cover ($P < 0.001$) than ram lambs. Scottish Blackface lambs had higher carcass fat score ($P < 0.001$) and lower conformation score ($P < 0.001$) than TXSB lambs. Scottish Blackface lambs had greater intramuscular fat (IMF) ($P < 0.001$), while TXSB lambs had greater cooking loss ($P < 0.05$). A tendency for lower WBSF values in SB ($P < 0.10$) and wether lambs ($P < 0.10$) compared to TXSB and ram lambs, respectively. For colour measurements, Lightness (L*) and yellowness (b*) values were lower ($P < 0.001$) in SB than TXSB lambs. Redness (a*) and B* were higher in ram lambs than in wether lambs.

**Table 1** Carcass and meat quality attributes of Scottish Blackface (SB) and Texel cross Scottish Blackface (TXSB) ram and wether lambs

| | Breed | | Sex | | | P Value | |
|---|---|---|---|---|---|---|---|
| | SB | TXSB | Ram | Wether | SEM | Breed | Sex |
| Carcass Conformation[1] | 2.63 | 3.38 | 2.92 | 3.10 | 0.068 | <0.05 | <0.05 |
| Carcass fat score[2] | 3.77 | 3.27 | 3.07 | 3.91 | 0.077 | <0.001 | <0.001 |
| Carcass weight, kg | 20.71 | 25.74 | 23.14 | 23.31 | 0.20 | <0.001 | NS |
| Intramuscular fat | 3.28 | 2.52 | 2.68 | 3.19 | 0.098 | <0.001 | NS |
| WBSF (N)[3] | 34.15 | 37.20 | 37.10 | 34.20 | 1.530 | <0.10 | <0.10 |
| Cooking loss (%) | 27.55 | 30.35 | 29.21 | 28.69 | 0.624 | <0.05 | NS |
| Total Collagen (g/kg) | 3.23 | 2.68 | 2.94 | 2.96 | 0.091 | <0.05 | NS |
| Colour measurement | | | | | | | |
| L* | 44.02 | 45.27 | 44.72 | 44.58 | 0.221 | <0.001 | NS |
| a* | 19.29 | 19.77 | 18.14 | 20.13 | 0.250 | NS | <0.001 |
| b* | 6.74 | 7.05 | 6.73 | 7.06 | 0.129 | <0.001 | <0.001 |

[1]Carcass Conformation EUROP Scale transformed to 5, 4, 3, 2 and 1, respectively, [2] Carcass fat score=1 to 5 scale (1=low fat cover, 5=excess fat tissue), [3] WBSF= Warner Bratzler Shear Force, Colour measurements: L* = lightness, 0(black) to 100 (white); a* = redness, +a (red) to –a (green); b* = yellowness, +b (yellow) to –b (blue)

**Conclusion** Differences were found in carcass and meat quality attributes between ram and wether lambs. However, both sexes produced meat which would be deemed acceptable by consumers. Given superior production performance of ram lambs shown by Claffey *et al*., (2017), coupled with the minimal differences in meat quality attributes reported in this study there seems to be little advantage to castrating lambs to improve meat quality, though castration may still be required as a management tool in some systems.

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**References**

Claffey, N.A., Fahey, A.G. and Diskin, M.G. 2017. Advances in Animal Biosciences 8, 101.

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# Identification of novel regulatory polymorphisms in the promoter region of differentially expressed genes related to carcass and meat quality traits in lamb

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**Application** The regulatory SNPs identified in the promoter region of four differentially expressed genes will enable insight into the molecular mechanism of differential gene expression that is fundamental to variation in ovine carcass and meat quality.

**Introduction** Improvement of the lamb carcass and meat quality will enhance the industry's capacity to meet consumer needs. Genetic variation exists for a variety of meat quality traits including carcass fatness, conformation and intramuscular fat content. Single nucleotide polymorphisms (SNPs) in the promoter region of genes have been associated with gene expression and meat quality phenotypes (Sweeney *et al*., 2015). However, the traits that affect meat quality can be measured only after slaughter and require expensive and time-consuming procedures. Therefore, the identification of regulatory genetic polymorphisms robustly associated with carcass and meat quality traits will enhance genetic gain in the breeding programme. The differentially expressed genes for fatness (*LPL, MMP1* and *MSTN*) examined were reported earlier (Alam *et al*., 2017). The aim of this experiment was to identify SNPs in the promoter region of the previously identified differentially expressed genes.

**Material and methods** From 64 pairs of twin lambs, eight pairs of the most divergent twins (High or Low) were selected based on fatness. The promoter regions of the candidate genes (differentially expressed between twins) were sequenced in forward and reverse directions. SNPs were identified by multiple sequence alignment, using Molecular Evolutionary Genetics Analysis (MEGA) software.

**Results** The number of SNPs identified by *in silico* analysis in the genes and affected transcription factors are summarised in Table 1. The *in silico* analysis of the promoter region of *MSTN* gene identified CCAAT/enhancer binding protein alpha (C/EPBα) binding motif where the single nucleotide polymorphism changed C/EPBα motif into Mating-type M-specific polypeptide Mc (mat1-Mc) motif. In the *MMP1* promoter, one SNP leads to the loss of the TFIID binding motif. The transcription-factor binding motif for histone gene transcription factor (H4TF-1) is changed to the signal transducer and activator of transcription 5 (STAT5) motif due to the occurrence of one SNP in the motif.

**Table 1** Summary of genes, number of SNPs identified and affected transcription factor

| Gene (Gene ID) | Product length (bp) | No. of SNPs identified | No. of transcription factors affected by SNP | Transcription factor affected by SNPs |
|---|---|---|---|---|
| Myostatin *(MSTN*, 443449*)* | 1182 | 6 | 1 | CCAAT/enhancer binding protein alpha |
| Matrix metallopeptidase 1 *(MMP1*, 101120355) | 974 | 10 | 1 | TFIID |
| Low density lipoprotein receptor *(LDLR*, 443535) | 1394 | 9 | 1 | H4TF-1 |
| Lipoprotein lipase *(LPL,* 443408) | 1029 | 6 | | |

**Conclusion** The identified novel regulatory polymorphisms in the cis-regulatory binding site of the promoter, especially those affecting the transcription factor binding sites and those in the enhancer and repressor regions, have the potential to contribute to our understanding of the underlying molecular mechanisms of differentially expressed genes. The association study between novel SNPs and carcass and meat quality traits is in progress.

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**References**

Alam, T., Campos, V., Hanrahan, J.P., Allen, P., Keady, T., Hamill, R.M. and Sweeney, T. 2017. In Proceedings 63rd International congress of meat science and Technology, 13-18th August 2017, Cork, Ireland, 75.
Sweeney, T., O'Halloran, A., Hamill, R.M., Davey, G., Gil, M., Southwood, O., and Ryan, M. 2015. Meat Science 100, 32–40.

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# The impact of three contrasting dairy cow management strategies on the vitamin D content of milk

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**Application** Housed dairy cows offered either fresh grass or grass silage produce milk with lower concentrations of vitamin D than grazing cows. This highlights the need to develop strategies to increase vitamin D concentrations in the milk of housed cows.

**Introduction** Humans obtain Vitamin D through endogenous dermal synthesis upon exposure to UVB light, from naturally occurring food sources, and from supplements and fortified food products. However, Vitamin D deficiency is well recognised as a public health issue, with recent research (Cashman *et al*., 2016) suggesting that 40.4% of the EU population is Vitamin D deficient. While oily fish and eggs represent good naturally occurring sources of Vitamin D, these are not widely consumed. In addition, people living at more northerly latitudes have limited Vitamin D synthesis for a considerable part of the year. Consequently, there is interest in increasing the Vitamin D content of more commonly consumed foods, such as milk. This study investigates the effect of dairy cow management regime on Vitamin D concentrations in milk.

**Material and methods** Three management strategies were examined in a 24-week (April – September), continuous design experiment involving 114 Holstein Friesian dairy cows (38 cows/strategy), as follows: i. housed and offered grass silage (Silage), ii. housed and offered fresh grass (Zero-grazing), iii. full-time grazing (Grazing). Concentrate feed levels were common across all three treatments (8.0 and 6.0 kg/day for multiparous and primiparous cows, respectively). Cows within the Silage and Zero-grazing treatments were housed in cubicle accommodation. These cows were offered either fresh silage or fresh herbage once daily. Cows on treatment Grazing were managed within a rotational paddock grazing system, and given access to fresh herbage daily. On two occasions during the experiments (6 July and 20 September) milk samples were taken from all cows during two consecutive milkings, bulked in proportion to milk yield, and the samples subsequently analysed for vitamin D3, vitamin D2, 25-hydroxyvitamin D3 (25-(OH)-D3) and 25-hydroxyvitamin D2 (25-(OH)-D2). Data were analysed with linear mixed model methodology using the REML estimation method. A factorial arrangement of management and date were fitted as fixed effects with cow fitted as a random effect.

**Results** Both management strategy and sampling date had a significant effect on vitamin D3, D2 and 25-(OH)-D3 concentrations (P<0.001). Compared to Silage and Zero grazing, milk from Grazing had higher mean concentrations of vitamin D3 and its metabolite 25-(OH)-D3 on both sampling dates, and higher vitamin D2 concentrations on the first sampling date. Management strategy x date interactions were observed for vitamins D3 and D2 (P<0.001) but not 25-(OH)-D3. Concentrations of 25-(OH)-D2 were below the detection limit in all samples.

**Table 1** Effect of management strategy on milk production and the Vitamin D content of milk during the two measurement periods

| | Date | Treatment | | | SEM | Significance | | |
|---|---|---|---|---|---|---|---|---|
| | | Zerograzing | Silage | Grazing | | Management (M) | Date (D) | M x D |
| Vitamin D3 | 6 July | 0.037$^{a}$ | 0.040$^{a}$ | 0.203$^{c}$ | 0.0094 | <0.001 | <0.001 | <0.001 |
| (µg/kg) | 20 Sep | 0.041$^{a}$ | 0.036$^{a}$ | 0.088$^{b}$ | | | | |
| Vitamin 25-(OH) | 6 July | 0.058$^{a}$ | 0.058$^{a}$ | 0.094$^{d}$ | 0.0036 | <0.001 | <0.001 | NS |
| D3 (µg/kg) | 20 Sep | 0.048$^{b}$ | 0.049$^{b}$ | 0.082$^{c}$ | | | | |
| Vitamin D2 | 6 July | 0.021$^{ab}$ | 0.022$^{ab}$ | 0.046$^{c}$ | 0.0024 | <0.001 | <0.001 | <0.001 |
| (µg/kg) | 20 Sep | 0.026$^{b}$ | 0.018$^{a}$ | 0.018$^{a}$ | | | | |

Values within each variable which do not share the same superscript are significantly different (p<0.05) as determined by Fishers LSD test.

**Conclusion** Housed cows, irrespective of diet, produced milk with lower concentrations of Vitamin D3 and 25-(OH)-D3 than grazing cows. With grazing cows, Vitamin D concentrations were significantly lower in September than July.

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**References**

Cashman, K.D., Kiely, M., Kinsella, M., Durazo-Arvizu, R.A., Tian, L., Zhang, Y., Lucey, A., Flynn, A., Gibney, M.J., Vesper, H.W. *et al*. 2016. American Journal for Clinical Nutrition 103, 1033–44.

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# Multi-year evaluation of the effect of stocking rate and breed on milk production per hectare within intensive grass-based production systems

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**Application** Increasing stocking rate (SR) resulted in greater grass production and utilisation, and consequently, milk production per hectare (ha). Furthermore, Jersey x Holstein-Friesian (JxHF) cows produced more milk solids (MS) compared with Holstein-Friesian (HF) cows.

**Introduction** Successful grazing systems are dependent on achieving a balance between competing objectives of high grass intake and milk production per cow, and increasing grazing intensity to maximise grass utilisation and milk production per ha (McCarthy *et al*., 2014). A combination of cows capable of achieving high intakes of grazed grass and SR capable of maximising grass utilisation are critical to overall systematic performance. The objective of the experiment was to quantify the effect of SR and breed on milk production per ha within intensive grass-based production systems.

**Material and methods** A total of 533 lactation records from 246 high genetic merit spring calving dairy cow (68 HF and 71 JxHF, respectively) were available over a 4 year period. Cows from each breed were randomly assigned to one of three SR treatments defined in terms of body weight (BW) per ha; low (LSR; 1,200 kg BW/ha), medium (MSR; 1,400 kg BW/ha) and high (HSR; 1,600 kg BW/ha). Milk yield was recorded daily and milk constituents weekly. Body weight was measured fortnightly. Different grazing intensities were imposed on each SR, with target post-grazing residual heights of 4.5-5.0, 4.0-4.5, and 3.5-4.0 cm for LSR, MSR, and HSR, respectively. The effect of SR, breed and their interactions on milk production per cow and per ha, BW, and grazing characteristics were analysed using mixed models (PROC MIXED) in SAS.

**Results** Milk and MS yield per cow was greatest for LSR (5,309 and 459 kg, respectively), intermediate for MSR (5,030 and 432 kg, respectively), and lowest for HSR (4,862 and 414 kg, respectively). Conversely, milk and MS yield per ha was greatest for HSR (16,340 and 1,387 kg, respectively), intermediate for MSR (14,866 and 1,275 kg, respectively), and lowest for LSR (12,934 and 1,126 kg, respectively). Milk production efficiency (MS per kg BW) decreased as SR increased. Milk yield was greatest for HF cows (5,233 kg) compared with JxHF (4,900 kg), while MS was greater for JxHF cows (439 kg) compared with HF cows (430 kg). Jersey x Holstein-Friesian had greater milk production efficiency compared with their HF counterparts.

| | Holstein-Friesian | | | Jersey x Holstein-Friesian | | | | Significance | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | LSR | MSR | HSR | LSR | MSR | HSR | S.E.M. | SR | BR | SR*BR |
| Per cow | | | | | | | | | | |
| Milk yield (kg) | 5,531 | 5,158 | 5,011 | 5,086 | 4,902 | 4,712 | 62.0 | <0.001 | <0.001 | 0.266 |
| MS yield (kg) | 458 | 425 | 408 | 459 | 433 | 419 | 4.9 | 0.034 | <0.001 | 0.365 |
| Per ha | | | | | | | | | | |
| Milk yield (kg) | 13,170 | 14,920 | 16,257 | 12,698 | 14,812 | 16,153 | 184.0 | <0.001 | 0.042 | 0.566 |
| MS yield (kg) | 1,097 | 1,226 | 1,346 | 1,155 | 1,324 | 1,429 | 15.6 | <0.001 | <0.001 | 0.408 |
| MS yield (kg/kg BW) | 0.90 | 0.82 | 0.80 | 0.95 | 0.88 | 0.85 | 0.030 | <0.001 | <0.001 | 0.859 |

**Conclusion** The greater productivity per ha demonstrated at higher SR resulted from increasing BW per ha, increased grazing intensity, and higher grass utilisation. At similar BW per ha, JxHF cows produced more MS per cow, and consequently, achieved greater milk production per ha.

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**References**

McCarthy, J., McCarthy, B., Horan, K.M., Pierce, N., Galvin, A., Brennan, A. and Delaby, L. 2014. Journal of Dairy Science 97, 1693-1706.

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# The effect of cow genotype on milk production and fertility performance in a spring milk production system

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**Application** Cow genotype had a significant effect on total milk and milk solids yield and milk composition. Twenty-four day submission rate, pregnancy rate to first service and six week in calf rate were significantly different amongst genotype.

**Introduction** Declining reproductive performance in the dairy herd has become an increasingly difficult issue in the past few decades. As a result, there has been increased interest in crossbreeding, particularly in pasture-based milk production systems (Buckley *et al*., 2014). The aim of this study was to assess the milk production and fertility performance of three genotypes within a spring calving milk production system; Holstein-Friesian (HF), Jersey x HF (JEX) and a three way cross of Norwegian Red x Jersey x HF (3WAY).

**Material and methods** Data from 360 cows (120 in 2014, 2015 and 2016, respectively; 40 of each genotype each year) was available for analysis. Cows were managed within one of four grazing treatments (tetraploid-only, diploid-only, tetraploid-white clover and diploid-white clover), stocked at 2.75 cows/ha, with equal numbers of each genotype (n = 10) across each grazing treatment. Within breed, cows were assigned to treatment based on calving date, parity and economic breeding index (EBI). Milk yield was recorded daily and milk composition was analysed weekly by taking milk samples from a consecutive evening and morning milking. Fertility performance was monitored by recording all artificial inseminations and natural services throughout the breeding season and ultrasound scanning to determine pregnancy status when the breeding season concluded. Milk data were analysed using Proc MIXED and fertility data using Proc LOGISTIC in SAS (SAS 9.4). Cow genotype, grazing treatment, parity and year were included as fixed effects in the model.

**Results** The effect of cow genotype on milk production and fertility performance is presented in Table 1. Holstein-Friesian had the greatest milk yield, JEX were intermediate and 3WAY lowest. Milk solids yield was similar for HF and JEX and for HF and 3WAY, whereas JEX had a greater milk solids yield than 3WAY. Milk composition (fat and protein %) was greater for JEX and 3WAY (4.76% and 3.79% and 4.71% and 3.80% for JEX and 3WAY, respectively compared with HF (4.43% and 3.64%). The fertility performance amongst genotype was not significantly different in terms of overall in calf rate. Twenty-four day submission rate was lower for JEX compared with HF and 3WAY. Differences also existed amongst genotype for pregnancy rate to first service and six week in calf rate, with JEX having a greater pregnancy rate to first service compared with HF and 3WAY and the JEX and HF having a greater six week in calf rate compared with 3WAY (Table 1).

**Table 1** Cow genotype effect on milk production and fertility performance 2014-2016

| | HF | JEX | 3WAY | SE | *P*-value |
|---|---|---|---|---|---|
| Milk yield (kg) | 5656$^{a}$ | 5452$^{b}$ | 5277$^{c}$ | 51.9 | <0.0001 |
| Milk solids (kg) | 455$^{a}$ | 466$^{a,b}$ | 448$^{a,c}$ | 4.0 | 0.009 |
| 24-day submission rate (%) | 97.5$^{a}$ | 91.6$^{b}$ | 97.4$^{a}$ | | 0.040 |
| Pregnancy to 1$^{st}$ service (%) | 64.4$^{a}$ | 78.2$^{b}$ | 61.5$^{a}$ | | 0.008 |
| Six week in calf rate (%) | 88.1$^{a}$ | 89.9$^{a}$ | 83.8$^{b}$ | | 0.041 |
| In calf rate (%) | 95.8 | 95.0 | 93.2 | | NS |

**Conclusion** The similar performance of the HF and JEX in terms of milk solids yield and fertility was in contrast with previous research (Prendiville *et al*., 2011) but indicates the suitability of both for spring calving pasture-based milk production systems. And although the 3WAY had lower milk solids yield than JEX and lower fertility performance than HF and JEX, their performance was adequate for this system.

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**References**

Buckley, F., Lopez-Villalobos, N. and Heins, B.J. 2014. Animal 8, s1, 122-133.
Prendiville, R., Shalloo, L., Pierce, K.M. and Buckley, F. 2011. Irish Journal of Agricultural and Food Research 50, 123–140.

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**078**

# Incorporating international and national breeding values for the UK Limousin beef breed

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**Application** Increased reliabilities for UK Limousin beef breeding values through the incorporation of international proofs and the inclusion of overseas progeny information. The breeders benefit from the phenotypes recorded in other countries.

**Introduction** United Kingdom is an active member of the International Evaluations for beef and dairy cattle (Interbeef and Interbull). In the Interbeef evaluations data from nine countries are used in order to calculate the Estimated Breeding Values (EBVs) for weaning weight (AWW). Until now, none of the nine countries were using combined international and national proofs in their breeding programmes. The aim of this project was to develop the mechanism of incorporating international AWW EBV into the national evaluation and evaluate its impact on the reliabilities of the EBVs of animals widely used abroad.

**Material and methods** Three times per year UK send the pedigree and performance records for the Limousin beef breed, which are used along with the data provided by Czech Republic, Denmark, Finland, France, Germany, Ireland, Sweden and Switzerland. As a result of this international evaluation, 199,183 records return to the UK for national (180,194) and foreign (18,989) Limousin animals. Currently international proofs are produced for one trait only (weaning weight), but research to analyse more traits is ongoing. In the UK national evaluation, a multi-trait model is implemented and account is taken of the genetic correlations of AWW with many other traits like birth weight, fat/muscle depth or calving ease. In order to incorporate the international proofs for AWW, the impact on correlated traits should also be accounted for. Initially combined EBVs for AWW were computed using the formula published by Mrode *et al*., (1996), then combined EBVs were calculated for all the correlated with AWW traits, using the following formula:

$$\begin{pmatrix} \widehat{U_1} \\ \widehat{U_2} \\ \vdots \\ \widehat{U_n} \end{pmatrix} = \begin{pmatrix} g_{1d} & g_{1m} \\ g_{2d} & g_{2m} \\ \vdots & \vdots \\ g_{nd} & g_{nm} \end{pmatrix} \begin{pmatrix} g_{dd} & g_{dm} \\ g_{md} & g_{mm} \end{pmatrix}^{-1} \begin{pmatrix} \widehat{U_d} \\ \widehat{U_m} \end{pmatrix}$$

where $U_1$ to $U_n$ are UK traits, correlated with AWW, $g_{1d}$ to $g_{nd}$ are the genetic covariances between direct AWW and adequate trait, $g_{1m}$ to $g_{nm}$ are the covariances between maternal AWW and adequate trait, $g_{dd}$, $g_{md}$ and $g_{mm}$ are the direct variance, covariance and maternal variance for AWW, respectively. $U_d$ and $U_m$ are the differences of combined and UK national EBVs for direct and maternal AWW.
The reliabilities (RD) for the combined EBVs were computed using Mrode *et al*., (1996) formula:

$$RD = \frac{1}{(1 - UK\ rel)} + \frac{1}{(1 - ITB\ rel)}$$

The increases in the reliabilities of correlated traits with AWW were calculated as the difference between combined and UK reliabilities for AWW multiplied by the square of the genetic correlation between the given trait and AWW.
International proofs both for the UK and foreign animals were rebased to the same base as for the UK national evaluation before the combination procedure.

**Results** Correlations between obtained blended AWW EBVs and UK national EBVs are 0.89 and 0.68 for direct and maternal AWW, respectively. This indicates undoubtedly that the international values resulted in re-ranking of animals. Average increase of reliability for direct AWW is 0.13, with the change range from 0 up to 0.83. Very high change is observed for the animals widely used abroad, with many progeny records overseas but with few or no progeny in the UK. Results obtained for maternal AWW show the average increase of reliabilities of 0.13, with the range from 0 to 0.76.
The correlations of combined and UK national breeding values for the correlated with AWW traits ranges between 0.97 and 1.00, with reliability correlations 0.86 to 1.00 (lowest values are for strongly correlated with AWW 400-day weight).

**Conclusion** Until now, it was not possible to directly compare the results of the UK national evaluation with the international proofs calculated by Interbeef. This method improves the reliabilities and allows a direct comparison between UK and foreign animals.

**Acknowledgements** The authors gratefully acknowledge British Limousin Cattle Society for the use of the data.

**References**

Mrode, R.A., Swanson, G.J.T. and Winters, M.S. 1996. Animal Science 63 (3), 373-379.

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079

# Immune-associated traits measured in milk as proxies for blood serum measurements in dairy cows

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**Application** Using routinely collected milk samples as a proxy for blood offers a low-cost non-invasive approach to collect valuable immune-associated data to help improve animal welfare and provide an indication of the animal's health status.

**Introduction** Previous work highlighted that immune-associated (IA) traits measurable in blood are associated with health, productivity and reproduction in dairy cows (Eckersall *et al*., 2006; Banos *et al*., 2013; Denholm *et al*., 2017). Similar relationships have been observed in IA traits from milk (van Knegsel *et al*., 2007; Ploegaert *et al*., 2010; de Klerk *et al*., 2015). Focusing on natural antibodies (NAb), haptoglobin (Hp) and tumour necrosis factor alpha (TNF-α), the present study analysed longitudinal IA data in order to determine whether routinely collected milk samples could provide a less invasive method of obtaining an immune profile relevant to dairy cow health and productivity, thus reducing the requirement for blood sampling.

**Material and methods** Data were collected from 546 Holstein-Friesian dairy cows. All cows were from the experimental Langhill herd based at the SRUC Dairy Research Centre, and were involved in an on-going selection experiment for genetic line x feeding system study (Pollott and Coffey, 2008). Concurrent milk and serum samples were collected at 15 time points (5,354 total samples) between April 2013 and March 2015 and included summer and winter samplings. Whole blood was collected into plain Vacutainers (BD) and allowed to coagulate before centrifugation at 2,000 × g for 10 min. Milk was centrifuged at 3,000 × g for 30 minutes and the skimmed milk fraction was retained. All samples were stored at -20°C prior to analysis and analysed with ELISA for natural antibodies binding keyhole limpet hemocyanin ($NAb_{KLH}$) and lipopolysaccharide ($NAb_{LPS}$), Hp and TNF-α. Univariate and multivariate analyses were carried out using repeated measures mixed linear animal models with a pedigree relationship matrix fitted to account for the genetic relationships between animals.

**Results** Analyses revealed significant ($P<0.01$) positive phenotypic correlations between milk and serum measures of NAb ($0.59 \leq R \leq 0.75$), Hp ($R=0.37$) and TNF-α ($R=0.12$). Milk and serum NAb were also found to have a strong genetic correlation ($0.77 \leq R \leq 0.94$, $P<0.01$) and were both genetically correlated with cow lameness (0.66 and 0.79 for milk $NAb_{KLH}$ and serum $NAb_{LPS}$ respectively) and other cellular immune measures such as %lymphocytes, %PBMC, %neutrophils and %eosinophils. Clinical mastitis was found to be phenotypically correlated with both milk and serum Hp ($0.09 \leq R \leq 0.23$). Serum Hp was also strongly genetically correlated with other cellular IA traits such as $\%NKp46^+$ ($R=0.35$) and %PBMC ($R=-0.90$). Similarly, genetic correlations were found to exist between serum TNF-α and $\%NKp46^+$ ($R=0.22$), %PBMC ($R=0.41$) and %lymphocytes ($R=0.47$). All milk and serum IA traits were repeatable ($P<0.05$) within animal ranging from 0.09 (serum Hp) to 0.47 (serum $NAb_{LPS}$). Between-animal variation was highest in milk and serum NAb (0.36 – 0.47) and significant estimates of heritability were also observed in milk and serum NAb (0.17 – 0.37).

**Conclusion** Outcomes from the present study suggest that IA traits present in the milk of dairy cows are heritable, repeatable and have the potential to describe IA profiles in the blood, especially in the case of NAb. The strong genetic correlations found between the milk and serum NAb suggests there is potential for using NAb in the milk as a marker for NAb in the blood. Furthermore, the relationships found between NAb, Hp and TNF-α with both health and cellular IA traits are promising and warrant further study. Presently, milk samples are routinely collected for milk recording purposes, and hence, offer a less invasive and cost-effective way to sample and collect valuable and informative IA trait data for use in a variety of areas of predictive modelling.

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**References**

Banos, G., Wall, E., Coffey, M.P. *et al*. 2013. PLoS One. 8, e65766.
Denholm, S.J., McNeilly, T.N., Banos, G. *et al*. 2017. Journal of Dairy Science 100, 2850–2862.
Eckersall, P.D., Young, F.J., Nolan, A.M. *et al*. 2006. Journal of Dairy Science 89, 1488–1501.
de Klerk, B., Ducro, B.J., Heuven, H.C.M. *et al*. 2015. Journal of Dairy Science 98, 2746–2752.
van Knegsel, A.T.M., de Vries Reilingh, G., Meulenberg. S. *et al*. 2007. Journal of Dairy Science 90, 5490–8.
Ploegaert, T.C.W., Wijga, S., Tijhaar, E. *et al*. 2010. Journal of Dairy Science 93, 5467–73.
Pollott, G.E. and Coffey, M.P. 2008. Journal of Dairy Science 91, 3649–3660.

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**080**

# A meta-analysis of genome wide association data of residual feed intake and related traits in Irish beef cattle

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**Application** Identification of DNA based biomarkers for residual feed intake (RFI) and related traits could contribute to genomic selection of feed efficient animals as well as aid in understanding the underlying biology of feed efficiency (FE).

**Introduction** FE is a measure of how well an animal converts feed to produce, i.e. milk or meat. FE is an economically important trait as improvement in FE allows an animal to maintain production levels, while consuming less feed, thus improving profitability. There are several metrics by which FE can be measured including RFI. RFI is independent of production traits and thus an attractive measure of FE (Arthur and Herd, 2008). RFI is not widely adopted, as it is an expensive trait to measure. The inclusion of genetic variants associated with RFI in the Irish national genomic selection breeding programme would improve RFI in the national herd. This study aims to examine genome wide associations between single nucleotide polymorphisms (SNPs) and RFI and its component traits in Irish beef cattle.

**Material and methods** 1,684 Irish animals were genotyped using the custom International Dairy and Beef (IDB) version three SNP chip (Mullen *et al.*, 2013). A further 338 animals were genotyped on the Illumina Bovine HD genotyping chip and imputed to IDBv3 density using FImpute version 2.2. All genotypic data was uploaded to Golden Helix version 8.7.1. Data underwent QC analysis within the SNP Variation Suite environment and linkage disequilibrium (LD) pruning was carried out. Genotypic data was then merged with collated phenotypic data on these animals which had undergone feeding trials and for which RFI was calculated. Following merging of phenotypic and genotypic data files, 1,492 animals were eligible for analysis (Aberdeen Angus n = 102, Belgian Blue n = 177, Charolais n = 387, Limousin n = 537, Simmental n = 289).
Genome-wide association studies (GWAS) for each trait of interest per breed were carried out using a mixed linear model. This resulted in the generation of 5 sets of GWAS results for each of the three traits of interest, i.e., RFI, average daily gain (ADG), and daily feed intake. Following GWAS, meta-analysis was carried out to analyse marker effects per trait across all breeds using METAL (Willer *et al.*, 2010). Ingenuity Pathway Analysis (IPA) was performed on SNPs reaching nominal significance.

**Results** Markers that had P-values less than $5x10^{-5}$ were deemed to have genome wide association significance as per the Wellcome Trust significance threshold. Most significant markers for each trait are represented in Table 1.

**Table 1** Top 2 most significant SNPs for RFI or related traits

| rsID | P-value | Trait | Chromosome_Megabase | Nearest Gene |
|---|---|---|---|---|
| rs386023985 | $4.32x10^{-11}$ | ADG | 19_49 | *ERN1* |
| rs135897656 | $5.83x10^{-10}$ | ADG | 3_119 | *CSF2RA* |
| rs55617218 | $1.33x10^{-5}$ | Feed intake | 19_14 | *HNF1B* |
| rs109691080 | $2.61x10^{-5}$ | Feed intake | 1_6 | *MAP3K7CL* |
| rs43555985 | $8.28x10^{-6}$ | RFI | 8_69 | *GFRA2* |
| rs41638273 | $1.08x10^{-5}$ | RFI | 2_6 | *SLC40A1* |

IPA identified pathways such as growth hormone signalling and cell cycle regulation as playing a role in the divergence of RFI, ADG, and feed intake in this cohort.

**Conclusion** Following validation, the SNPs identified via association studies in this study may be included in the Irish national breeding program as markers for RFI and related traits. The genes and pathways identified as implicated in the divergence of the traits of interest will be further investigated to enhance our understanding of the biology underpinning FE in beef cattle.

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**References**

Arthur, J.P. and Herd, R. 2008. Revista Brasileira de Zootecnia 37, 269-279.
Mullen, M.P., McClure, M.C., Kearney, J.F., Waters, S.M., Weld, R., Flynn, P., Creevey, C.J., Cromie, A.R. and Berry, D.P. 2013. Interbull, August 23-25 2013, Nantes, France. Bulletin No.47
Willer, C.J., Li, Y. and Abecasis, G.R. 2010. Bioinformatics 26, 2190-2191.

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**081**

# Vetnomics: a comprehensive online course upskilling practising vets in the contemporary field of genomics in animal breeding

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**Application** Development of an online course designed to upskill practicing vets in the field of genomics in animal breeding, encapsulating years of research, delivering confidence in scientific evidence and at the acquisition of CPD points.

**Introduction** The increasing potential and applicability of genomics in livestock breeding (Womack, 2005; Haley & de Koning, 2006; Goddard, 2012) has simultaneously highlighted the need for training to level the field with regards to genomic knowledge across stakeholders, retain confidence in emerging technologies and optimise their use. Vets serve as a fundamental, trustworthy interface with farmers. Through the Vetnomics course, practicing vets can revise and improve their knowledge of genetics and genomics in animal breeding, complementing and developing the advice they can offer farmers and keeping up in this fast moving area.
This project is in collaboration with partners including; XLVet Training Services (who will ultimately package and deliver the training modules), AHDB (contributing knowledge and experience from their key role in national genetic and genomic evaluations) and Edinburgh Genetic Evaluation Services (EGENES; routinely handling UK-wide performance and pedigree data for farmed animals and producing UK genomic breeding values). Despite the target audience being vets, their clients, interested farmers and breeding companies could all derive benefit from accessing such a course and further improve across-stakeholder communication. The overall aim of this project was to deliver a comprehensive overview of the field of genomics, to complement veterinary training and satisfy their need to develop their progressive knowledge as obligatory under the RCVS Code of Professional Conduct.

**Material and methods** Initially the course syllabus was derived drawing on years of expertise, experience and research in the field of animal breeding from SRUC and EGENES. Lectures were subsequently planned, scripted, recorded and edited using consistent lecture format and structure, regular definitions and further reading opportunities. Lectures were designed to be stimulating, digestible and ultimately for delivery via the web. Despite the course forming a continuous guide through genomics, each lecture serves as a stand-alone guide to each subject area facilitated by an initial overview of any essential prior knowledge required to optimise its understanding. Lectures were delivered by experts in each area and filmed to include the person speaking where appropriate. This novel presentation method utilises audio visual equipment provided under an Agrimetrics UK grant.

**Results** Through a series of 17 core c. 40 minute lectures the constructed course guides the audience through the relevant theory and extensive practicalities of genomics in livestock, with modules covering everything from good practices in the process of tissue sampling to the value of big data, from the importance of phenotype recording to available breeding values for previously difficult to measure traits. The course essentially lays the biological foundations for genetics, guides the audience through relevant theory and provides strong emphasis on the application of genomics in practice across a whole range of farmed species, elaborating on terminology likely experienced in the field, approaches to best interpret data and maximise on the services offered to farmers. The key learning outcomes of this course are 1) To understand and give confidence in the current applications of genomics in animal breeding, 2) To understand the information required in utilising genomic information and how to get the most benefit from it in animal breeding and 3) To understand how best to interpret genomic information and facilitate on farm management and breeding decisions.

**Conclusion** The Vetnomics course encompasses years of research from SRUC Animal breeding and Genomics and EGENES and is dedicated to delivering a comprehensive guide through genomics to its target audience. The authors have scheduled discussions with numerous vets and stakeholders to review course content and ensure revisions are implemented prior to the course release. This release is scheduled early in 2018.

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**References**

Haley, C. and de Koning, D.J. 2006. Animal Genetics 37, Suppl. 1, 10–12.
Womack, J.E. 2005. Genome Research 15, 1699–1705.
Goddard, M.E. 2012. Animal Production Science 52. 73–77.

**082**

# Comparative performance of Holstein-Friesian dairy cows of contrasting economic breeding index

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**Application** Validates the impact of Ireland's dairy breeding objective, Economic Breeding Index (EBI), on improving performance

**Introduction** Genetic selection of dairy cattle in Ireland is based on total merit selection using the EBI (Veerkamp *et al*., 2002). Monitoring phenotypic trends in traits is prudent to ensure the continued suitability of a particular breeding objective. The objective was to evaluate the milk production, body weight, body condition score and fertility performance of two genotypes of Holstein-Friesian of divergent genetic merit based on EBI, across three pasture-based feeding treatments.

**Material and methods** Two genotypes of Holstein-Friesian based on the Irish total merit index, EBI, were evaluated across four years (2013–2016). In each year, 90 genotyped cows representative of the top 1% (ELITE; mean EBI €154; Milk sub-index €37, Fertility sub-index €80, (ICBF, May 2017)and 45 cows representative of the mean nationally (NatAv; mean EBI €51; Milk SI €17, Fertility SI €13, ICBF, May 2017) based on their genomic EBI proof were evaluated. In year 1 all animals were primiparous and by year 4 20% parity 1, 20% parity 2, 27% parity 3 and 33% parity 4 animals were present in each genotype. Mean calving date was February 15 (±16 d) and 18 (±18 d) for ELITE and NatAv cows, respectively. By mid-March of each year, cows were randomly assigned to one of 3 feeding treatments feeding treatments based on parity, calving date and pre-experimental yield of milk solids (mean of 2 weeks). Control (CTL), Low Grass Allowance (LGA), and High Concentrate (HC) treatments, had target post-grazing residual sward heights (PGRSH) of 4.5-5, 3.5-4, and 4.5-5cm respectively, and total concentrate allowances of 300, 300 and 1200kg per cow per lactation respectively. Parity structure and calving date were maintained similar for each treatment group. Weekly milk production was established from daily recording (morning and evening) while milk fat and protein concentrations were determined weekly from one successive p.m. and a.m. milk samples. Bodyweight of each animal was recorded weekly using a calibrated electronic scale (Dairymaster, Causeway,Co.Kerry). Body condition score (BCS) was measured fortnightly for the first ten weeks of lactation, and monthly thereafter, on a 1 to 5 scale. Reproductive efficiency indicators investigated included SR24; proportion of cows submitted in the first 24 days of the breeding season, PREG1; proportion of cows pregnant to first service, PREG6W; proportion of cows pregnant in the first 6 weeks of the breeding season and PREG12W; proportion of cows pregnant after the 12 week breeding season. All pregnancy variables are based on ultrasonic imaging at day 150. Analysis of reproductive efficiency indicators, and mean BCS, body weight (BW) and milk production variables was conducted using a repeated measures model in PROC GENMOD or PROC MIXED of SAS (SAS Institute, 2017), respectively. Genotype, feeding treatment, calving day of year and parity were included as fixed effects. Cow nested within genotype was included as a random effect while year was included as a repeated effect. Interactions between genotype, feeding treatment and parity were also investigated.

**Results** Mean lactation length was 284 and 283 days in the Elite and NatAv, respectively. Milk volume (5384kg vs. 5601 kg) was lower ($P<0.01$) but milk fat (44.7g/kg vs. 42.0g/kg) was higher ($P<0.01$) and milk protein content (37.2g/kg vs. 35.4g/kg) was higher ($P<0.001$) with ELITE compared to NatAv. Milk solids (fat plus protein) yield tended to be higher with ELITE cows (441 kg vs. 432 kg; $P=0.06$). Elite cows tended to be lighter (507kg vs. 515kg; $P=0.08$) and maintained higher BCS (2.92 vs. 2.74; $P<0.001$) throughout lactation. Three week submission rate (92% vs. 86%; $P<0.05$), pregnancy rate to first service (0.6 vs. 0.46; $P<0.001$), six-week in-calf rate (0.73 vs. 0.58; $P<0.001$) and 12-week in-calf rate (0.92 vs. 0.81; $P<0.001$) were higher for ELITE compared with NA. No genotype × feeding treatment interaction was observed for any of the traits.

**Conclusion** The results provide confidence that genetic selection based on EBI delivers more productive (higher value milk) and more fertile dairy cows. In addition the study provides leadership, that the decline in fertility evidenced in the Holstein-Friesian population caused by aggressive selection for milk yield, may be reversed when appropriate selection pressure on suitable fertility traits is applied, while simultaneously improving milk solids potential.

**References**

Veerkamp, R.F., Dillon, P., Kelly, E., Cromie, A.R. and Groen, A.F. 2002. Livestock Production Science. 76**,** 137-151.
ICBF. May 2017. Irish Cattle Breeding Federation (ICBF) 2017 [Online].
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**083**

# Genome wide association study of health related traits in dairy and beef cattle on Irish commercial farms

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**Application** Calves are born with a predetermined genetic potential. SNPs associated with enhanced immunity in beef and dairy calves could be applied in a genomic selection breeding programme to breed for improved health traits in cattle.

**Introduction** Genome-wide association studies (GWAS) using single nucleotide polymorphism (SNP) markers provide a powerful approach for mapping QTL of important health traits on the genome (Thompson-Crispi *et al*., 2014). Here, health related data was collected for dairy and beef calves on commercial farms in Ireland over the first 6 months of life and associated with SNP data using GWAS, to analyse genetic predisposal towards superior or inferior immunity.

**Material and methods** In total, 1,392 suckler (n = 111 farms) and 2,090 dairy (n = 84 farms) calves were born between July 2014 – June 2016 and followed until 6 mo. of age. Blood samples collected from calves with associated morbidity and mortality information were genotyped using the v3 IDB SNP chip (Mullen *et al*., 2013), which included 260 immune SNPs added from literature (e.g. Fisher *et al*., 2011). A total of 2400 calves with appropriate genotyping data and phenotypic records were carried forward for GWAS. Quality control was carried out whereby SNPs were removed from analysis if they had a call rate of less than 0.80 or a minor allele frequency of less than 0.05. Animals were removed from analysis if they had a call rate of less than 0.95. Following QC, 2,400 animals and 36,664 markers remained for analysis. Further filtering resulted in 7 main breeds for analysis: Angus (n=273), Belgian Blue (n=63), Charolais (n=384), Holstein Fresian (n=1,047), Hereford (n=112), Limousin (n=368) and Simmental (n=116). GWAS were carried out in Golden Helix using a mixed linear model method, EMMAX with gender of calves defined as a co-variate. This mixed model can be summarised as: $y=X\beta+Zu+\varepsilon$, where y is an n x 1 vector of the observed phenotypes (IgG serum concentration, BRIX reading, ZST reading, Total protein, Pneumonia, Scour, All diseases, Other diseases, Navel infection, Joint illness), X is an n x f matrix of fixed effects and β is a f x 1 vector which represents the coefficients of fixed effects, while Z is an n x t matrix relating the random effect to the specified phenotype.

**Table 1** SNPs associated with incidence of scour during the first 6 month of life

| Calf Breed | Gene | Marker | Ch[1] | Position | P-Value | Minor Allele Frequency |
|---|---|---|---|---|---|---|
| Aberdeen Angus | *CADM2* | HAPMAP58994-RS29021251 | 1 | 32738897 | 5.78E-07 | 0.018315018 |
| Belgian blue | *PARD3B* | ARS-BFGL-NGS-81996 | 2 | 119772030 | 4.66E-09 | 0.150793651 |
| | *PAX3* | UA-IFASA-5029 | 2 | 111206088 | 3.96E-07 | 0.095238095 |
| | No Info | ARS-BFGL-NGS-108811 | 2 | 119668743 | 8.07E-07 | 0.214285714 |
| | *CAB39* | HAPMAP54936-RS29013679 | 2 | 119387551 | 5.08E-06 | 0.222222222 |
| | No Info | HAPMAP41038-BTA-62514 | 27 | 23957015 | 7.17E-06 | 0.182539683 |
| Charolais | LOC104976682 | ARS-BFGL-NGS-86309 | 22 | 30988035 | 1.83E-06 | 0.122395833 |
| Hereford | No Info | HAPMAP49389-BTA-96192 | 7 | 106170981 | 3.16E-07 | 0.03125 |
| | *RORA* | ARS-BFGL-NGS-111546 | 10 | 49480741 | 1.45E-06 | 0.397321429 |
| | No Info | ARS-BFGL-NGS-10713 | 7 | 76021523 | 4.05E-06 | 0.205357143 |
| Simmental | No Info | HAPMAP41388-BTA-95947 | 8 | 3470459 | 5.10E-07 | 0.017241379 |
| | *OLA1* | HAPMAP33359-BTA-133718 | 2 | 22604508 | 1.73E-06 | 0.004310345 |
| | *ADNCR* | BTA-24463-NO-RS | 13 | 22021716 | 5.77E-06 | 0.012931034 |
| | No Info | ARS-BFGL-NGS-115663 | 1 | 11743590 | 5.06E-06 | 0.034482759 |
| | LOC107131301 | BTB-00871695 | 23 | 43535918 | 1.62E-05 | 0.081896552 |

A total of 413 significant SNPs were identified across the 7 breeds, at a cutoff value of $P <= 5x10^{5}$, associated with overall disease occurrence, scour, pneumonia, joint disease, navel infection, and total IgG level. Table 1 lists the SNPs associated with incidence of scour during first 6 months of life. A polymorphism in the *PAX3* gene was identified as being associated with scour and pneumonia in Belgian Blue calves. A significant SNP in the *CAB39* gene was also identified in Belgian Blue calves to be associated with incidence of scour. This gene encodes a component of a complex that binds and activates *STK11/LKB1*, a serine threonine kinase complex involved in B-cell differentiation.

**Conclusion** A large number of significant SNPs were identified across the 7 breeds. Although none of the research SNPs added to the IDBv3 SNP chip by our group were identified as significant, a number of other research SNPs on the IDB chip were identified as associated with health traits. An example includes a SNP in *RBM26* that encodes an RNA binding protein thought to play a role in innate immune response. These SNPs are particularly important for further mining for the genetic basis of immune response development and health traits in cattle and used for trait prediction across the breeds to identify possible genetic determinants of predisposal for disease traits such as pneumonia in beef and dairy cattle.

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**References**

Fisher, C.A., Bhattarai, E.K., Osterstock, J.B., Dowd, S.E., Seabury, P.M. *et al*., 2011. PloS one, 6(11), p.e27744.
Mullen, M.P., McClure, M.C., Kearney, J.F., Waters, S.M., Weld, R., Flynn, P., Creevey, C.J., Cromie, A.R., Berry, D.P. 2013. Interbull Bulletin, 47.
Thompson-Crispi, K.A., Sargolzaei, M., Ventura, R., Abo-Ismail, M., Miglior, F., Schenkel, F., Mallard, B.A. 2014. BMC Genomics.

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**084**

# Mathematical growth functions to model live-weight in sheep

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**Application** Successful application of growth functions to sheep live-weight data will facilitate the development of animal growth prediction tools for farmers.

**Introduction** In sheep production systems lamb days to slaughter is determined via age and growth rate while replacement females must reach a target live-weight before mating. Animal growth is defined as the relationship between age and the lifetime live-weight of an animal (Fitzhugh, 1976). Mathematical growth functions have the ability to condense the weight-age relationship into biologically interpretable parameters. The objective of the current study was to compare alternative growth functions fitted to sheep live-weight data with particular emphasis on the ability to predict future live-weight.

**Material and methods** A total of 831,031 live-weight records from 246,157 ovine animals in 4,497 Irish flocks between the years of 2011 and 2017, inclusive, were obtained from the national database. Live-weight is recorded at specific time-points including: birth, forty days and weaning as lambs and again as ewes, usually on an annual basis. Animals 550 days of age were deemed to have reached their asymptotic live-weight; only animals with a minimum of five live-weight records, including one within the first seven days of life and one after 550 days were retained for analysis. The final dataset consisted of 47,525 live-weight records from 6,347 females in 289 sheep flocks. The growth functions of von Bertalanffy, Gompertz, Richards, Logistic and Brody were fitted to the live-weight data of each animal individually. Each growth function included a dependent variable ($Y_t$) representing the observed weight of the animal at t days of age and three predicted parameter estimates *A* ((kg) represents asymptotic mature weight), *B* (the constant of integration), *K* ((kg/d per kg mature weight) represents the maturing rate), (and *m* (represents the point of inflection in relation to *A*) for Richards). The goodness to fit for each growth function to the data was quantified using the $R^2$ and the root mean square error (RMSE) statistic. To test the accuracy of each model to predict future live-weight, live-weight data post 100 days of age in 25% of the animals were masked. Three models were then used to predict the masked weights. The $R^2$ and the RMSE between the predicted and actual weight of the masked live-weights were used to determine accuracy of prediction for each model.

**Results** Only animals that converged with biologically sensible parameters for each growth function were retained for the goodness of fit calculations. Twenty-one animals failed to converge for the Richards function and were therefore omitted. The population mean values (SD in parenthesis) of the model parameters varied by growth function; mean values for parameter *A* ranged from 67.90-69.43kg (13.70-14.20kg) for the von Bertalanffy, Gompertz and Richards growth functions. Mean parameter *k* values ranged from 6-10g/d per kg mature weight (2-6g/d per kg mature weight). Summary statistics of the goodness to fit for each model to the live-weight data in the initial exploratory dataset are presented in Table 1. The mean $R^2$ and RMSE values ranged from 0.83 to 0.98 and 3.33 kg to 9.76 kg, respectively. The Gompertz growth function predicted the masked weights least accurately with an $R^2$ = 0.20. The Richards fixed effect model had the largest $R^2$ between actual and predicted weights of 0.70 with an RMSE of 19.29kg. This indicated that it was the best model for the prediction of futuristic live-weights.

**Table 1** Growth functions for the exploratory dataset with corresponding $R^2$ and RMSE values

| Function | $R^2$ | RMSE (kg) |
|---|---|---|
| von Bertalanffy | 0.98 | 3.33 |
| Gompertz | 0.97 | 3.60 |
| Richards | 0.89 | 9.76 |
| Logistic | 0.96 | 4.27 |
| Brody | 0.83 | 9.32 |

**Conclusion** Results show the prediction of sheep live-weight is extremely promising in striving to produce growth prediction tools for farmers, with the Richards growth function having the highest accuracy at predicting futuristic live-weight.

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**References**

Fitzhugh, H.A, 1976. Animal Science 42, 1036–1051.

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**085**

# The genetic basis of pneumonic lesions and pleurisy in New Zealand lambs

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**Application** High density genotyping revealed several regions of the sheep genome associated with either pneumonic lesions or pleurisy in New Zealand (NZ) lambs, which included genes involved in the immune response.

**Introduction** Pneumonia is an important issue for sheep production in NZ and internationally, leading to reduced growth rate and a predisposition to pleurisy (Alley, 2002). Previous work has established that the heritability of pneumonic lesions at slaughter in NZ mixed breed progeny tested lambs is 0.07 ± 0.02 (McRae *et al*., 2015). Genomics, through tools such as genome-wide association studies, can also be used to further increase our understanding of the genetic mechanisms underlying the host response to disease, and compare these mechanisms between breeds or species. Discovering regions of the genome associated with susceptibility may also lead to development of new diagnostic tools and alternative treatments. The objective of this study was to identify loci associated with pneumonic lesions and pleurisy in NZ progeny test flocks.

**Material and methods** The lungs from 7,994 progeny-test lambs were scored for presence and severity of pneumonic lesions and pleurisy at slaughter using the methodology of McRae *et al*., 2015. Before analysis both pneumonic lesion score and pleurisy values were scaled by the incidence rate per contemporary group (flock, birth year, sex, weaning mob and slaughter date). Data analysis models for both traits included contemporary group as a fixed effect. A subset of animals (N = 3,488) were genotyped using the Illumina Ovine Infinium® HD SNP BeadChip (606,006 markers). The heritability of lung lesion score and pleurisy were calculated using the genomic relationship matrix, and genome-wide association analyses were conducted using Efficient Mixed-Model Association eXpedited (EMMAX) using identity-by-state (IBS) and haplotype trend regression (HTR) with a three-SNP sliding window.

**Results** At slaughter, 31% of lambs had pneumonic lesions, with more than half of an individual lobe affected in 9% of lambs. The incidence of pleurisy was 8%. Heritability estimates for pneumonic lesions and pleurisy were 0.16 (± 0.03) and 0.05 (± 0.02), respectively. Five SNPs were significantly associated with pneumonic lesions at the genome-wide level (Table 1), and an additional 31 SNP were suggestively significant. Several SNPs that reached suggestive significance ($p<1.86x10^{-6}$) were in regions that contained genes involved in processes including DNA repair and the innate immune response including *EYA4, ATAD5* and *RFC4*. Both EMMAX and HTR analyses of pleurisy data showed a significant peak on chromosome 2 (Table 1), located downstream from the transcription factor *SP3*. SP3 activates or supresses the expression of numerous genes, including several genes with known functions in the immune system.

**Table 1** SNPs significantly ($p <9.31x10^{-8}$) associated with consolidated pneumonia score and pleurisy in NZ lambs

| Phenotype | HTR P-Value | EMMAX P-value | Chr | Position | RSID |
|---|---|---|---|---|---|
| Pneumonia | 4.75E-08 | n.s. | 3 | 197,825,391 | rs424070250 |
| | 6.45E-08 | n.s. | 6 | 77,843,695 | rs399606595 |
| | 1.63E-09 | n.s. | 8 | 7,733,798 | rs429357466 |
| | 7.65E-09 | n.s. | 8 | 7,743,164 | rs400905064 |
| | 3.36E-08 | n.s. | 13 | 8,848,881 | rs417728121 |
| Pleurisy | 1.53E-08 | 3.10E-09 | 2 | 134,984,962 | rs398681238 |
| | n.s. | 2.32E-08 | 2 | 134,985,148 | rs424471052 |
| | 7.66E-08 | n.s. | 2 | 134,979,525 | rs428634189 |

**Conclusion** At slaughter, a third of lambs examined showed evidence of pneumonic lesions in the lung. High density genotyping revealed several regions associated with both pneumonic lesions and pleurisy in NZ lambs, which included genes involved in the immune response. One region associated with pleurisy was identified using two independent methods. These results support the hypothesis that the genomic basis of disease resistance is multifaceted, which is consistent with the complex aetiology of the disease. Furthermore, the identification of SNPs associated with both pneumonic lesions and pleurisy furthers our understanding of the genetic mechanisms underlying the host response to respiratory disease in sheep.

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**References**

Alley, M.R. 2002. New Zealand Veterinary Journal 50, 99-101.
McRae, K.M., Baird, H.J., Dodds, K.G., Bixley, M.J. and Clarke, S.M. 2016. Small Ruminant Research 145, 136-141.

086

# An investigation in to the relationship between ewe body condition score change, ewe breed, and litter size of flocks enrolled in the Irish Central Progeny Test programme

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**Application** The effect of BCS change on ewe performance differs between breeds and is influenced by ewe age, mating body condition score (BCS) and progeny breed.

**Introduction** Monitoring changes in ewe body reserve status using body condition score allows for the subjective assessment of individual animal and flock nutritional status without the need for sophisticated feed intake or blood metabolite measurements. The effect of BCS and BCS change during the production cycle on flock performance has been extensively discussed in the literature (Kenyon *et al*., 2011). However, research results on the effect of ewe BCS on flock performance are not always consistent (Kenyon *et al*., 2014). The objective of this study was to investigate if an interaction exists between ewe BCS change and ewe breed and litter size. This aims to explain some of the inconsistencies reported in the literature on the effect of ewe BCS on flock performance and provide information on the drivers of ewe BCS change during the production cycle through multivariate regression analysis.

**Material and methods** Flock performance and BCS measurements were collected from two commercial and one research flock comprising of approximately 1500 ewes in total. The data set was compiled over three production seasons from October 2012 through to July 2015 and all flocks were involved in the Sheep Ireland Central Progeny Test programme. Ewe BCS was assessed twice during year one of the study at mating and weaning (approximately day 100 post-partum) and four times during the following two seasons; mating, mid-gestation (approximately day 85 of gestation), mid-lactation (approximately day 40 of lactation) and weaning. Ewe BCS was assessed on a scale of one to five (Jefferies, 1961) in 0.1 interludes. All ewes were mated in October of each production year following oestrus synchronisation using intervaginal progestogen pessaries followed by an intramuscular injection of 500 i.u. PMSG. All farms involved operate grass based production systems whereby sheep grazed perennial ryegrass (Lolium perenne) based swards for approximately nine months of the year. Ewes in each flock were housed and offered a grass silage based diet with concentrate supplementation during the final two months of gestation and returned to grass within one week of parturition. Ewe litter size was recorded at parturition. Belclare, Suffolk and Texel ewes used were crosses of their breed. The breed type 'Terminal' refers to the combination of data from Charollais (176 ewes), Rouge (37 ewes) and Vendeen (211 ewes) ewes. These data were analysed using a generalised least square means procedure in SAS version 9.4 following tests for the assumptions of normality.

**Results** As indicated by a multivariate regression analysis year of study ($P = 0.01$), ewe breed ($P = 0.01$), ewe age ($P = 0.01$), litter size ($P = 0.01$), mating BCS ($P = 0.01$), farm ($P = 0.02$) and progeny breed ($P = 0.02$) all influenced the change in ewe BCS from mating to weaning. Ewes with a litter size of 1 at parturition gained BCS from mating to mid-gestation whereas ewes with a litter size of 2 or 3+ at parturition lost BCS during this period ($P = 0.01$). When all ewe breeds were considered together, ewes with an average litter size of 2.0 and 2.5+ gained more BCS from weaning to mating than ewes with and average litter size of 1 and 1.5 ($P = 0.01$). When these data were disaggregated by breed there was no difference in BCS change from weaning to mating and average litter size for Belclare ewes ($P = 0.74$) but there were differences in Suffolk ($P = 0.02$), Terminal ($P = 0.01$) and Texel ($P = 0.03$) ewes. The effect of BCS change on ewe performance differs between breeds and between ewes of different ages. This was particularly evident in the interaction between ewe breed type, litter size and BCS change from mating to weaning.

**Conclusion** In conclusion the farm, year of study, ewe breed, progeny breed, litter size, ewe age and mating BCS will all influence the BCS change of the ewe from mating to weaning. The effect of BCS change on ewe performance differs between ewe breed types and litter size. The increase in BCS from weaning to mating in ewes with an average litter size of 2.0 and 2.5+ may be a trigger for ewes from some breeds having a higher average litter size. However, the average litter size of the maternal breed within the data set appeared unaffected by BCS change at this time. Further analysis is warranted to investigate how the interaction between ewe breed and BCS change effects other flock performance parameters such as weaning weights.

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**References**

Jefferies, B. 1961. Tasmanian Journal of Agriculture 32, 19-21.
Kenyon, P.R., Maloney, S.K. and Blache, D. 2014. New Zealand Journal of Agricultural Research 57(1), 38-64.
Kenyon, P.R., Morris, S.T., Stafford, K.J. and West, D.M. 2011. Animal Production Science 51, 557-564.

087

# Concentrations of adiponectin, leptin, ghrelin and resistin in goat colostrum and mature milk from seven breeds

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**Application** The present study found that adiponectin, leptin, resistin and ghrelin hormones are secreted biologically in goat colostrum and milk. Further studies will assess their relationships with fat metabolism and immune system development.

**Introduction** The importance of passive or humoral immunity, through absorption of colostral antibodies or immunoglobulin is well recognised. White adipose tissue, considered a passive site of lipid storage, is reported to have a role in metabolic and endocrine functions; secreting a range of hormones known as adipokines such as adiponectin, leptin, ghrelin and resistin (Hussein *et al.*, 2015). In addition, these major adipokines have been found in human milk and in some livestock species (Guzel *et al*., 2017). There is a paucity of literature regarding hormones that may be associated with energy metabolism in goats, thus the objective of this study was to measure the concentration of adiponectin, leptin, ghrelin and resistin in goat colostrum and mature milk from seven different breeds.

**Material and methods** Seventy colostrum and milk samples were obtained from seven goat breeds (n=10 goats per breed); (Majorera (MAJ), Palmera (PAL), Tinerfeña (TNF), Del Guadarrama (GU), Florida (FL), Payoya (PY) and Verata (VE)). The goats' diet was fed according to INRA recommendations and was balanced for energy and protein levels. Colostrum samples (50 mL) were collected immediately post-partum (PP) and milk samples (50 mL) were collected on day 30 PP. Each sample was divided into four aliquots. All aliquots were preserved by freezing at -20°C until analysis. Hormonal concentrations were determined in skimmed colostrum and milk. Skimming was performed by centrifugation at 4600 rpm, 10 minutes, 4°C. Colostrum and milk hormone concentrations were measured using commercially available ELISA kits (Leptin and adiponectin (Cusabio Biotech kits); Resistin and ghrelin (MyBioSource kits). Samples were analysed in duplicate. Statistical analysis was performed using SAS, Version 9.4 (SAS Institute Inc., Cary, NC). The PROC MIXED procedure of SAS with repeated measures was used to evaluate the concentration of hormones in colostrum and milk.

**Results** A breed effect was found for all hormones except milk ghrelin concentration (Table 1). Leptin concentration was greater in milk, and in colostrum, for the GU and PY, respectively, with no differences between colostrum and milk concentration for the other breeds. Adiponectin concentration was greater in colostrum than in milk for all breeds except for the PY breed. Ghrelin concentrations were greater in colostrum than in milk for the PAL, TNF and VE breeds. Resistin concentration was greater in milk than in colostrum for GU, PY and VE breeds.

**Table 1** Leptin, adiponectin, ghrelin and resistin concentration in colostrum and milk (day 30 PP) from seven goat breeds[1]

| | Leptin (ng/ml) | | | Adiponectin (µg/ml) | | | Ghrelin (ng/ml) | | | Resistin (ng/ml) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Breed | C | M | SEM | C | M | SEM | C | M | SEM | C | M | SEM |
| MAJ | $0.83^{a}$ | $0.07^{a}$ | 0.29 | $73.80^{x,b}$ | $11.06^{y,b}$ | 8.20 | $0.10^{a}$ | 0.07 | 0.01 | $26.99^{ab}$ | $23.68^{ab}$ | 1.60 |
| PAL | $0.47^{a}$ | $0.10^{a}$ | 0.14 | $82.20^{x,b}$ | $11.02^{y,b}$ | 2.59 | $0.10^{x,a}$ | $0.06^{y}$ | 0.01 | $18.36^{a}$ | $24.68^{abc}$ | 2.53 |
| TNF | $1.17^{a}$ | $1.19^{b}$ | 0.07 | $175.71^{x,c}$ | $7.53^{y,ab}$ | 4.40 | $0.17^{x,ab}$ | $0.07^{y}$ | 0.01 | $41.27^{b}$ | $25.22^{abc}$ | 5.93 |
| GU | $0.98^{x,a}$ | $1.20^{y,b}$ | 0.03 | $8.38^{x,a}$ | $4.29^{y,a}$ | 1.20 | $0.14^{ab}$ | 0.16 | 0.01 | $16.77^{x,a}$ | $26.23^{y,abc}$ | 2.00 |
| FL | $4.72^{b}$ | $1.10^{b}$ | 1.37 | $123.03^{x,bc}$ | $9.27^{y,b}$ | 20.32 | $0.17^{ab}$ | 0.13 | 0.05 | $38.77^{b}$ | $35.08^{c}$ | 3.18 |
| PY | $2.22^{x,ab}$ | $1.11^{y,b}$ | 0.33 | $11.38^{a}$ | $9.29^{b}$ | 1.06 | $0.16^{ab}$ | 0.14 | 0.01 | $16.20^{x,a}$ | $23.10^{y,a}$ | 1.79 |
| VE | $0.42^{a}$ | $0.09^{a}$ | 0.12 | $79.63^{x,b}$ | $9.75^{y,b}$ | 4.86 | $0.21^{x,b}$ | $0.15^{y}$ | 0.01 | $10.78^{x,a}$ | $34.63^{y,bc}$ | 4.00 |
| SEM | 0.78 | 0.03 | | 12.30 | 0.89 | | 0.02 | 0.03 | | 3.94 | 2.57 | |

[1] n=10 goats per breed; C: Colostrum, M. Milk, SEM: Standard Error of Means, $^{x,y}$Lsmeans within a row (for each hormone) with different superscripts differ significantly ($P<0.05$), $^{a,b}$ Lsmeans within a column with different superscripts differ significantly ($P<0.05$). The values are expressed as Lsmeans (SEM).

**Conclusion** These data confirm that adiponectin, leptin, ghrelin and resistin are present in goat colostrum and milk. The function of these adipokines in colostrum will require further investigation as they may have important roles, such as energy intake and immune system development, in the neonatal kid goat.

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**References**

Guzel, S., Yibar, A., Belenli, D., Cetin, I. and Tanriverdi, M. 2017. Turkey Journal of Veterinary Medicine Science 79, 602-607.
Hussein, Y.S., Maha, A.E., Hannan, A.T. and Mogeda, K.M. 2015. Egyptian Journal of Chemistry and Environmental Health 1, 588-612.

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# The impact of maternal genetic indexes and country of origin on lambing performance through comparing New Zealand and Irish ewes

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**Application** The selection of elite genetics through the maternal genetic index will result in superior production including lambing traits, while a comparison of the maternal genetic indexes used in Ireland and New Zealand will facilitate validation of the Sheep Ireland €uro–star index.

**Introduction** Genetics are responsible for over 50% of production gains on farm. Although genetic gain is cumulative and permanent, the progress in the maternal index reported to date in Ireland has been low (€0.28/lamb/year) compared to the corresponding gains reported in New Zealand (€1.16/lamb/year) (Santos *et al*., 2015). Similar to SIL-ACE (New Zealand's national sheep index), the Sheep Ireland €uro-star indexes were introduced to aid farmers in selecting breeding animals, in an attempt to assess the benefits of careful genetic selection in Ireland. However, to date Irish and New Zealand elite ewes have not been compared in a common environment. Furthermore, the Irish replacement €uro-star index, used to select maternal type ewes in Ireland, has not been validated. The aim of the present study is to evaluate the effects of maternal genetic indexes on ewe maternal performance across three diverse strains, with specific focus given to lambing traits.

**Material and methods** The study began in October 2015 and will run for four years. Three groups of animals (n=60), comprising of two main breeds (Texel and Suffolk), were assembled; representing high genetic merit New Zealand ewes (NZ), high genetic merit Irish ewes (Elite Irish), and low genetic merit Irish ewes (Low Irish). In early October, ewes were mated using artificial insemination. Within 2 hours after lambing ewes were scored for lambing difficulty using a 1 to 4 scale (where 1 = lambed without assistance and 4 = significant assistance was required) and mothering ability on a scale of 1 to 3 (where 1 = good and 3 = poor), depending on the attentiveness of the ewe to the lamb(s) and her willingness to follow the lamb(s) immediately after birth. The number of times a ewe and her progeny required additional help after lambing was recorded as the number of visits per ewe. The total number of lambs born alive was recorded in conjunction with lamb birth-weight which was measured using a standard portable scale. Lamb mortality involved recording lambs that died in the first month of life. Data was analysed using a linear mixed model in PROC MIXED (SAS Inst. Inc., Cary, NC, USA) where breed (Texel or Suffolk), genetic strain (NZ/Elite Irish/Low Irish), sex, birth-type and year were included as fixed effects. Ewe parity was included as the repeated effect and sire of the lamb was included as a random effect in the model.

**Results** Lambing difficulty differed by strain with New Zealand ewes requiring less assistance at lambing compared to Elite Irish and Low Irish ewes ($P$=0.03; Table 1). Relative to lambs born to Elite Irish ewes, lambs born to Low Irish ewes had a greater predicted probability of dying (2.91 ± 0.614; $P$=0.07) within one month of birth. There was no significant effect of strain on lamb birth weight. To date, there has been no significant effect of strain on the number of lambs born per ewe, mothering ability and the level of assistance required by the ewe in the immediate post-partum period.

| | No. per treatment | New Zealand | Elite Irish | Low Irish | SEM | $P$ value |
|---|---|---|---|---|---|---|
| Lambing Difficulty Score | $n$=120 | 1.73[a] | 2.23[b] | 2.20[b] | 0.230 | 0.03 |
| Birth Weight (kg) | $n$=195 | 5.24 | 5.44 | 5.26 | 0.137 | 0.29 |
| No. lambs born | $n$=195 | 1.80 | 1.63 | 1.62 | 0.168 | 0.70 |
| Mothering Ability | $n$=115 | 1.40 | 1.46 | 1.30 | 0.119 | 0.14 |
| No. of visits per ewe | $n$=58 | 1.05 | 0.98 | 1.23 | 0.207 | 0.24 |

**Table 1** The effect of ewe strain (New Zealand/Elite Irish/Low Irish) across a range of lambing performance traits

**Conclusion** Results to date demonstrate that NZ ewes require less assistance at lambing when compared to both groups of Irish ewes, while greater lamb mortality was recorded in the lambs born to Low Irish ewes. However, as only two years of data have been collected and analysed thus far, it is anticipated that differences may be detected as more data accumulates.

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**References**

Santos, B., McHugh, N., Byrne, T., Berry, D. and Amer, P. 2015. Journal of Animal Breeding and Genetics 132, 144-154.

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# An analysis of the factors effecting lamb mortality using the Teagasc BETTER lowland sheep farms

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**Application** Lamb birth weight, ewe age and lamb sex are key factors influencing the level of lamb mortality. There is a large variation in the levels of mortality among the progeny of different sires within breeds

**Introduction** Globally, lamb mortality accounts for 15-20% of all lambs born, most of these deaths occur in the first day post-partum (Nowak *et al*., 2000; Nowak and Poindron, 2006), with birth trauma, hypothermia, starvation and infectious disease (Dwyer *et al*., 2016) key causations. Previous studies have indicated that traits linked to mortality rates are heritable and as such are useful when included in estimated breeding values (Southey *et al*., 2001). Recently, data from a progeny evaluation study on the Teagasc BETTER sheep farms has highlighted the potential influence of sire breed on lamb mortality rates and the variation in mortality rates of progeny between sires of the same breed. The objective of this paper is to identify the key factors within flock structure affecting lamb mortality rates and investigate the variation in lamb mortality attributable to different sires.

**Material and methods** In October 2013 and 2015 approximately 1400 commercial cross bred ewes of mixed age and breed were randomly divided into single sire mating groups across five farms and mated to pure bred Belclare, Suffolk and Texel rams. All rams were performance recorded rams selected to be either rated as being in the top or bottom 20% for terminal characteristics based on the Sheep Ireland Euro-Star Index (Pabiou *et al*., 2014). Rams were joined with ewes for 17 days before mating groups were collapsed to avoid issues arising from infertile or sub fertile rams. All lambs were born indoors and lambs born alive or dead were recorded to their dam. Ewes and lambs were gathered again at seven weeks and 14 weeks (weaning) post-partum and lamb live weights were recorded. Lamb live weight and live weight gain were adjusted to 49 and 100 days post-partum for all lambs based on live weight gain to the date of weighing. Lamb mortality was defined as lambs recorded as born dead or recorded as dead up to 14 weeks post-partum. Performance data for the three years was analysed using a generalised least square means procedure in SAS 9.4 with ewe as the experimental unit and year of study treated as a repeated measure. A logistic regression analysis was carried out to generate P values for the factors effecting lamb mortality. Descriptive data presented was calculated using frequency and means procedures.

**Results** A stepwise logistic regression indicated that lamb birth weight ($P< 0.01$), ewe age ($P<0.01$), lamb sex ($P<0.02$) and sire breed ($P<0.02$) all influenced the level of lamb mortality recorded on the farms. Birth type had no effect on lamb mortality in this model ($P>0.10$). As presented in Table 1 the level of mortality recorded varied within breed types. Sire breed did not have a significant effect on progeny weaning weights with a mean weaning weight of 31.7 kg, 32.4 kg and 32.0 kg for the Belclare, Suffolk and Texel cross lambs respectively ($P>0.51$)

**Table 1** Descriptive statistics for data used in analysis

| | No. rams used | No. progeny recorded | % Mortality | Morality range (%) |
|---|---|---|---|---|
| Belclare | 7 | 371 | 6.7 | 1.1 - 13.0 |
| Suffolk | 17 | 684 | 10.7 | 0.0 – 20.3 |
| Texel | 7 | 171 | 7.6 | 0.0 – 17.9 |

**Conclusion** The variation in lamb mortality appears to be greater within breed than between breeds. However, lamb birth weight, ewe age and lamb sex all have a significant impact on lamb mortality.

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**References**

Dwyer, C., Conington, J., Corbiere, F., Holmøy, I., Muri, K., Nowak, R., Rooke, J., Vipond, J. and Gautier, J.M. 2016. Animal 10(3), 449–459.
Nowak, R. and Poindron, P. 2006. Reproduction Nutrition Development 46, 431-446.
Nowak, R., Porter, R. H., LévY, F., Orgeur, P. and Schaal, B. 2000. Reviews of Reproduction 5, 153-163.
Pabiou, T., Byrne, T., Wall, E. and McHugh, N. Proceedings of the 10th World Congress of Genetics Applied to Livestock Production. 2014.
Southey, B.R., Rodriguez-Zas, S.L. and Leymaster, K.A., 2001. Journal of Animal Science 79(9), pp.2298-2306.

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# Drivers and barriers for adoption of Electronic Identification (EID) technology by commercial sheep farmers

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**Application** The results of the current study give us insight into what factors influence adoption of EID technology on sheep farms and can be used to develop actions to enhance uptake. Considering the upcoming legislation on mandatory EID of all cattle in the UK, these findings may also be useful to increase adoption of EID technologies among cattle farmers.

**Introduction** The UK is one of the greatest producers of lamb meat worldwide. However, sheep farming has seen a decline in profitability in past decades in the UK when compared to other livestock sectors. Increased recording of farm and flock data could contribute to the identification of farm resources used less efficiently (Kaler & Green, 2013; Redman, 2016). Technologies such as EID tools facilitate insertion and use of flock records, but anecdotal evidence suggests the uptake of these tools by sheep farmers has not been significant (Morgan-Davies & Lambe, 2015). The aims of this study were to i) investigate uptake and sheep farmers opinions about use of EID recorded information for flock management in the UK, ii) explore the association between farmers beliefs and other farmer and farm characteristics and EID adoption, and iii) analyse the association between levels of lameness on sheep farms and use of EID technology.

**Material and methods** Commercial sheep farmers from England and Wales were sent a postal questionnaire in September 2015 (n=2000). Questionnaire included questions on farm and farmer characteristics, flock husbandry practices, flock lameness levels, current use of EID tools and farmer's beliefs on the technology. Husbandry practices and farmer's opinions associated with adoption of EID technology were explored. A descriptive analysis of the results was performed, followed by exploratory factor analysis and multivariable logistic regression modelling, to identify farmer beliefs and management practices significantly associated with adoption of EID technology. Analysis was performed using Stata 14 (Statacorp, USA).

**Results** 439 farmers replied to the questionnaire, generating a response rate of 22%. Among these, 87 had used EID technology for recording flock information, 97 planned to adopt it in the future, and 222 farmers had neither adopted it nor intended to do it in the future. Three factors representing farmer's beliefs were identified via Exploratory Factor Analysis – practicality, usefulness, and external pressure and negative feelings, and all three factors were significantly associated with adoption/intention to adopt EID technology for flock management ($p \leq 0.05$). Although technology cost was considered important by both groups, adopters were significantly more likely than non-adopters to perceive EID as practical and useful ($p \leq 0.05$). In contrast, non-adopters were more likely than adopters to consider that 'government pressurise farmers to adopt technology'. Farmers with higher IT knowledge, using other technologies on farm and who intended to intensify flock production in the future were significantly more likely to adopt EID technology ($p \leq 0.05$). Moreover, flocks managed with EID tools in the previous year had significantly lower flock lameness levels ($p \leq 0.05$). Factors such type of farm (upland or lowland), farmer age or flock size seem to be less relevant for adoption of EID technology.

**Conclusion** These findings provide insights on drivers and barriers of adoption of EID tools by sheep farmers. Enhancing farmer's familiarity with technologies and communicating evidence of the beneficial effects of use of EID recording information on flock performance are likely to contribute to the uptake of this technology by sheep farmers.

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**References**

Redman, G. 2016. Melton Mowbray: Agro Business Consultants.
Kaler, J. and Green, L. 2013. Preventive Veterinary Medicine 112(3–4), 370–377.

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# Detection of lameness in sheep using different machine learning algorithms

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**Application** Lameness in sheep is a widespread problem in the UK sheep farming industry. We developed algorithms that can successfully identify lame sheep using sensor data; this monitoring system to automatically detect lameness in sheep has a huge potential to improve sheep health and welfare.

**Introduction** Lameness is a significant problem in the sheep industry, causing losses of around £80million/year. Lameness is characterised by changes in behaviour especially in the stance and gait of the animal (Kaler *et al*., 2009). Recent advances in bio-telemetry technology have made it possible to monitor and obtain a better understanding of the physiology and behaviour of wild and farm animals, including sheep. Accelerometer and gyroscope based sensors can be used to automatically detect changes associated with lameness in sheep. In the present study, we developed a machine learning algorithm using accelerometer and gyroscope sensor data to detect and classify lameness in sheep.

**Material and methods** For this study, data was collected in October 2016 using a total of 19 sheep representing a range of body condition, age and breed. Sheep were sprayed with coloured spray to facilitate identification. The locomotion score of sheep were captured in two 2h sessions on each day. A custom-made wearable device based on the Intel® Quark™ was attached to the existing electronic identification tag via a tape and lightweight plastic tie for data collection. The device had a triaxial gyroscope and accelerometer. Data was segmented in consecutive windows of 7 seconds. Different machine learning algorithms such as: random forest, neural network, support vector machine, linear regression and average perceptron were used for the classification of different lameness scores in sheep. The different machine learning algorithms were evaluated using 1 to 22 feature characteristics. A comparison of the performance of the different algorithms was obtained using the overall accuracy, precision, recall, F-score, and specificity values. An individual sheep level prediction of lameness was obtained using the ratio of windows where a sheep was classified as lame over the total number of windows in a period of time.

**Results** The best performance for the classification of lameness at the window level was obtained by the random forest algorithm with a precision ranging from 65% when using only one feature characteristic to 69% when using 7 features and 75% using all features. Recall for random forest algorithm ranges from 76% using 2 features to 79% using 8 features. The maximum precision obtained by the rest of the algorithms was below 62%. However, other algorithms did show high recall values up to 84% (i.e., averaged perceptron). The maximum F-score, which combines precision and recall metrics, was obtained by the random forest algorithm. The algorithm could significantly differentiate between lame and non-lame sheep.

**Conclusion** The results from this study show that by using machine learning algorithms combining different features from accelerometer and gyroscope based sensors, it is possible to accurately classify lameness in sheep. The evaluations of the different algorithms show that random forest algorithm is better suited for this classification. Further work is needed with respect to feature characterisation and importance.

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**References**

Kaler, J., Wassink, G.J. and Green, L.E. 2009. Vet Journal 180, 189-194.

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# Cardiovascular measures during routine practices for feed efficiency assessment in beef cattle

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**Application** Cardiovascular function monitored during rest and stress may serve as a proxy for feed efficiency in cattle.

**Introduction** Assessment of cattle feed efficiency via physiological indicators of residual feed intake (RFI; kg DM/day) (Pollak *et al*., 2012) has potential to aid in meeting increasing meat demand while lowering environmental footprint, from both breeding and management perspectives. Further investigation of the relationship between cardiovascular function against productive performance could advance its application as a proxy for RFI. Feed efficient (Low-RFI) cattle have been shown to have lower heart rates (HR) during rest, but higher HR in response to acute stress (Munro *et al*., 2016). Thus, objectives were to further assess the relationships of cardiovascular function and RFI in cattle as part of regular feedlot husbandry practices, in an ongoing effort to determine the potential of cardiovascular measures in commercial settings.

**Material and methods** Beef cattle (Cohorts A to D) were housed in facilities equipped with feed intake recording systems (Cohorts A, B and D; Insentec BV, Marknesse, The Netherlands and, Cohort C; GrowSafe Systems Ltd., Airdrie, Canada). Feed intake was recorded over 115 ± 23 days based on *ad libitum* access to total mixed rations. Productive performance was measured, on 28 day intervals, including body weight (BW) and body composition via ultrasound (back and rump fat thickness, rib eye area and marbling). Average daily dry matter intake (ADMI) was modelled across all cohorts using BW, average daily gain, body composition measures and cohort using the GLM procedure. Residual feed intake was the residual of the ADMI model calculated for each animal. Heart rate recording of the cohorts A and B began at 2200 h and stopped the following day upon transportation and stunning in the abattoir, while recording of cohort C occurred overnight, from 1600 h to 0530 h. Heart rate was averaged on 5 second intervals (Polar RC3 GPS, Polar Electro, Kempele, Finland). Average HR over the entire segment; overnight ($HR_{OVA;}$ 2200 h and 0500 h), transport ($HR_{TRA;}$ initial 20 minutes of transport; Cohort A and B only) and abattoir ($HR_{ABA;}$ initial 10 minutes of abattoir arrival; Cohort A and B only), and average of the highest ($HR_{OVH}$, $HR_{TRH}$, $HR_{ABH}$; BPM) and lowest ($HR_{OVL}$, $HR_{TRL}$, $HR_{ABL}$; BPM) twenty percent of HR were calculated for each animal within each segment. A frequency-domain analysis was completed separately on each HR segment and recording. Area under the amplitude/frequency curve from 0.01 to 0.1 Hz was calculated as an indicator of low-frequency (LF) power and sympathovagal balance (Ori *et al*., 1992) (overnight, $LF_{OV}$; transport, $LF_{TR}$; abattoir, $LF_{AB}$). All modelling and statistical analyses were completed using the SAS software. Residuals for HR were determined by the GLM procedure. Significant fixed effects and covariates were identified by the GLMSELECT procedure using the backward and BIC options. The CORR procedure determined Pearson's correlation of various traits. Partial $R^2$ was determined using the REG procedure, summed across linear and quadratic effects to determine the portion of explained variation in RFI attributed to each trait. Models, effects and correlations were deemed significant when $P \leq 0.05$.

**Results** Overnight and transport HR and LF traits were not correlated with productive performance. Positive correlations were observed between $HR_{ABH}$ and DMI (0.34), and $LF_{AB}$ and DMI (0.29), but abattoir HR traits were not correlated with RFI. The heart rate model was able to explain a significant amount of the variation in RFI ($R^2$= 0.157). Transport HR traits explained the largest portion (39 %) of the variation in RFI accounted for by the heart rate model, followed by abattoir (34 %) and overnight HR (27 %) traits (Fig. 1).

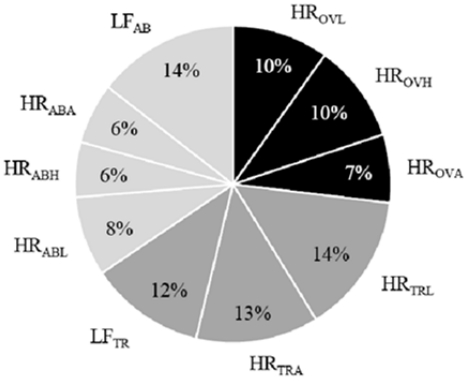


**Figure 1** Contribution of HR traits towards explained variation in RFI (light grey, abattoir HR; black, overnight HR; dark grey, transport HR).

**Conclusion** Resting HR has a positive relationship with RFI indicating a greater metabolic efficiency in feed efficient cattle, while changes in stress severity and duration with abattoir and transport HR support a more specific energy conserving stress response. Cardiovascular measures have potential to be developed as robust feed efficiency proxies for the bovine, without hindrance to performance or health.

**Acknowledgements** We thank the Elora Beef Research Centre and Maritime Beef Test Station.

**References**

Munro, J., Schenkel, F., Physick-Sheard, P., Fontoura, A., Miller, S., Tennessen, T., Montanholi, Y. 2016. Animal 11:3, 452–460.
Ori, Z., Monir, G., Weiss, J., Sayhouni, X. and Singer, D.H. 1992. Cardiology Clinics 10, 499–537.
Pollak, E.J., Bennett, G.L., Snelling, W.M., Thallman, R.M. and Kuehn, L.A. 2012. Animal Production Science 52, 92–99.

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# Beef Monitor: tracking beef cattle growth and predicting carcass characteristics of live animals

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**Application** The Beef Monitor system accurately monitored individual animal growth and predicted liveweight, cold carcass weight, saleable meat yield and EUROP classifications through passive weighing and 3D imaging of live cattle.

**Introduction** Beef producers currently assess the performance of their cattle through visual assessment or by weighing through a crush. This can lead to animals being retained on farm too long, thus not achieving the optimal market price and increasing production cost to the farmer. Video imaging analysis (VIA) is increasingly used in abattoirs to grade carcases and there is potential for 3d imaging to be used on farm to predict carcass characteristics of live animals. The Beef Monitor (BM) system combines a water trough with a non-intrusive automated weighing system and 3D camera technology to track growth and predict carcass characteristics of live animals. The objectives of this study were to validate the use of the BM system and to develop artificial neural networks (ANNs) to predict liveweight and carcass characteristics from images of live animals.

**Material and methods** To validate the accuracy of the weigh crate 39 steers were weighed weekly in a crush and these weights were related to the average of all weights recorded on the same day for each beast by the BM crate. Seven BM systems were installed on commercial and research farms in Scotland and a variety of breeds (steers and heifers) were placed behind the system for 1-3 months pre-slaughter. Images and weights were passively collected from individual animals at each visit to the trough. A further trial was conducted at the abattoir where live animals were weighed and 3D images taken immediately pre-slaughter. Cold carcass weights (CCW) were provided by the abattoir, and saleable meat yield (SMY), fat and conformation grades were determined by VIA of carcass images. ANNs were developed to predict liveweight and carcass characteristics from 40 measurements obtained from the 3D image (heights, widths and lengths) and 20 calculated areas, volumes and ratios. ANN performance was assessed by regression for liveweight, CCW and SMY, and prediction of the correct EUROP grade for fat and conformation.

**Results** The relationship between weights of 39 steers measured weekly in the crush and the average of weights measured in the BM crate on the same days had an $R^2$ of 0.99 (n = 224, Figure 1).
Predictions for all animals, including sex and breed as factors gave the following $R^2$ values: Liveweight $R^2$ = 0.72 (n = 40930), CCW $R^2$ = 0.91 (n = 1655) and SMY: $R^2$ = 0.80 (n = 1655, Figure 2).
Prediction of EUROP fat grade had 63% accuracy, and for EUROP conformation grade prediction accuracy was 69% (n = 1655).

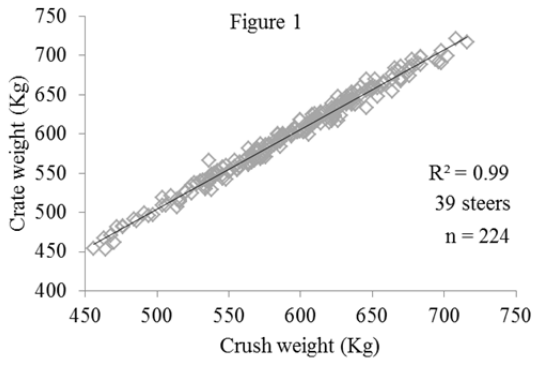


**Figure 1** Relationship between weights measured in a crush and the average weighed in the BM crate for the same day.

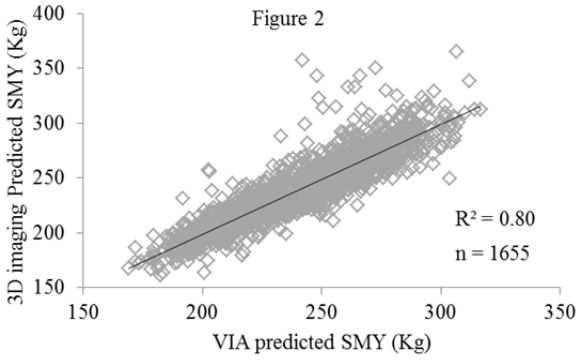


**Figure 2** Relationship between SMY predicted by VIA of carcasses and SMY predicted by 3D imaging of live animals

**Conclusion** The BM system can provide accurate weights for individual animals on a daily basis without the need for manual handling. 3D imaging of live cattle can be used to predict carcass characteristics on farm, presenting an opportunity to improve the efficiency of beef production enterprises through marketing of animals at the optimal time.

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# Effect of sampling frequency, window size and sensor positon in the classification of sheep behaviour

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**Application** Automated behavioural monitoring systems have the capacity to improve the health and welfare of farm animals. However such systems require an optimal set of parameters that can reduce energy requirements while maintaining high levels of performance.

**Introduction** Recent advances in bio-telemetry and machine learning techniques have made possible to monitor and classify different activity in wild and farm animals. A behavioural monitoring system can be potentially used to detect changes in behaviour that are associated with health and welfare changes in farm animals. However, current systems are limited by the processing power and memory on each device. Therefore, an optimisation on resources such as sampling frequency, window size and sensor position can help to overcome the limitations. Currenrly, there are no studies in precsion livestock technology that have evaluated these parameters simulatoneosly. Aim of this study was to simultaneously evaluate the effect of sampling frequency, window size and sensor position in the performance of a sheep behavioural classifier for lying, standing and walking.

**Material and methods** In this study data was collected for eight days in October 2016. A total of six sheep were selected with range of body condition (ranging from 2.5 to 4), age (18 months to 4 years) and breed. The sheep's locomotion were captured in two 2h sessions on each day. Sensors were custom-made developed by Intel using the Intel® Quark™. The device encompasses a triaxial gyroscope and accelerometer. Sensors were attached to sheep at two locations 1) the existing electronic ear tag via a tape and lightweight plastic tie and 2) to neck collar using tape and lightweight plastic tie. Data was collected using sampling frequencies of 8Hz, 16Hz and 32HZ and each sheep was used for multiple sampling frequencies. Data was segmented using window sizes of 3s, 5s and 7s. For each time window a set of 44 feature characteristics. A random forest algorithm with a 70% training set and 30% test set was used for the classification of lying, standing and walking. The performance of the classifier was evaluated using metrics such as overall accuracy, precision, recall, F-score and specificity. The performance of the classifier as a function of sensor position was assessed using a McNemar's test.

**Results** The comparison of the performance of the classifier for the different factors shows that the best performance for walking, standing and lying in sheep (accuracy 95%, F-score 93-97%) was obtained using a combination of 32Hz and 7s for both ear and collar sensors. However, comparable results (accuracy 93%, F-score 89-94%) on the performance of the classifier were obtained using a combination of 16Hz sampling frequency and 7s window. When comparing across the different window sizes, the worst accuracies were obtained for 3s and the best for 7s. The activity with the highest performance was lying. In general terms, the classification performance increased with increasing sampling frequency and window sizes.

**Conclusion** The results of this study show that using a random forest algorithm it is possible to accurately (92-95%) classify biologically relevant behaviours in sheep such as walking, standing and lying. Results using a 16Hz and 32Hz sampling frequency were comparable, thus suggesting significant benefits in the power consumption using 16Hz sampling frequency. There were no significant differences in accuracy of the classification between ear and collar. Therefore, an ear-mounted sensor, which can be easily integrated to existing tag identifiers, can accurately classify behaviours in sheep.

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# Blue light from light-emitting diodes (LEDs) directed at a single eye elicits a dose-dependent suppression of melatonin and affects milk production in dairy cows

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**Application** Wearable head masks that direct blue light at a single eye can suppress melatonin (MT) secretion in dairy cows. In pasture-based spring-calving multiparous dairy cows, this suppression in melatonin increases milk production during early lactation.

**Introduction** In confinement systems, providing dairy cows with a long-day photoperiod (18 hours light, 6 hours dark) suppresses circulating MT concentrations and increases milk production (Dahl *et al*., 2012). The research reported herein had two objectives. First, we wanted to determine the minimum light intensity directed at a single eye required to suppress circulating MT concentrations to those observed under indoor lighting systems in dairy cows. Second, we wanted to examine the effect of blue light treatment on milk production in spring-calving pasture-based dairy cows.

**Material and methods** Study 1: Five non-lactating Holstein-Friesian cows were housed in metabolism stalls during November 2015. Following a 14-day environmental conditioning period comprising 8 hours of light, 16 hours of dark (LD8:16), where mean (± SD) white light intensity levels by day (08:00 – 16:00) at eye level were 237 ± 68 Lux, cows were exposed to each of the following light intensities for 8 additional hours (16:00 – 00:00) using a 5 x 5 Latin Square design; <1 Lux; 70 Lux; 125 Lux; 175 Lux and 225 Lux. Light was administered via head worn masks fitted with LEDs emitting short wavelength blue light (465 nm) to the right eye. Each treatment night was followed by a break night, where the animals again received LD8:16. Two days after completion of the different light intensity treatments, all cows were exposed to the indoor lighting system until 00:00 (LIGHTS ON). Blood samples were collected from indwelling jugular catheters at 16:00, 17:00, 18:00, 20:00, 23:00, 00:00 and 01:00 on treatment nights and at 18:00 and 22:00 on break nights and plasma later assayed for MT by radioimmunoassay. MT data were log transformed, and analyzed using mixed models with treatment, period, hour and treatment x hour as fixed effects and cow as a random effect.
Study 2: 40 spring-calving cows were blocked for parity, calving date and Economic Breeding Index for milk production and randomly assigned to either a CONTROL treatment or blue light to a single eye (LIGHT) treatment from day of calving through 32 weeks of lactation (Feb-Nov 2017). The LIGHT treatment cows were fitted with light masks providing 225 Lux of blue light to the right eye that activated on a timer from dusk until 00:00. Milk production data were recorded daily using electronic milk meters, and collapsed into weekly means. These data were transformed to generate a normal distribution, and analysed using mixed models with repeated measures. Fixed effects included treatment, week, parity (primiparous or multiparous) and two-way interactions, and calving date was included as an adjustment variable. Cow within treatment was included as a random variable.

**Results** Study 1: A dose-dependent effect of treatment on mean circulating MT concentrations (and 95% CI) between 16:00 and 00:00 was observed [9.1 (6.8, 12.2), 4.9 (3.7, 6.6), 4.4 (3.2, 5.8), 3.3 (2.5, 4.4), 1.8 (1.4, 2.5) and 1.9 (1.4, 2.5) ng/ml for 0, 70, 125, 175, 225 Lux and LIGHTS ON treatments, respectively]. Only the 225 Lux mask treatment did not differ from LIGHTS ON, and hence 225 Lux was deemed as the minimum light intensity required.
Study 2: Across all cows, mean milk production (and 95% CI) during 32 weeks of lactation was not affected by treatment (20.3 (19.3, 21.3) vs 20.9 (19.8, 22.0) kg/d, P = 0.4; CONTROL and LIGHT, respectively). Within multiparous cows, a treatment by week interaction was detected (P = 0.048), whereby LIGHT treatment increased milk production during the first 12 weeks of lactation (25.8 (24.3, 27.3) vs. 28.0 (26.5, 29.5) kg/d; P = 0.04), but had no effect thereafter (21.0 (19.7, 22.4) vs. 21.8 (20.5, 23.2) kg/d; P = 0.4).

**Conclusion** These studies have identified the minimum blue light intensity administered to a single eye required to acutely suppress circulating concentrations of MT to those observed under indoor lighting regimes. They provide the first evidence of an effective application of this technology for simulating a long day photoperiod to significantly increase daily milk yield in grass-based multiparous dairy cows during the first 12 weeks of lactation.

**References**

Dahl, G.E., Tao, S. and Thompson, I.M. 2012. Journal of Animal Science 90, 755-760.

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# Use of thermal imaging for the assessment of pyrexia in pre-weaned artificially reared calves

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**Application** Thermal imaging has the potential to be used as a non-invasive method in the prediction of body temperature of calves.

**Introduction** Body temperature is often monitored in cattle for the diagnosis of ill health usually by inserting a thermometer into the rectum. This is an invasive process and can be subject to error. Thermal imaging therefore represents a non-invasive means of assessing body temperature. Objects emit heat in the form of infra-red energy and the hotter an object is, the more infra-red energy is given off. Thermal imaging devices detect this infra-red energy and create an electronic image based on different temperatures. However, (thermal) environmental factors, such as ambient temperature and wind speed can also influence thermal image temperature (Church *et al*., 2014). The aim of this study was to assess the use of thermal imaging in the diagnosis of pyrexia in pre-weaned artificially reared calves.

**Material and methods** One hundred Holstein bull calves and 25 Holstein heifer calves were used in this study with all calves housed in a group Igloo system (*Holm & Laue*) and grouped by sex of calf, with each group containing a maximum of 14 calves and fed via an automatic milk feeder (*H&L 100*). Calves commenced on the study at 8.0 days ±1.0 (mean ± sd) as they entered the group housing system. Measurements were taken daily (between 0800 h and 1000 h) until day 40 of age when the weaning process commenced. All calves were collected and placed within a temporary pen (5m x 1.9m) at the front of the group pen where they remained until the measurements had been taken after which they were returned to their group pen. From here, each calf was individually placed in a modified weigh crate located in the alley outside the group pen. Each calf then had its temperature taken rectally with a digital thermometer (*Genia Digiflash*). The thermal image was taken from the area in and around the medial canthus of the left eye of each calf using a FLIR SC620 thermal camera. Attached to the modified weigh crate, 0.75m from ground level, was a Kestrel 4000 weather meter and readings for ambient temperature ($^{o}$C), relative humidity (%) and wind speed (m/s) were recorded at the time the thermal image for each calf was taken. Water vapour density (g/m$^3$) was calculated using relative humidity and ambient temperature. The thermal images were downloaded and processed using ThermaCAM™ Researcher Professional 2.10 software. To obtain the maximum temperature from the required part of the thermal image, a circle 2.5cm in diameter was imposed over that section and the maximum temperature extracted. Data was analysed using each observation as a repeated measure in REML with calf as the random effect.

**Results** Measurements were taken over ambient temperature range -0.8 to 22.6 $^{o}$C and wind speed 0 to 2.3 m/s. Initial analysis of the data (Table 1) has shown that environmental parameters such as ambient temperature and wind speed have a significant effect on the use of thermal imaging in the prediction of rectal temperature. The sex of the calf also proved to be significant.

**Table 1** Prediction of rectal temperature

| Fixed Effect | Estimate | S.E. of Estimate | P value |
|---|---|---|---|
| Intercept | 27.300 | 0.553 | <0.001 |
| Thermal Image temperature ($^{o}$C) | 0.323 | 0.015 | <0.001 |
| Ambient temperature ($^{o}$C) | -0.035 | 0.011 | <0.001 |
| Relative humidity (%) | -0.001 | 0.002 | 0.586 |
| Water vapour density (g/m$^3$) | 0.023 | 0.021 | 0.274 |
| Wind speed (m/s) | 0.154 | 0.022 | <0.001 |
| Sex (F) | -0.177 | 0.043 | <0.001 |
| Age (days) | -0.001 | 0.001 | 0.312 |

**Conclusion** Thermal imaging looks like it has the potential to be used in the prediction of body temperature in pre-weaned artificially reared calves. However, further analysis needs to be undertaken to examine whether or not ambient temperature, wind speed and sex of the calf should be included when developing a predictive equation.

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**References**

Church, J.S., Hegadoren, P.R., Paetkau, M.J., Miller, C.C., Regev-Shoshani, G., Schaefer, A.L. and Schwartzkopf-Genswein, K.S. 2014. Research in Veterinary Science 96, 220-226.

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# Combining offline and online classifiers for the classification of sheep behaviour

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**Application** One of the greatest challenges for a robust and long-term implementation of an automated behavioural classifier in animals is how to transfer knowledge for smart devices into changing conditions. By combining a static offline classifier with an online classifier it can be possible to develop a flexible system that can accurately classify relevant behaviours in sheep under changing conditions.

**Introduction** Behavioural activity classifier algorithms in humans and animals require large amounts of labelled training data under different conditions. This represents one the biggest challenges for the development of a flexible system that can accurately classify behaviour under very diverse circumstances (Cook *et al.* 2013). This challenge can be tackled by transferring knowledge learned from static offline classifiers and combining it with online information to improve and extend the classification capabilities of a system. In this study, we present a two stage classification algorithm in sheep that combines a static offline classifier with online information to improve the classification performance under new conditions.

**Material and methods**
In this study data was collected for a total of 30 days in 2017. A total of 26 sheep were selected via stratified random sampling from a flock of 140 animals at the University of Nottingham. Sheep were assessed for body condition, age and breed. Sheep behavioural activities were recorded twice a day in a morning and afternoon sessions. Sheep were fitted with a custom-made wearable device based on the Intel® Quark™ which was attached to the existing electronic identification tag. The device had an implemented machine learning algorithm that automatically classified walking, standing and lying in sheep. The algorithm was developed as a static offline classifier using data collected previously. The device can also provide a measure of the magnitude of the acceleration over a window. Behavioural classification labels and the magnitude of the acceleration outputs from the sensor were combined in a second online classifier. The results of the two-stage classifier were compared against ground truth behavioural observations for its validation. The performance of the two-stage classifier was validated using the overall accuracy, precision, recall, F-score, and specificity values metrics.

**Results** The two-stage classifier showed a significant improvement in the classification of the three different behaviours when compared to the performance of the static offline classifier only. Overall accuracy increased from 55.58% using only offline classification information to 82.96% using the two-stage classifier. The best improvement in the classification were for walking, increasing from 44.72% to 91.38%, and standing increasing from 41.32% to 75.81%. Sensitivity increased from 51.05% to 63.69% while specificity increased from 71.15% to 83.04%.

**Conclusion** The results show that by combining a static offline classifier with online information it is possible to improve the performance of a classifier and to incorporate changing conditions that can affect a behavioural monitoring system. A two stage classifier system offers a huge potential for a flexible monitoring system that adapts to changing and new circumstances.

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**References**
Cook, D., Feuz, K.D. and Krishnan, N.C. 2013. Knowledge and information Systems 36(3), 537-556.

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**098**

# Assessing and monitoring soil quality in Irish grassland soils

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**Application** Soil quality is essential for sustainable land management and grass production. Soil quality was assessed across Irish grassland soils and a land use intensity index was developed to help advise and monitor soil quality and related soil functions.

**Introduction** Irish agriculture is dominated by grass based animal agriculture which enables Irish farmers to produce milk and meat products in a sustainable manner and to compete on world markets. Agriculture today is faced with delivering increased primary productivity to meet the growing global demand for food security. At the same time, society expects that any emphasis placed on increasing food outputs is met with an equal emphasis on sustainability particularly as historically the intensification of agriculture, although not always, has often been associated with deleterious impacts on the environment. Animal grazing, may lead to serious damage to vegetation and soil. Many studies have demonstrated that grazing contributes to breakdown soil aggregates, making the soil more subject to decreased porosity and compaction (Novikoff, 1983; Pluhar *et al*., 1987). This has as consequence the damage of soil structure which is a key factor that supports all soil functions (Avondo *et al*., 2013). On the other hand, the presence of livestock has the power to return organic manure on the soil, which contributes to a high overall soil quality. In order to protect and manage these grassland and soil resources sustainably requires managing the landscape in its totality with due recognition of its complex integrated nature. Moreover, within Ireland, Food Wise 2025 objectives to intensify agriculture are coupled with greening objectives of the Common Agricultural Policy amongst other environmental policy. Thus, any intensification of agriculture must be achieved in a sustainable manner. On the light of this, the SQUARE project aimed to assess the interactions of different management practices on the overall soil quality with a special focus on livestock grazing for the multi-faced role that this management system has on the overall soil quality. Within SQUARE, soil structural quality through physical parameters and visual methods was assessed, and the combination of management activities and soil functional quality was evaluated.

**Material and methods** Within SQUARE a survey of grassland soils was undertaken representative of the main soil and five major agro-climatic regions of Ireland. Detailed analysis of soils and herbage were taken from 38 grassland farms where a series of samples and measurements were conducted using soil profile pits and *in-situ* and laboratory analysis. The different functions were assessed through combined measurement of commonly used parameters of soil quality. In order to assess the impact of management operation within those functions and aiming to study synergies and trades off between them, a land use intensity index (LUI) was developed using management information collected through on-farm questionnaires on management practices. This detailed questionnaire aimed to capture all the common farming practices carried out in Irish grassland soils. The derived land use intensity index (LUI) was based upon three management intensity components: (i) intensity of Fertilisation (Fi), (ii) frequency of Mowing (Mi) and (iii) intensity of Livestock Grazing (Gi).

**Results** The ability of LUI components and aggregated LUI to capture changes for each soil function is shown in Table 1. The symbol (+) indicates higher LUI/components explanatory power for the different soil functions. Soil structure has been added due to its importance as a key driver of soil functionality.

**Table 1**

| | Fi | Gi | Mi | LUI |
|---|---|---|---|---|
| Primary productivity | + | | + | |
| Water purification and regulation | | | | |
| Carbon sequestration | + | | | |
| Biodiversity | + | | + | + |
| Nutrient cycling | + | + | + | + |
| Structure | + | | | + |

**Conclusion** Initial findings indicate that the LUI components are efficient metrics to assess the influence of management across soil functions. Biodiversity and Nutrient Cycling were highly affected by intensive grassland management. The remaining soil functions were only partially explained by the LUI. Fi showed the highest ability to capture changes across functions whereas the Gi explained changes only for Nutrient Cycling function. Soil structure quality showed a greater sensitivity to fertilising operations (Fi), and also the combined effects of fertilising, grazing and mowing indices together, represented in the LUI.

**References**

Novikoff, G., 1983. Ambio. 12, 102–105.
Avondo, M., *et al*., 2013. Italian Journal of Agronomy 8, 3–19.
Pluhar, J. J., *et al*., 1987. Journal of Range Management 240-243.

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# Impact of soil-enhancers and plant bio-stimulants on the microbial profile of wilted ryegrass forage: A field experiment

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**Application** Increased biotic stress makes crops susceptible to undesirable microbial growth. Consequently, yield loses, fungal proliferation and possible mycotoxin production create undesirable conditions for optimum ensiling. The application of these products will limit the stress factors and enhance the proliferation of a desirable microbial environment, thus forage will ferment more quickly and remain stable.

**Introduction** The use of soil-enhancers and plant bio-stimulants for agricultural use has grown steadily in the last decade. It is estimated that the current market will have an annual growth of 12.5 % by 2018 (Calvo, 2014). Bio stimulants are originated from either natural or biological sources and when applied in small quantities on plants and/or rhizosphere, they increase the development and growth of the plant as well as the nutrient uptake, soil structure and overall plant response (Calvo, 2014). Crop quality is a fundamental parameter to ensuring optimum forage growth along with limiting abiotic stress and eventually resulting to a desirable microbial environment for ensiling. The aim of the present study was to assess the effect of two commercial products on the natural microbial communities that are involved with the process of ensiling in an open field experiment. These products include a soil-enhancer (SE) by the commercial name Soil-Set® aid and a plant bio-stimulant (PB) by the commercial name Imprograin® and were provided by Alltech (Sarney, Dunboyne, Ireland). The initial hypothesis was that the current application would alter the microbial population and nutritional value of the pre-ensiled ryegrass crop and therefore the silage produced from it.

**Material and methods** Both SE and PB were tested on perennial ryegrass mixture (Hybrid silage lay, Cotswold grass seeds, Gloucestershire, UK). The seeds were planted on a 1.5 ha field (Rothamsted Research, North Wyke, Devon, UK). SE was added to the soil after sowing (2 L/ha) while PB was sprayed on the crops after the first cut (1 L/ha). The field was split into 24 plots (24*26 m each) as a randomized block design with 6 blocks and a 2x2 factorial treatments structure. Experimental treatments consisted of an untreated control; soil treated with SE; plants treated with PB; and soil and plants treated with both SE and PB. Focus was set on Enterobacteriaceae (ENB), Lactic acid bacteria (LAB), *Pseudomonas spp.* (PSE) and yeasts. Microbial populations were enumerated using 10-fold serial dilutions of ryegrass mixed with ¼ strength Ringer's solution (BR0052, ThermoFisher, UK). Defined growth media was used for each microorganism. Following 24 h growth, plates with microbial colonies between 30 and 300 were used for calculating microbial numbers. Total enumerated colonies underwent a logarithmic transformation ($Log_{10}$) to follow normal distribution. All data were analysed with analysis of variance (ANOVA) using the 18th version of GenStat Software (VSNI). Significance level was set at 95% ($P<0.05$).

**Results** As shown in Table 1 microbial population varied significantly between treatments. ENB and LAB varied between control and PB-treated crops ($P=0.009$, $P=0.011$ respectively), while PSE varied across all 4 treatments ($P<0.001$). In terms of yeasts, PB-treated crops contained the lowest population in comparison to all the other treatments ($P<0.001$).

**Table 2** Average $Log_{10}$(CFU/g) between wilted ryegrass treatments. Superscripts a, b, c and d indicate significant differences between the means. Means not sharing a letter are significant. Standard error of differences = S.E.D.

| Average Log10 (CFU/g) | Control | SE | PB | SE+PB | SED | P |
|---|---|---|---|---|---|---|
| ENB Log10(CFU/g) | $^{b}4.25$ | $^{ab}3.85$ | $^{a}3.63$ | $^{ab}4.03$ | 0.159 | 0.009 |
| LAB Log10(CFU/g) | $^{a}3.89$ | $^{ab}3.97$ | $^{b}4.36$ | $^{b}4.32$ | 0.148 | 0.011 |
| PSE Log10(CFU/g) | $^{a}6.44$ | $^{b}6.46$ | $^{d}6.50$ | $^{c}6.48$ | 0.004 | <0.001 |
| Yeasts Log10(CFU/g) | $^{c}6.48$ | $^{b}6.45$ | $^{a}6.38$ | $^{b}6.44$ | 0.009 | <0.001 |

**Conclusion** PB-treated crops contained the highest numbers of LAB and the lowest numbers of ENB and yeasts trend which are associated with favourable crop ensiling conditions. The difference between ENB and LAB is crucial as both compete for carbohydrate based sources during the ensiling process. Higher starting LAB numbers will result in a more rapid pH decline and create a more stable fermentation process. As a part of this study the harvested ryegrass will be ensiled in big bales to monitor the microbial community changes during ensiling and after during aerobic exposure.

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**References**

Calvo, P., Nelson, L. and Kloepper, J.W. 2014. Plant and Soil 383, 3-41

# Integrated soil fertility management for grass based dairy systems

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**Application** Declining soil fertility levels on Irish grassland farms has the potential to threaten the sustainability of dairy production systems. A more soil specific and spatially targeted approach to nutrient management shows potential to increase nutrient use efficiency, grass productivity, environmental performance and profitability on Irish farms.

**Introduction** Soil fertility and nutrient management are two of the key factors driving productivity and environmental sustainability on grass based farms. Farmers regularly manage the fertility of soils by applying fertilisers and organic manures to build-up or maintain the supply of nutrients required by grass in order to meet the requirements of grazing animals. However, experienced farmers will know that not all soils (or fields) have the same production potential (or suitability for certain crop types) or respond in terms of their soil fertility status to the nutrients that are applied. This poses a challenge for individual farmers and their advisors when planning nutrient and fertiliser management strategies for their farms. In a real scenario, not all fields, although having similar soil test results, respond equally to similar nutrient application rates. This is because different soil types possess different characteristics and qualities to receive, store and supply nutrients for grass growth. In 2014 Teagasc finalised the 3rd edition National Soil Map of Ireland, which provides an overview of soil types found across the country at a scale of 1:250,000. This provides a useful summary of the type of soils to be found within a particular catchment area. In 2015, Teagasc launched the N&P online web-based management system for providing nutrient management planning advice to farmers. This system is based on detailed crop response trials to establish the key relationships between soil nutrient tests and crop production requirements. The combination of the data used in both of these systems could be utilised to derive nutrient management advice. The work summarised here aimed to understand how and to what extent the knowledge on nutrient requirements in agricultural production systems combines with the major characteristics defining the variability in soils.

**Material and methods** Twenty one dairy farms were selected based on general soil drainage class to evaluate nutrient management practices at the field and paddock scale. Each farm was intensively soil sampled on a per paddock basis in 2015, 2016 and 2017. Grass growth and herbage quality was measured throughout each growing season for each year in every individual paddock. These data were coupled with biophysical data including soil type, drainage, weather etc. and management data (i.e. fertiliser and slurry applications, grazing management etc.) in order to evaluate nutrient flows, recoveries, losses and efficiencies within and between farms.

**Results** Results highlight high spatial variability in soil fertility status within farms. In particular, low pH levels are impeding grass production by reducing the nutrient availability of both stored nutrients in the soil and freshly applied nutrients. Current nutrient management practices do not address variability issues at the sub-field scale. The requirement for increasing applications of lime, P and K indicated that farmers are not prioritising nutrient management in line with increasing stocking rates. In 2016 the average grass uptake values per paddock (n=384) was 356.02 kg /ha N, 37.29 kg/ha P and 277.62 kg/ha K in 2016.The average nutrient balance (= nutrient inputs – nutrient offtakes) per paddock was 133.60 kg/ha N, -1.6 Kg/ha P and 14.83 kg/ha K in 2016.

**Table 1** Nitrogen, phosphorus and potassium balance at the field and farm scales across 10 dairy farms

| | Average field/paddock scale nutrient balance (kg/ha) | | | |
|---|---|---|---|---|
| Sample size | Year | Nitrogen | Phosphorus | Potassium |
| n=384 | 2015 | 133.67 | -4.52 | 9.29 |
| n=384 | 2016 | 133.60 | -1.64 | 14.83 |
| | Average farm scale nutrient balance (kg/ha) | | | |
| n=10 | 2015 | 75.47 | 2.41 | 18.25 |
| n=10 | 2016 | 67.34 | 1.03 | 12.32 |

**Conclusion** A more targeted approach to nutrient management on grassland a farms is required to correct soil fertility and increase nutrient sustainability of the farming systems. Increased knowledge transfer (advisor/education support) will be necessary to achieve better distribution of nutrient across farms to achieve high levels of nutrient efficiency and increase grassland productivity and carrying capacity.

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# Using NMP-Online to improve nutrient management on Irish grassland farms

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**Application** In the last 10 years the soil fertility on Irish Farms has declined. NMP online was developed to aid farmers in achieving improved outcomes in relation to grass output, environmental performance and compliance with regulation

**Introduction** The introduction of regulation associated with the Nitrates and Water Framework Directives put in place a very restrictive regulatory framework in relation to the application of both Nitrogen and Phosphorus. When combined with increased fertiliser prices and an income crisis this resulted in a very significant fall in soil fertility which threatened the achievement of industry growth objectives. NMP online was developed support farmers to achieve improved soil fertility and compliance with regulation. In its first year NMP Online has been used to prepare nutrient management plans with 30% of Irish farmers.

**Material and methods** To deal with the new complexity in managing soil fertility following the implementation of the Water Framework and Nitrates Directives a variety of tools were prepared which were mainly spreadsheet based. These systems, for the most part, focussed on meeting statutory requirements. However, the complexity of the statutory requirement led to the production of plans which were not suitable for guiding farmers and the majority of farmers ignored these plans when it came to applying their chemical and organic fertilisers. Teagasc undertook a process of consultation with farmers. The outcome of this process was a request for a divergence in output within a nutrient management planning system with tabulation based outputs for regulators but a much more visual output for farmers integrating mapping and graphical outputs to support the day to day actions at farm level. In developing NMP online Teagasc had three main objectives:-

- To improve nutrient management at farm level in support of more efficient, competitive and profitable farming systems
- To improve the efficiency and quality of plans produced to meet the statutory requirements
- To improve environmental outcomes, particularly in relation to water quality and gaseous emissions

NMP Online is available to all advisers and consultants working in Ireland. It facilitates the efficient production of high quality nutrient management plans and provides a basis for improving soil fertility management on Irish farms.
NMP Online utilises a data-set of farm information to allow a consultant to work with a farmer to create a nutrient management plan. Much of the required data is available from other sources and the system integrates with a number of databases to speed up the planning process. Connected databases include DAFM land parcel data, DAFM livestock information, and soil analysis results. Completion of the plan involves a sequential process. Some of the components are optional in completing the plan (mapping and winter housing and storage).

**Results** In its first year of operation NMP online has become the mandatory system for the development of nutrient management plans for agri-environmental scheme application and for complying with the nitrates derogation. This has resulted in NMP being used to prepare 40,000 plans in 2018. It is expected that this will increase to 60,000 in 2018. Feedback from farmers has been very favourable in relation to the format of the plans.

**Conclusion** NMP Online has been developed as a comprehensive Nutrient Management Planning and has been used extensively. However, the optimisation of the use of the system to deliver improved outcomes in terms of soil fertility status, improving water quality protection and reduction of GHGs remains a challenge. The focus on system development and rollout in the immediate future needs to focus on:

- Further improvement in outputs to support practice change at farm level
- Increasing the efficiency in preparation of plans to allow a review process to become a key part of the annual engagement between advisers and clients

NMP Online is a key tool for Irish Agricultural to achieve industry output and environmental targets. Ongoing development of the system and its integration into farmers adviser interactions will need significant focus.

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# The effect of nitrogen application rate and grass ploidy on herbage production and sward clover content in frequently grazed swards

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**Application** Perennial ryegrass ploidy does not affect sward white clover content. Increasing N application rate generally reduces sward clover content.

**Introduction** Perennial ryegrass (*Lolium perenne* L.) ploidy and N fertiliser application rate have been shown to influence sward white clover (*Trifolium repens* L.) content and herbage production (e.g. Davies *et al*., 1991; Swift *et al*., 1993; Enriquez-Hidalgo *et al*., 2016). The objective of this study was to determine the effect of ploidy and nitrogen (N) application rate on sward clover content and herbage production under frequent, tight grazing.

**Material and methods** The experiment was undertaken at Teagasc, AGRIC, Moorepark, Fermoy, Co. Cork, Ireland on a free-draining, acid brown earth of sandy loam to loam texture. The experiment had a 2 × 2 × 5 factorial design with four replicates. Treatments were two sward types (grass-only and grass-clover); grass was either a tetraploid (Kintyre) or diploid (Aberchoice) perennial ryegrass sown with or without clover (cv. Iona); swards received 0, 60, 120, 180 or 240 kg N/ha. Plots were grazed 8-9 times per year. Measurements were taken for a three year period (2014 - 2016) and included pre-grazing and annual herbage mass (HM), post grazing sward height and sward clover content. Data were analysed using Proc Mixed in SAS.

**Results** There was no effect of ploidy on annual herbage production, pre-grazing HM or sward clover content. Herbage production increased as N application rate increased (Table 1) in both sward types. Treatments receiving 120, 180 and 240 kg N/ha had significantly greater (P<0.05) herbage production than those receiving 0 kg N/ha. Increasing N rate increased pre-grazing HM in grass only and grass clover swards. Clover inclusion increased herbage production in all treatments except for the treatment receiving 240 kg N/ha. Nitrogen rate had a significant effect on annual herbage production and sward clover content which was greatest on the treatment receiving 0 kg N/ha and least on the one receiving 180 kg N/ha.

**Table 1** Effect of ploidy (tetraploid or diploid), sward type (grass-only or grass-clover) and N rate (0, 60, 120, 180, 240 kg N/ha) on annual herbage production, pre-grazing herbage mass (kg DM/ha) and sward clover content (%)

| | Grass-only | | | | | Grass-clover | | | | | S.E | P Value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Nitrogen rate (kg N/ha) | 0 | 60 | 120 | 180 | 240 | 0 | 60 | 120 | 180 | 240 | | N rate | Sward Type | Ploidy |
| Annual herbage production (kg DM/ha/year) | 7178 | 9012 | 9995 | 10395 | 11153 | 8346 | 9510 | 10618 | 10540 | 11098 | 360.6 | <0.001 | <0.05 | NS |
| Pre-grazing herbage mass (kg DM/ha) | 1011 | 1260 | 1395 | 1449 | 1621 | 1257 | 1329 | 1480 | 1469 | 1545 | 49.3 | <0.001 | <0.05 | NS |
| Average clover content (%) | 33 | 25.6 | 23.2 | 17.6 | 25.8 | | | | | | 2.41 | <0.05 | - | NS |

**Conclusion** There was no effect of ploidy on herbage production or sward clover content. In general, increasing N application rate reduces sward clover content.

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**References**

Davies, D.A., Fothergill, M. and Jones, D. 1991. Grass and Forage Science 46, 39-49.
Enriquez-Hidaalgo, D., Gilliland, T. and Hennessy, D. 2016.Grass and Forage Science 71, 559-574.
Swift, G., Vipond, J.E., McClelland, T.H., Cleland, A.T., Milne, J.A. and Hunter, A. 1993. Grass and Forage Science 48, 279-289.

# Commercial farm dry matter production performance of grass varieties on Irish grassland farms

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**Application** New on-farm evaluation of grass varieties has the ability to identify profitable phenotypic traits and redirect the next generation breeding towards more suitable varieties for intensive dairying.

**Introduction** Grass is Ireland's greatest renewable feed resource and provides the main feed for Irish ruminant livestock. The phenotypic improvement for this forage offers a cost-effective mechanism to increase profitability and reduce environmental cost of animal production**.** Plot based ryegrass evaluations are conducted under simulated grazing studies to identify varieties with the best phenotypic performance. The objective of the current study was to evaluate the on-farm performance of the varieties under a commercial management regime giving a realistic platform to identify superior varieties for grazing systems.

**Material and methods** This was a longitudinal study of grass performance on 475 paddocks across 79 grassland farms during the period January 2013-December 2016. Farms which varied in agroclimatic regions on differing soil types operating grass-based spring calving herds were identified and enrolled. PastureBase Ireland is a web-based management tool which has the dual function of providing real-time decision support for farmers while acting as a national database which captures information for benchmarking and research purposes (Hanrahan *et al*., 2017). Within PastureBase Ireland (PBI) the drainage characteristics of each paddock were also categorised. A range in pH across all farms was noted with a pH average of 6.30 operating at between 170 and 250kg N/ha/yr across paddocks with an additional allowance of 250kg of inorganic N/ha/yr. Farmers with a proven history of grassland measurement and management based on the PBI records indicated the quality of the data collected. All farmers were trained to take grass measurements and were part of a discussion group that met monthly during the grazing season. It operates at the paddock scale as the basic unit of measurement as did this experiment. All information (varieties sown, altitude aspect drainage soil status etc.) is recorded by the farmer in the bed interface. All measurements on PBI are calculated on a per hectare basis. Grass cover estimations are entered on a weekly basis during the main growing season using a plate meter or by visual assessment (O'Donovan *et al*., 2002). Grass varieties were sown in monoculture in individual paddocks from 2011 to 2016 on all of the 79 commercial farms according to stringent guidelines which ensured successful sward establishment. All paddocks for reseeding were sprayed with Glyphosate to ensure that all botanical species were removed. Surface trash was removed pre cultivation. A range of cultivation methods were used to form a fine and firm seed bed. When a suitable seed bed was achieved, varieties were sown at a seeding rate of 34.5kg/ha. N, P and K were applied at the time of sowing. Varieties with a range in heading dates were selected and all farms were allocated tetraploid and diploid varieties. Grass DM production was measured from January 1$^{st}$ to December 31$^{st}$ annually. Grazing and silage DM yields were assessed prior to grazing or harvest and were recorded in PBI. In the data analysis, newly reseeded swards in their first year of analysis were excluded from the data set. The effect of variety on DM production was estimated using a mixed linear model in PROC MIXED (SAS inst. Inc., Cary, NC, USA) with paddock nested within farm included as a repeated measure with a compound symmetry covariance structure assumed among paddocks within a farm. Pearson's correlation coefficient was used to determine the strength of the linear association for total and grazing herbage production.

**Results** There was a variety by year interaction ($P < 0.001$) for total and the grazing DM yield. There was a significant ($P < 0.001$) effect of variety on total and grazing DM yield. The highest performing variety for total DM yield was AberMagic (13653kg ± 435kg) and the lowest yielding variety was Glenveagh (11993kg ± 367kg). The highest yielding variety with respect to grazing DM was Astonenergy (12024kg ± 357kg) and the lowest yielding variety was Dunluce (10101kg ± 431kg).

**Conclusion** This early work is part of a long term program evaluating varieties on commercial farms. This data shows that commercial farm variety performance can be reliably estimated from on-farm evaluations. Results show that on-farm evaluation is effective in identifying suitable varieties for intensive grazing regimes and will direct variety choice and traits for breeding in the future.

**References**

Hanrahan, L., Geoghegan, A., O'Donovan, M., Griffith, V., Ruelle, E., Wallace, M., and Shalloo, L. 2017 Computers and Electronics in Agriculture 136, 193-201.
O'Donovan M., Dillon, P., Rath, M., and Stakelum. G. 2002 Irish Journal of Agricultural and Food Research 41(1), 17-29.

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# Spatial variation in baled silage quality and faecal origin microbes

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**Application** Results impact management practices in silage bale preparation and storage conditions. Work highlights the opportunity for improved forage quality and reduced health risks through achieving and maintaining anaerobic conditions.

**Introduction** The role of preserved feed within on-farm microbial cycling has consistently been underestimated. The importance of bacterial survival and proliferation within preserved feed is critical in the return of faecal origin microbes to livestock. This stage in microbial cycling effects feed quality, animal health and can elicit selective pressure on microbial population diversity. This study aimed to investigate spatial variation in baled silage quality and faecal origin microbes.

**Material and methods** Cut grass was baled before being wrapped (4 layers of plastic) less than 24 h after wilting in field. Silage bales were labelled to identify source fields and positions in stack, where they were maintained for 8 weeks. Silage sampling was conducted alongside standard farm feeding practice. Bales were selected by farm staff for consecutive days' feeding (n=5). The bale position in stack, field label and qualitative metrics for assessing silage bale quality were noted. Quantitative assessment of bale wrap integrity was measured with a custom-built pressure gauge, providing comparative assessment of anaerobicity. Coring was conducted through the bale wrap, with the number of wraps counted to confirm farm practice, in the sampling pattern indicated in Figure 1. Cores were taken to the centre of the bale (60 cm ± 5 cm). Variation was defined by bale, section (side or centre), position (top, mid or base) and depth (inner or outer). Individual cores to constituent depths were placed in separate sealed bags. Sterility was maintained between sampling points. Core samples were mixed and sub-sampled apportioned: 5 g for faecal indicator organisms (FIO) colony forming units (CFU), 10 g for pH, 100 g for DM and ash component testing with the remainder for forage quality analysis by NIRS as described by Parker *et al*., (1998). FIO CFU was determined in 5 g sub-samples to which 45 ml Ringer's buffer solution was added before maceration in sterilised blender capsules and serial dilution for standard membrane filtration as described by Hodgson *et al*., (2016). The variation of FIO CFU $g^{-1}$ was calculated by blocked general ANOVA. Correlation of NIRS forage quality analysis to the calculated metrics of pH, DM and ash was completed by linear mixed modelling. Variation in forage quality parameters was calculated by blocked general ANOVA to microbial results.

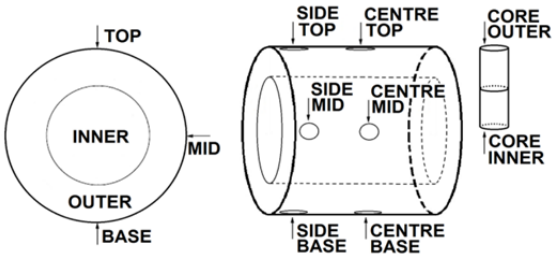


**Figure 1** Individual silage bale core sampling plan

**Results** Most significant source of variation resulted from between 'bale' differences for CFU of *Escherichia coli* (*E. coli*) and intestinal enterococci (In-ent) (0.001). This was treated as a blocking factor in subsequent analysis. The 'section' was not significant in variation of *E. coli* or In-ent CFU. The 'position' at top, middle or bottom within the bale as it was stacked was significant in CFU of *E. coli* (0.010) but not in In-ent, both had lowest CFU at top and highest at base position. The 'depth' had significant differences (0.001) for both, highest CFU of *E. coli* in outer and for In-ent in the inner depth.
The accuracy of NIRS forage quality analysis was confirmed by linear mixed modelling to be significantly associated to calculated pH (0.001), DM (0.010) and ash (0.001). As with microbial analysis, the greatest source of variation for forage quality indicators resulted from between 'bale' differences and this was again accounted for by blocking. Protein % was significantly different between 'depths' (0.050) with the higher content associated with the outer depth and showed a trend across positions with lowest content at the top and highest at the base. ME MJ/kg also indicated a trend across positions with lowest concentration at the base and higher at the top. pH showed no significant differences across stratum.

**Conclusion** The 'outer' depth represents approximately 75% of the silage bale volume. This horizon has critical economic importance to farmers. This study indicates an association of increased *E .coli* CFU and higher Protein % in outer 'depth' with the converse being true for In-ent CFU. As the differences between bales was indicated as the predominant source of variation, there is significant opportunity to achieve mutual benefits to health and production through improved stakeholder management during wrapping and stacking. Higher resolution testing may indicate significant differences in 'position' supporting the identified trends.

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**References**

Parker, R. S. *et al*., 1998. Animal Feed Science and Technology 72, 155-167.
Hodgson, C. J. *et al*., 2016. Journal of Environmental Management 183, 325–332.

# The effect of establishment method and maturity at harvest on yield and chemical composition of ensiled lucerne grown in south-east England

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**Application** Sowing in spring rather than autumn improved establishment of lucerne. Cutting earlier than guideline harvest maturity (early flower) moderately improved feed value at 1st cut but impaired regrowth, yield, and fermentation.

**Introduction** Interest in forage legumes for livestock systems has been growing due to higher fertiliser prices and increasing emphasis on sustainable farming practices. Lucerne (*Medicago sativa*) is the most popular legume forage crop globally; however, there is a need for a modern assessment of best practice for growing lucerne under UK conditions. Agronomic challenges of lucerne for UK growers include (i) high weed burden at establishment (ii) short harvest window for optimal digestibility (Tyrolova and Vyborna, 2008) and (iii) difficulty in ensiling the harvested material due to high buffering capacity (McBurney *et al*., 1983). The aim of the present study was to investigate the effect of establishment method (at one site in South England) and the effect of maturity at harvest on yield and chemical composition of resulting lucerne silage and sward regrowth. We hypothesised that lucerne silage feed value might be improved (i.e. more protein, less fibre) by cutting earlier than the currently recommended harvest maturity, but that regrowth would be compromised.

**Material and methods** A split-plot design was used in which twelve main 2.5m x 10m lucerne trial plots (variety daisy) were established (University of Reading, Berkshire, UK) by three differing establishment methods using four plots per method: Spring (S); Spring undersown in Barley (S+B) and Autumn (A) in Year 1. In Year 2, all plots were split to test the effect of harvesting at either an early (pre-flower, E) or recommended (10 - 30% flower, R) growth stage at each of two cuts. Cut material was ensiled in polyethylene mini-silos, allowed >90 d to ferment and subsequently analysed for chemical composition. Data from each cut were analysed separately using Mixed Models procedure of SAS version 9.4, testing for fixed effects of establishment, maturity and random effects of plot.

**Results** Within the establishment year, plots sown using the S and S+B methods gave a similar dry matter (DM) yield at 3.2 Tonnes (T) DM/ha whereas A sown plots did not establish in time to produce usable biomass. For spring establishment, undersowing with barley tended to reduce initial weed burden by 248 g/kg DM relative to no cover crop ($P < 0.06$). In Year 2, the combined 1st and 2nd cut yield of A sown plots was 7.4 T DM/ha less than that of both S sown plots (main effect of establishment: $P < 0.001$). At both 1st cut and 2nd cut, E harvest maturity reduced DM yield ($P < 0.03$; Table 1) relative to R. Early cut silage contained greater crude protein concentrations relative to R, however this effect was only significant at 1st cut. At both cuts, harvesting at E reduced or tended to reduce lactate concentration in the resulting silage ($P < 0.07$). The time required for regrowth between 1st and 2nd cut was 60 and 38 d for E and R cut material respectively.

**Table 1** The effect of harvest maturity on yield and chemical composition of lucerne silage

| | Maturity | | | |
|---|---|---|---|---|
| Item | E | R | SEM | *P* value |
| First Cut, g/kg DM | | | | |
| Yield, T DM/ha | 3.19 | 6.61 | 0.241 | 0.001 |
| Crude protein | 147 | 126 | 7.1 | 0.001 |
| NDF | 422 | 445 | 9.0 | 0.130 |
| pH | 5.11 | 4.91 | 0.069 | 0.086 |
| Lactate | 11.1 | 33.1 | 6.29 | 0.049 |
| Second Cut, g/kg DM | | | | |
| Yield, T DM/ha | 2.97 | 3.68 | 0.291 | 0.023 |
| Crude protein | 181 | 177 | 3.3 | 0.250 |
| NDF | 434 | 418 | 11.4 | 0.158 |
| pH | 5.19 | 5.44 | 0.170 | 0.135 |
| Lactate | 15.4 | 26.2 | 5.85 | 0.067 |

E, early; R, recommended; DM, dry matter; T, tonnes; NDF, neutral detergent fibre.

**Conclusion** Optimal establishment of lucerne at our site in southern England was achieved by sowing in spring rather than autumn. Autumn sown plots had poor yields and high weed burdens, perhaps due to delayed root development requiring plant resources in Year 2 as has been observed in previous studies (Sim *et al*., 2015). When harvesting 1st cut silage in Year 2, taking an early cut slightly increased crude protein concentration relative to the recommended harvest maturity but at a cost of reduced yield, slower regrowth, and poorer fermentation characteristics.

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**References**

McBurney, M. I., Vansoest, P. J. and Chase, L. E. 1983. Journal of the Science of Food and Agriculture 34, 910-916.
Sim, R. E., Moot, D. J., Brown, H.E. and Teixeira, E. I. 2015. European Journal of Agronomy 68, 69-77.
Tyrolova, Y. and Vyborna, A. 2008. Czech Journal of Animal Science 53, 330-335.

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# Effects of nitrogen fertiliser application rate to spring grass on intake, digestion and nitrogen balance in beef cattle

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**Application** Reducing fertiliser nitrogen (N) application rate is a means of lowering N excretion in beef cattle consuming spring grass.

**Introduction** Cattle production has been identified as a major contributor to N pollution of air and water (Dijkstra *et al*., 2013). Beef cattle partition dietary N into meat or excrete it in faeces and urine. Nitrogen intake is the main driver of N excretion but, in contrast to dairy cows, less is known about the relationships between nutrition and N excretion in beef cattle. Fertiliser N is the primary input influencing grass production, which is the cornerstone of many beef production systems in temperate climates. For moderately-intensive grass-based beef production systems fertiliser N use ranges from 200 to 250 kg N/ha with 50 to 100 kg N/ha typically applied during spring (Drennan and McGee, 2009). Therefore, the objectives of this study were to examine the effects of fertiliser N application rate to spring grass on herbage chemical composition, and intake, digestion and N-balance of beef cattle.

**Material and methods** Treatments, applied between April 25 and May 26 2016 to *Lolium perenne* dominant swards, comprised of two inorganic fertiliser N application rates - 15 (Low N - LN) and 80 (High N - HN) kg N/ha/rotation - harvested ca. 21 d post N application. Herbage was harvested daily at 08.00 h to a stubble height of 4 cm using a zero-grazer. Sixteen Charolais steers (475±18.4 kg) were used in a completely randomised block design experiment. The experimental period lasted 22 d with 14 d of dietary adaptation and 8 d of sampling in purpose-built metabolism stalls. Herbage was offered *ad libitum* daily during the adaptation phase and during the sampling phase, at 0.9 of the pre-determined dry matter (DM) intake in two equal meals. Total faeces and urine (in 9 M sulphuric acid) were collected on a 48 and 24 h basis, respectively. Blood samples were obtained by jugular venepuncture immediately before, 3 and 6 h post-feeding on d 12 and 22; plasma urea concentrations for each sampling time were averaged giving one value per treatment. Data were statistically analysed using the GLM procedure of SAS. The statistical model had fixed effects for treatment and block.

**Results** Herbage DM and CP concentration was 17 g/kg lower and 20 g/kg DM higher, respectively, for HN compared to LN (Table 1). Herbage DM intake or *in vivo* (DM) digestibility did not differ (P>0.05) between treatments. Nitrogen intake was 19 g/d greater for HN compared to LN (P<0.05). Nitrogen digestibility and faecal N loss did not differ (P>0.05) between treatments. Plasma urea concentration (P<0.05), total N and urine N loss (P< 0.01) was 1 mmol/l, 16 and 14 g/d greater for HN compared to LN. The quantity of N retained and therefore, N-use efficiency did not differ (P>0.05) between HN and LN (31 vs. 32 %).

**Table 1** Herbage composition (s.d.), intake, digestibility, nitrogen balance and plasma urea concentration in beef cattle consuming spring grass with low (LN) and high (HN) rates of fertiliser nitrogen applied.

| | LN | HN | s.e.m. | P-value |
|---|---|---|---|---|
| Herbage composition | | | | |
| Dry matter (DM) (g/kg) | 190 (3.1) | 173 (11.7) | - | - |
| Crude protein (g/kg DM) | 124 (10.9) | 144 (4.6) | - | - |
| DM intake (kg/d) | 7.6 | 7.4 | 0.20 | 0.454 |
| *In vivo* DM digestibility (g/kg) | 797 | 784 | 7.2 | 0.233 |
| Nitrogen (N) balance | | | | |
| N intake (g/d) | 151 | 170 | 5.3 | 0.041 |
| Digestible N (g/kg) | 678 | 701 | 16.9 | 0.353 |
| N loss (g/day) | 101 | 117 | 1.9 | 0.007 |
| Urine N loss (g/d) | 53 | 67 | 2.4 | 0.006 |
| Faecal N loss (g/d) | 48 | 50 | 1.4 | 0.217 |
| Retained N | | | | |
| g/day | 50 | 53 | 5.6 | 0.780 |
| g/kg N intake | 323 | 309 | 27.2 | 0.738 |
| g/kg N absorbed | 467 | 441 | 36.0 | 0.613 |
| g/kg bodyweight | 11 | 11 | 1.1 | 0.752 |
| Plasma urea (mmol/l) | 3.9 | 4.9 | 0.23 | 0.019 |

**Conclusion** Increasing the rate of fertiliser N application to spring grass resulted in higher herbage N concentration, N intake, plasma urea concentration and urine N excretion but had no effect on DMI, DM digestibility and N-use efficiency.

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**References**

Dijkstra, J., Oenema, O., van Groenigen, J.W., Spek, J.W., van Vuuren, A. M. and Bannink, A. 2013. Animal 7, 292-302.
Drennan, M.J. and McGee, M. 2009. Livestock Science 120, 1-12.

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# The effect of white clover on herbage production over a 3-year period

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**Application** Swards containing white clover had a dry matter yield advantage over grass-only monocultures. However, this yield advantage decreased as white clover proportion decreased over the experimental period (2014-2016).

**Introduction** The majority of previous studies into the incorporation of white clover (*Trifolium repens* L.) in perennial ryegrass (*Lolium perenne* L.) swards in dairy production systems have been at low nitrogen fertiliser (N) (< 150 kg N/ha) and low stocking rate (SR; < 2 livestock units/ha) levels whereas high N levels and high SR favour optimum herbage dry matter (DM) production in grass-only (GO) systems. Increasing artificial N decreases sward white clover proportion in grass-clover (GC) swards (Carlsson and Huss-Danell, 2003), and increasing SR on GC swards can result in damage to white clover plants. However, under high N levels, a high SR can minimise the negative effects of increased grass growth on white clover proportions (Harris and Clark, 1996). The objective of this study was to examine the effects of white clover inclusion on herbage DM production in an intensive animal grazing system, under high N fertiliser (250 kg N/ha) and high SR (2.75 LU/ha) levels over a 3-year period (2014-2016).

**Material and methods** A 3-year farm-systems grazing study (43.6 ha) commenced in 2014 on 1 and 2-year old swards. The study consisted of four sward types: tetraploid only; diploid only; tetraploid + clover; diploid + clover, amounting to two GO swards and two GC swards. Four diploid cultivars (30 kg/ha) and four tetraploid cultivars (37 kg/ha) were sown with and without white clover (50% Chieftain; 50% Crusader; 5 kg/ha) in five blocks. Nitrogen fertiliser application was 250 kg N/ha/year and each sward type was stocked at 2.75 cows/ha. Herbage production was estimated visually and a weekly grass wedge profile was produced using a decision support tool, PastureBase Ireland (Hanrahan *et al*., 2017). Sward white clover proportion in each GC treatment was determined at each grazing by hand plucking 10-15 herbage samples above 4 cm, separating the components and calculating the proportions of each component of the sward on a DM basis. Analyses were undertaken on all variables (herbage production, sward white clover proportion) using the mixed model (PROC MIXED) and regression (PROC REG) procedure in the statistical package SAS 9.3.

**Results** Herbage production and sward white clover proportion (2014-2016) is presented in Table 1. Grass-clover swards produced an additional 1,468 kg DM/ha when compared with GO swards ($P < 0.001$; GO: 15,487 kg DM/ha; GC: 16,954 kg DM/ha). Total herbage DM production differed between years ($P = 0.009$) and sward white clover proportion decreased from 2014 to 2016 by 0.18 ($P < 0.001$). In 2014, every 0.01 increase in sward white clover proportion ($R^2 = 0.257$; $P < 0.001$) increased herbage DM production by 56 kg DM/ha. However, this contribution to herbage DM production was lower in 2015 and 2016 ($R^2 = 0.091$; $P = 0.007$, and $R^2 = 0.003$; $P = 0.607$, respectively).

**Table 1** Herbage production and sward white clover proportion (2014-2016)

| 2014 | Grass-only | Grass-clover | S.E. | *P*-value (clover inclusion) |
|---|---|---|---|---|
| Herbage produced (kg DM/ha) | 14,830 | 17,319 | 341.8 | <0.001 |
| Sward white clover proportion | - | 0.36 | 0.012 | - |
| 2015 | | | | |
| Herbage produced (kg DM/ha) | 16,143 | 17,429 | 423.6 | 0.0036 |
| Sward white clover proportion | - | 0.24 | 0.010 | - |
| 2016 | | | | |
| Herbage produced (kg DM/ha) | 15,487 | 16,114 | 531.0 | 0.0994 |
| Sward white clover proportion | - | 0.18 | 0.011 | - |

**Conclusion** Although white clover inclusion initially increased herbage DM production, this effect decreased as the experiment progressed. This may have been due to the high N fertiliser levels applied throughout the study, which increased the competitive ability of the perennial ryegrass component of the sward.

**Acknowledgements** The authors gratefully acknowledge the Teagasc Walsh Fellowship for financial support.

**References**

Carlsson, G. and Huss-Danell, K. 2003. Plant and Soil 253, 353-372.
Hanrahan, L., Geoghegan, A., O'Donovan, M., Griffith, V., Ruelle, E., Wallace, M. and Shalloo, L. 2017. Computers and Electronics in Agriculture 136**,** 193-201.
Harris, S. and Clark, D. 1996. New Zealand Journal of Agricultural Research 39**,** 149-158.

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# Alpha 1-acid glycoprotein serum concentration on day 7 *post-partum* as a potential prognostic biomarker for the development of purulent vaginal discharge in dairy cattle

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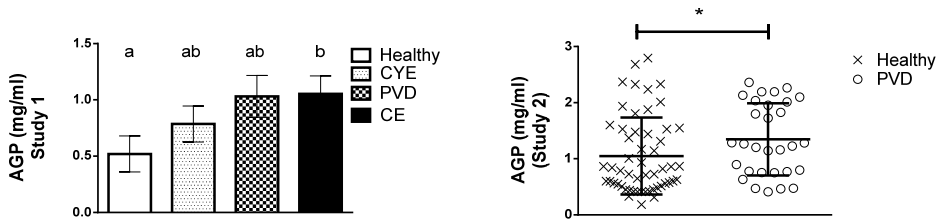
**Application** The prognosis of endometritis (the inflammation of the bovine endometrium) 14 days before diagnosis at 21 days *post-partum* (DPP) would permit early therapeutic intervention, reducing the use of antibiotics and disease impact.

**Introduction** Within the first week post-partum, uterine inflammation is ubiquitous in the dairy cow. If this inflammation is not resolved beyond 21DPP, the cow is said to have developed clinical endometritis (CE) (Sheldon, Lewis, LeBlanc, & Gilbert, 2006). The ability to predict cows at risk of developing uterine disease would facilitate earlier therapeutic intervention and improve the associated negative reproductive outcomes in dairy cows. Previous work by our group identified a greater concentration of the acute phase protein alpha 1-acid glycoprotein (AGP) with increased growth density of recognised uterine pathogens. Therefore, this study aimed to investigate the ability of AGP to identify cows within 7 DPP at risk of developing subsequent uterine disease.

**Material and methods** In Study 1, 60 mixed-parity Holstein-Friesian cows were classified on the basis of their presentation with clinical and subclinical forms of endometritis at 21 DPP. Diagnosis was based on vaginal mucus score (VMS) and percentage of uterine polymorphonuclear (PMN) cells identifying four groups – (i) Healthy (VMS 0, <18% PMN, n=19); (ii) Purulent vaginal discharge only (PVD: VMS ≥2 and <18% PMNs, n=14); (iii) clinical endometritis (VMS ≥2, ≥ 18% PMNs, n=19) and (iv) cytological endometritis only (CYTO: VMS ≤1, ≥ 18% PMNs, n=19). Plasma was obtained at 7 DPP and reproductive data from all enrolled cows was recorded also. In Study 2, serum was collected from 84 mixed-parity Holstein-Friesian cows, diagnosed as healthy (n=54) or PVD (n=30) according to the above criteria at 0 and 7 DPP. AGP was measured in all samples by ELISA kit (Dynex Technologies). PROC TRANSREG was used to normalise the data in SAS 9.4 and PROC MIXED was then used to compute the least means squares of fixed effects. A post hoc Bonferroni correction was included to adjust for multiple comparisons. Statistical significance was set at $P<0.05$.

**Results** In Study 1, AGP concentrations at 7 DPP were similar between healthy group (0.52 ± 0.16mg/ml) and the CYTO group (0.79 ± 0.16mg/ml, P =0.87). This was replicated between the healthy and PVD group (1.03 ±0.19mg/ml, P =0.06). Only the CE (1.05 ± 0.16mg/ml) had greater AGP concentrations than the control group (P =0.03). PVD cows had the longest calving-conception period (data not shown). In Study 2, elevated concentrations of AGP at 7 DPP were detected in the PVD than the control group (1.35± 0.1mg/ml vs. 1.05± 0.1mg/ml respectively; P =0.03).




**Graphs** The graph of study 1 illustrates the heightened AGP concentrations at 7 DPP in the CE group when compared to the healthy group (P <0.01). The graph of study 2 illustrates the divergence of 7 DPP AGP profiles of cows that will be diagnosed as healthy or PVD at 21 DPP (P=0.03).

**Conclusion** Results suggest that increased serum AGP concentration at 7 DPP identifies cows which subsequently develop PVD at 21 DPP. Future work will refine the specificity of AGP for PVD, which is associated with decreased reproductive performance and increased production losses.

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**References**

Sheldon, I. M., Lewis, G. S., LeBlanc, S., and Gilbert, R. O. 2006. Defining postpartum uterine disease in cattle. Theriogenology 65(8), 1516-1530. doi:10.1016/j.theriogenology.2005.08.021

# Prevalence of BVDV, BHV 1, Leptospirosis and Neosporosis, and associated risk factors in 161 Irish beef herds

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**Application** In recent years, the issue of herd health and, in particular, infectious disease has received increasing prominence as a potential factor influencing reproductive efficiency in Ireland and elsewhere.

**Introduction** There are limited data available, in Ireland or elsewhere, to determine the extent of exposure to various endemic diseases among beef cows and factors associated with exposure to causative pathogens. The objectives of this study were to determine the herd and within herd prevalence of BVDV, BHV-1, Leptospirosis and Neosporosis in a large scale study of commercial beef herds on the island of Ireland, and to examine herd level factors associated with exposure to these pathogens in these herds.

**Material and methods** In all, 161 beef cow herds from across the island of Ireland participated in this study. Over 6000 cows were serologically tested for BVDV, BHV-1, Leptospirosis and Neosporosis. Sera were screened using commercially available ELISA kits for BVDV (Svanovir BVD – Ab), BHV-1 (IDEXX IBR gE kit and IDEXX IBR gB X3) , Leptospirosis (PrioCHECK *L.hardjo* antibody kit) and Neosporosis (IDEXX Neospora antibody kit) in Department of Agriculture, Food and the Marine Regional Veterinary Laboratory service (RVL). In each case the tests were carried out according to the manufacturer's instructions. Descriptive statistics and linear regression analysis are presented, with herd prevalence of each of the four pathogens as the outcome variables of interest.

**Results** The average number of cows tested per herd was 35.5 (median 30). Herd level seroprevalence to Bovine Herpesvirus-1(BHV-1), Bovine Viral-Diarrhoea Virus (BVDV), Leptospirosis and Neosporosis was 90%, 100%, 91% and 67%, respectively, while the mean within herd prevalence for the these pathogens was 40%, 77.7%, 65.7% and 5.7%, respectively. The study confirms that the level of seroconversion for the four pathogens of interest increases with herd size. There was also evidence that exposure to one pathogen may increase the risk of exposure to another pathogen.

**Conclusion** Herd level seroprevalences were in excess of 90% for BVD, BHV-1 and Leptosporosis. Larger herds were subject to increased exposure to disease pathogens. This study suggests that exposure to several pathogens may be associated with the further exposure to other pathogens. The role of co-infection has been previously described. Another possible explanation is that infection with one of the pathogens could facilitate infection with another. The role of BVDV, in particular, as an immunosuppressive agent is well documented.

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**References**

Tiwari, A., VanLeeuwen, J.A., Dohoo, I.R., Stryhn, H., Keefe, G.P. and Haddad, J.P. 2005. Veterinary Microbiology 109, 147–158.
Ryan, E.G., Leonard, N., O'Grady, L., More, S.J. and Doherty, M.L. 2012. Irish Veterinary Journal 65, pp.8
Cowley, D.J., Clegg, T.A., Doherty, M.L., More, S.J. 2011. Acta Vet Scandinavica 53:40 doi: 10.1186/1751-0147-53-40.

# Identification of milk whey protein biomarkers of early pregnancy status in dairy cattle

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**Application** Several potential protein biomarkers of early bovine pregnancy (day 21) were discovered in (i) milk whey and (ii) extracellular vesicle (EV) enriched milk whey samples, using a global, label free, proteomics approach.

**Introduction** Current bovine pregnancy detection methods include rectal palpation, ultrasound scanning, Pregnancy Associated Glycoprotein ELISA assays, Pregnancy Specific Protein B ELISA assays, progesterone immunoassays, in-line progesterone testing and visual observation of oestrus (often with the use of aids to detection; e.g. tail paint/heat detection pads) (Balhara, *et al*., 2013). However, these assays suffer from limited sensitivity and results from these pregnancy test methods are not reliable until at least day 28 to 35 post artificial insemination (AI). Therefore, there is a need for the development of an early, reliable, pregnancy test, which is valid on or before day 21 of pregnancy, and would allow producers the opportunity to rebreed at the next oestrus. Ideally, this pregnancy test would utilise milk as the preferred analyte, as a sample can be easily obtained during routine milking and consequently, will not impose any additional stress on the cows.

**Material and methods** Ten dairy cows were initially oestrous synchronised, they went through one (control) oestrous cycle and were artificially inseminated during the following oestrus. Milk whey was collected on day 21 of the control oestrous cycle and on day 21 post AI. All cows were confirmed pregnant by ultrasound scanning on day 35 post AI. A Q-Exactive was used to analyse milk whey samples, and milk whey samples which were EV enriched using IZON Science qEV size exclusion columns, by liquid chromatography tandem mass spectrometry (LC-MS/MS). Subsequent analyses of the label-free quantitative data were performed in MaxQuant and Perseus.

**Results** The APOB, SPADH1and PLIN2 proteins were more abundant in milk whey from dairy cows at day 21 of pregnancy compared with day 21 of the control oestrous cycle ($P<0.05$) (Table 1). Furthermore, PIGR, PGD, QSOX1, MUC1, SHPRA and MD2 were more abundant in extracellular vesicle (EV) enriched milk whey at day 21 of pregnancy compared with day 21 of the oestrous cycle ($P<0.05$) (Table 1).

**Table 1** Proteins which were more abundant on day 21 of pregnancy compared with day 21 of the oestrous cycle in milk whey and EV enriched milk whey

| Protein ID | Sample type | Fold change* |
|---|---|---|
| APOB | Milk whey | 1.48 |
| SPADH1 | Milk whey | 1.36 |
| PLIN2 | Milk whey | 1.19 |
| PIGR | EV enriched milk whey | 1.34 |
| PGD | EV enriched milk whey | 1.30 |
| QSOX1 | EV enriched milk whey | 1.25 |
| MUC1 | EV enriched milk whey | 1.24 |
| SRPRA | EV enriched milk whey | 1.19 |
| MD2 | EV enriched milk whey | 1.18 |

*fold change increase on day 21 of pregnancy compared with day 21 of the oestrous cycle

**Conclusion** The milk whey proteins (APOB, SPADH1, PLIN2, PIGR, PGD, QSOX1, MUC1, SHPRA and MD2) have been identified as potential biomarkers of early pregnancy in dairy cows. As the fold change differences were small, they may be used, in combination with additional markers, for the development of an early pregnancy test for dairy cows.

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**References**

Balhara, A.K., Gupta, M., Singh, S., Mohanty, A.K., Singh, I. 2013. The Scientific World Journal 2013, 1-10

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# The effect of synthetic preimplantation factor on the bovine endometrial transcriptome

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**Application** The present study identifies how preimplantation factor (PIF) interacts with the bovine endometrial transcriptome, paving the way for future research to investigate the role of PIF in bovine pregnancy.

**Introduction** Preimplantation factor (PIF) is a novel peptide secreted from viable embryos as early as the 2-cell stage. Interaction of synthetic PIF (sPIF) with the human endometrium has been characterised, indicating the sPIF interacts with human endometrial stromal cells in culture. Synthetic PIF works to aid in the human embryo implantation process and acceptance by the maternal immune system (Paidas *et al*., 2010). There is limited knowledge related to bovine PIF, although a recent study identified that sPIF reduced native interleukin-6 secretion from bovine endometrial tissue in culture (Wonfor *et al*., 2017). The present study aimed to improve the current knowledge on the interaction of sPIF with the bovine endometrium through RNA-sequencing.

**Material and methods** Endometrium from bovine heifer uteri (n=7), in the follicular stage of the oestrous cycle, were sampled and intercaruncular tissue explants from each replicate were cultured with or without sPIF (100 nM) for 24 hours. Medium was replaced with fresh medium with or without sPIF (100 nM) for a further six hours. At the end of the incubation, tissue was stored in RNAlater. Total RNA was then extracted and quality assessed. Extracted RNA that was of a suitable quality was subjected to library preparation for RNA sequencing on the Illumina HiSeq 2500 platform (Illumina, San Diego, USA). Samples were sequenced in two different runs. Following sequencing, a previously described data analysis workflow was adapted for the sample set to determine differentially expressed genes (DEG) and biological pathways modulated by sPIF treatment (McCabe *et al*., 2012). Sequencing data was assessed for the effect of the treatment as well as the sequencing run. A p-adjusted value of <0.1 was used to determine statistically significant differential expression in the transcript data set. STRING (version 10.5) was used to assess protein to protein interactions, gene ontology (GO) category enrichment and KEGG pathway enrichment. A p-adjusted value of less than 0.05 was used to determine statistically significant over-enrichment of categories or pathways.

**Results** A total of 102 DEG were identified with 78 down-regulated and 24 up-regulated following treatment with sPIF; however, none were DEG over a 4-fold change in expression. There was a strong influence of animal replicates on the data variance, with the native gene expression differing between animals. There was no effect of sequencing run on the data ($P>0.1$). Two 'Cellular Component' GO categories were over represented following sPIF treatment: 'plasma membrane' and 'cell periphery' ($P<0.05$). Twenty-five KEGG pathways were over represented following sPIF treatment. Of interest to the immune function of sPIF, the two most significantly over represented pathways were 'TNF signalling pathway' ($P<0.001$) and 'NF-kappa β signalling pathway' with sPIF treatment downregulating 11 and 9 genes, respectively ($P<0.001$).

**Conclusion** Synthetic PIF interacts with the bovine endometrial transcriptome, although the response appears to be relatively weak compared to that identified in humans, with 20 out of >500 DEG being significantly changed >10-fold in human tissue (Paidas *et al*., 2010). Pathways identified have links to the immune response of the endometrium, suggesting that in the bovine endometrium, sPIF may also have an immune modulatory role. The tissue used in the present study was cyclic intercaruncular tissue; sPIF may have a greater effect on caruncular tissue or in combination with other pregnancy signalling molecules, such as interferon-τ.

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**References**

McCabe, M., Waters, S., *et al*., 2012. BMC Genomics 13, 193.
Paidas, M.J., Krikun, G., *et al*., 2010. American Journal of Obstetrics and Gynecology 202, 459 e1-8.
Ramu, S., Stamatkin, C., *et al*., 2013. Reproductive Biology and Endocrinology 11, 105.
Stamatkin, C.W., Roussev, R.G., *et al*., 2011. Reproductive Biology and Endocrinology 9, 63.
Wonfor, R.E., Natoli, M., *et al*., 2017. Theriogenology 102, 174-182.

# Taurine supplementation pre-freeze and post-thaw improves integrity and reduces oxidative stress in cryopreserved ram spermatozoa

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**Application** Investigating the optimal timing (pre-freeze, or post-thaw) and concentration of taurine supplementation can aid the development of ram sperm cryopreservation protocols to reduce oxidative stress and improve sperm integrity.

**Introduction** Reactive oxygen species (ROS) are required to regulate sperm capacitation, the acrosome reaction (AR) and improve fertilization rates (Aitken, 1997 and Kothari *et al*., 2010), however during cryopreservation ROS production increases due to oxidative stress. This increased ROS production has been associated with increased lipid peroxidation and subsequent loss of sperm function (Cassani *et al*., 2005). Antioxidants have shown some success in mitigating oxidative stress and taurine supplementation during cryopreservation improved some post-thaw sperm parameters (Banday, *et al*., 2017). This study aimed to optimise the concentration and timing of taurine supplementation during the cryopreservation of ram spermatozoa and determine the effect of treatment on sperm integrity and function.

**Material and methods** Ejaculates (n = 9) were collected from three Texel rams (n = 3/ram) by artificial vagina during the breeding season and cryopreserved individually in tris-citrate-glucose cryodiluent supplemented with taurine (0, 0.5, or 1.0 mg/ml) added pre-freeze (PF) or post-thaw (PT) generating seven treatments: 1) control 0 mg/ml, 2) PF 0.5 mg/ml, 3) PF 1 mg/ml, 4) PT 0.5 mg/ml, 5 )PT 1.0 mg/ml, 6) PF + PT 0.5 mg/ml and 7) PF + PT 1 mg/ml. The penetration of spermatozoa through artificial cervical mucus was assessed via vanguard distance at 0min and 60 min post-thaw. At 0, 30, 60 and 180 min post-thaw motility (subjectively via light microscopy), viability and acrosome integrity (propidium iodide and FITC-PNA stain with fluorescent microscopy) and ROS production (NBT assay and light microscopy) were measured. ROS production (% of 200 sperm) was assessed as the proportion of the sperm head that was positive for formazan (0, <50, >50 and 100%).

**Results** Compared to the control, all measures of sperm integrity improved with the addition of taurine ($P < 0.001$). Taurine added both PF+PT (0.5 and 1.0 mg/ml) had the most significant effect improving motility, acrosome integrity, viability, penetrability and % spermatozoa with <50% ROS post-thaw at almost all time points (table 1, $P < 0.001$). Supplementing with 1mg/ml taurine either PF or PT had an intermediate effect on the sperm parameters assessed, whilst 0.5mg/ml taurine PF or PT improved sperm function significantly but marginally.

**Table 1** Motility, viability, acrosome integrity, penetrability and ROS expression (mean ± SEM) in control and treated (1mg/ml taurine PF +PT) ram spermatozoa at 30 and 60 min post-tha

| Treatment | Time | Motility (%) | Viable (%) | Acrosome Intact (%) | Penetrability (cm) | ROS <50 (%) |
|---|---|---|---|---|---|---|
| Control | 30 min | 27.2 ± 1.470 | 41.3 ± 1.167 | 45.7 ± 0.866 | | 37.6 ± 1.029 |
| | 60 min | 13.0 ± 1.167 | 30.1 ± 1.620 | 33.6 ± 1.501 | 1.4 ± 0.066 | 30.3 ± 1.434 |
| PF+PT 1mg/ml | 30 min | 49.3 ± 0.957 | 57.8 ± 0.830 | 62.0 ± 0.928 | | 39.4 ± 0.689 |
| | 60 min | 38.1 ± 0.889 | 53.4 ± 0.959 | 56.8 ± 1.211 | 4.0 ± 0.076 | 35.2 ± 0.983 |

**Conclusion** This study determined the importance of antioxidant supplementation pre-freeze and post-thaw in cryopreserved ram spermatozoa. Concentrations of 1mg/ml were optimal and may protect the sperm from oxidative stress during cryopreservation (pre-freeze supplementation) and thawing (post-thaw). These findings may aid the development of cryopreservation protocols to improve sperm integrity post-thaw.

**References**

Aitken, R.J. 1997. Molecular Human Reproduction 3(3), 169-173.
Banday, M.N., Lone, F.A., Rasool, F., Rashid, M., and Shikari, A. 2017. Cryobiology 74, 25-30.
Cassani, P., Beconi, M.T., and O'Flaherty, C. 2005. Animal Reproduction Science 86(1), 163-173.
Kothari, S., Thompson, A., Agarwal, A., and du Plessis, S.S. 2010. Indian Journal of Experimental Biology s48(5), 425-35.

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# Effect of diet during the rearing and finishing phase on body growth and scrotal development of Holstein-Friesian bulls

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**Application** Ensuring optimal testicular growth is important for maximising semen production potential in bulls. Our data demonstrate that there is a strong relationship between body weight and scrotal circumference. The effects of an increased plane of nutrition during rearing on testicular development will remain into the finishing phase regardless of regime. This work further highlights the importance of early life nutrition on the sexual maturation of the bull.

**Introduction** Testicular development, measured as scrotal circumference is commonly used to define puberty, as proxy of reproductive potential in the bull and is influenced by early life plane of nutrition and thus growth rate (Byrne *et al*., 2016; Dance *et al*., 2015). There is a direct relationship between bodyweight and scrotal size (Coulter and Foote, 1977). The aim of this study was to characterize the effects of commonly used rearing and finishing strategies on scrotal growth in Holstein-Friesian bulls within the context of a pasture based production system.

**Material and methods** Spring-born HF bull calves (n = 69) were assigned at 111 (s.d. 10.4) days of age to one of two levels of concentrate supplementation (1 kg or 2 kg per head daily) during the first season at pasture. Bulls were housed at approximately 40 weeks of age after which they were reassigned within rearing treatment to one of two finishing diets (i) concentrate and barley straw offered on an *ad libitum* basis or (ii) grass silage offered *ad libitum* plus 5 kg concentrate per head daily. Bulls were group fed in slatted floor pens. Concentrate consisted of 580 g/kg barley, 260 g/kg beet pulp, 100 g/kg soya bean meal, 40 g/kg molasses and 20 g/kg minerals. Beginning at 36 weeks of age, bulls were weighed and scrotal circumference (SC) was measured monthly until 64 weeks of age. Bull age at collection of these data was recorded. Data were analysed using a repeated measures ANOVA (PROC MIXED, SAS v9.4). Level of concentrate supplementation, finishing diet and age at collection with their interactions, as appropriate, were included in the statistical model. The relationship between body weight and SC was modelled using regression (STEPWISE procedure) including fixed effects for rearing and finishing diets and age included as a co-variate. Results are presented as mean ± SEM, unless stated otherwise.

**Results** There was no interaction of supplementation level x finishing strategy on bodyweight. Bulls offered 2 kg compared with 1 kg during rearing were heavier at housing, as were bulls on concentrate *ad libitum* during finishing. Scrotal circumference was larger in bulls offered concentrate *ad libitum* vs a moderate finishing strategy following 2 kg concentrate during the first grazing season. There was no interaction of supplementation level x finishing strategy on SC. Finishing strategy had no effect on SC following 2 kg in the first grazing season per head daily; supplementation of 2 kg per head daily followed by a moderate finishing strategy resulted in a larger SC than supplementation of 1 kg per head daily followed by a moderate finishing strategy. Regression analysis showed that 83% of the variation in SC was accounted for by bodyweight (y = 19.75247 + 0.04217x; r = 0.8275; P < 0.001).

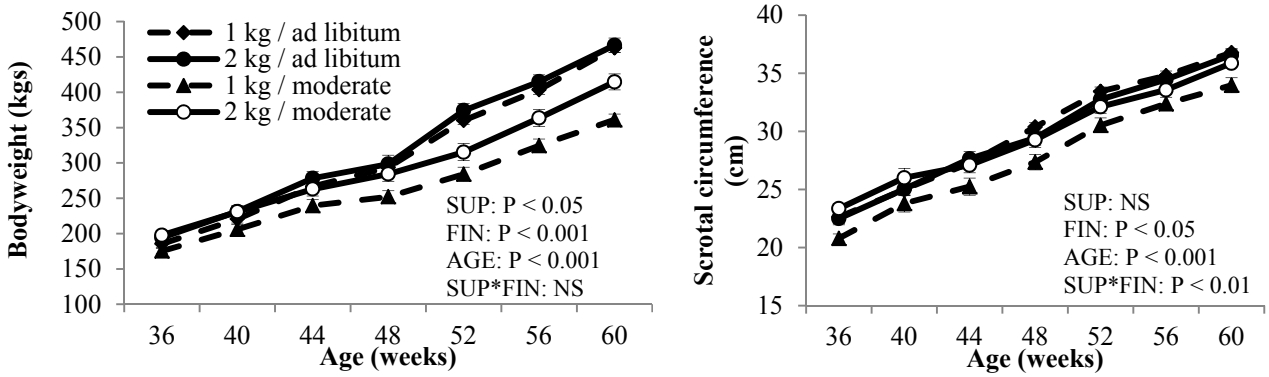


**Figure 1** Effect of diet during the rearing and finishing phase on bodyweight and scrotal circumference of Holstein-Friesian bulls

**Conclusion** Bulls offered 2 kg of concentrate during the first grazing season were heavier than bulls offered 1 kg. Offering 2 kg of concentrate per head daily during the first grazing season maintains scrotal growth irrespective of diet during the finishing phase. From the regression analysis, it was clear that bodyweight is closely related to SC in Holstein-Friesian bulls; therefore, increasing bodyweight by increasing plane of nutrition also leads to an increase in SC.

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**References**

Byrne, C.J., English, A.M., Fair, S., Lonergan, P., Kenny, D.A. 2016. Theriogenology 86 (suppl), 13.
Coulter, G. H., Foote, R. H., 1977. Journal of Animal Science 44, 1076-1079.
Dance, A., Thundathil, J., Wilde, R., Blondin, P., Kastelic, J. 2015. Journal of Dairy Science 98, 987-998.

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# Effect of number of cows in oestrus simultaneously on activity of dairy cows in oestrus

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**Application** In the dairy industry one of the major problems is poor oestrus detection. Developing a better understanding of factors that affect activity of cows in oestrus will improve the accuracy of oestrus detection.

**Introduction** Poor oestrous expression is considered the main contributing factor to poor reproductive efficiency in dairy cows where AI is used. Standing to be mounted is the primary sign of oestrus. However, only 50% of dairy cows are reported to show this sign (Van Eerdenburg *et al*., 2002). Now many dairy farms use activity monitors as an aid to oestrus detection. One of the factors contributing to the extent of oestrous expression is the number of cows in oestrus simultaneously (Sexual Group=SG). The aim of the present study was to investigate the effect of SG on activities such as the number of steps and lying time (h/d) during oestrus.

**Materials and methods** Lactating Holstein Friesian cows (n=92) were used for the study from 14 days post-partum from Feb-2015 to Feb-2016 (BCS 2.78±0.2 (mean±SEM), mobility score 2.13±0.5 and milk yield 31.6±9.3 kg/d) at the Harper Adams University (HAU) dairy unit. Cows were milked twice a day starting approximately 04:30 and 15:30 through a 40-point internal rotary milking parlour. Cows were housed in a free stall yard (cubicles 2.7x1.2 m) with sawdust covered, rubber mattresses and grooved concrete passageways. Cows were fed a total mixed ration *ad libitum* providing DM 43.5 %, ME 12.3 MJ/kg DM, CP 17.4% DM, NDF 33.5% DM, Oil 5% DM and Starch 23.1% DM. Cows were put out to graze during the day (10:00 to 14:15) in the summer and continuously housed during winter. Milk samples (40ml) were collected 3 times a week and stored at 4 ºC until analysis for progesterone concentration by ELISA (Ridgeway Science Ltd., UK). A cow was considered in oestrus when progesterone concentration in milk was <3 ng/ml, followed by an increase to >15 ng/ml. Activity was monitored continuously using IceQubes (IceRobotics Ltd., Edinburgh, UK) attached to the back left leg of each cow. The period during which activity increased to >80% above the mean activity for the preceding 3 days was considered the duration of oestrus (López-Gatius *et al*., 2008). The effects of group and time on the activity measures considered were analysed by repeated measures ANOVA with Tukey post hoc tests (GenStat 17th edition). HAU Research Ethics Committee approved the protocol.

**Results** Number of steps increased (Table 1), and lying times (h/d) were reduced significantly (P<0.001) on the day of oestrus (day 0) compared to 3 day before and 3 day after oestrus. Significantly (P<0.001) more steps and lower lying time were recorded when three or more cows were in oestrus simultaneously (SG3+) compare to when one cow was in oestrus (SG1). Moreover, significantly (P<0.001) more number of step were taken when two cows were in oestrus (SG2) compare to SG1. Lying time were lower on day of oestrus, but not significantly different between SG1, SG2 and SG3+ on the day of oestrus. There was a significant interaction between SG and time for both the number of steps/d and lying time (h/d).

**Table 1** Mean number of steps/d and lying time (h/d) 3 days before, on day of oestrus (0) and 3 days after oestrus and between the numbers of cows in oestrus simultaneously. One cow in oestrus (SG1; n=36), two cows in oestrus (SG2; n=50) and three or more cows in oestrus (SG+3; n=52).

| Activity | SG | Days from oestrus | | | | | | | SED | P value | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | -3 | -2 | -1 | 0 | 1+ | 2+ | 3+ | | SG | Days | SG X days |
| Steps per day | 1 | 1195 | 1207 | 1212 | 2923 | 1487 | 1369 | 1209 | 178.6 | <0.001 | <0.001 | <0.001 |
| | 2 | 1534 | 1487 | 1768 | 4113 | 1766 | 1524 | 1464 | | | | |
| | 3+ | 1959 | 1876 | 2090 | 5670 | 2307 | 2014 | 1980 | | | | |
| Lying time h/d | 1 | 10.0 | 10.0 | 10.4 | 7.6 | 10.8 | 10.8 | 10.7 | 0.518 | 0.954 | <0.001 | 0.005 |
| | 2 | 10.6 | 10.4 | 9.8 | 6.6 | 11.4 | 11.2 | 11.0 | | | | |
| | 3+ | 10.5 | 10.7 | 10.0 | 6.5 | 11.2 | 10.9 | 10.6 | | | | |

The duration of oestrus was significantly (P=0.004) longer in SG3+ (11.5±0.5h) compare to SG1 (9.1±0.4h); however, there was no significant differences in comparison to SG2 (10.1±0.4h). Significantly more steps were taken during the peak hour of oestrus in all SG groups (SG3+ 780.0 ±44.7, SG2 620.4±34.7, SG1 315.9±29.9 steps, P<0.001) compared to the 12 hours before and 12 hours after the peak of oestrus.

**Conclusion** Dairy cows in oestrus spent more time active at the expense of the lying time during the day of oestrus. The intensity of oestrus is greater and oestrus lasts longer with increasing numbers of cows in oestrus at the same time.

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**References**

López-Gatius, F., Mirzaei, A., Santolaria, P., Bech-Sàbat, G., Nogareda, C., García-Ispierto, I., Hanzen, C. and Yániz, J.L. 2008. Theriogenology 69, 1095-1103.
Van Eerdenburg, F.J.C.M., Karthaus, D., Taverne, M.A.M., Mercis, I. and Szenci, O. 2002. Journal of Dairy Science 85, 1150-1156.

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# Survey of breeding and reproductive management practices undertaken on suckler beef cow herds on the island of Ireland (2015-2016)

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**Application** Reproductive management measures undertaken in beef cow herds were determined in a survey (2015-2016).

**Introduction** Poor reproductive management has often been implanted as one the main reasons for the declining reproductive performance of the beef cow herd. In seasonal pasture based suckler beef production systems, reproductive efficiency is the main determinant of production and economic efficiency, requiring cows to calve compactly close to the onset of the grazing season. The current reproductive parameters for the suckler beef cow herd indicate and average inter-calving interval of 400 days (target 365 days), only 0.83 calves produced/cow/ year and less than 25 % of heifers calving for the first time between 22 and 26 months of age (Irish Cattle Breeding Federation). Indeed these statistics have grave implications for the future sustainability of suckler beef production throughout the island of Ireland. Whilst national statistics are useful, we have no information on the management practices or decisions underpinning this significant reproductive inefficiency at herd level. Therefore, the objective of the survey was to quantify the uptake of existing and new reproductive technologies as well as reproductive management practices on suckler beef cow herds on the island of Ireland.

**Material and methods** The survey was run from March 2015 to July 2016. Participation in the survey was voluntary and was not incentivised. The survey was carried out at Knowledge Transfer (KT) discussion groups, and also at suckler beef open days organised by Teagasc/AFBI. The total study population was 547 (representing 32 counties on the island of Ireland). Of these, 10 surveys could not be used due to substantial data gaps. For quality control purposes all participants were required to supply either a herd number or name on the survey. All participants were required to have breeding beef animals in their herd at the time of completion. Information on farmer age and herd size was also collected. For the purposes of this summary, questions relating to management of replacement heifers, reasons for culling and uptake of new and existing reproductive technologies were examined.

**Results** Quantified responses to questions relating to the uptake of new and existing reproductive technologies as well as general reproductive management are given in Table 1. The mean (range in parentheses) number of spring calving and autumn calving cows per herd was 34 (1-200) and 23 (1-220), respectively. The mean replacement rate (heifers only) was 19% (0-60%). A total of 53% of respondents breed replacements without purchasing any additional females. A total of 10 and 8% of respondents, respectively, purchase replacement heifers as calves/weanlings, or immediately before breeding. Animal age, poor fertility performance and poor breeding ability (ability to breed quality calves) were three of the main reasons cited by herd owners as to why cows were culled from the beef cow herd. Two of the main reasons cited for not using AI were heat detection, having a stock bull and not needing to use AI, and land fragmentation.

**Table 1** Quantified responses given to questions relating to the uptake of existing and new technologies as well as general breeding management practices

| Question | Respondents | Yes % |
|---|---|---|
| General breeding management | (n =) | |
| Use heat detection aids -Yes/No | 237 | 37 |
| AI usage (any level) - Yes/No | 512 | 65 |
| Ultrasound scan following breeding - Yes/No | 389 | 83 |
| Sexed semen usage (any level) -Yes/No | 380 | 7 |
| Commence breeding heifers before 17 months | 510 | 42 |
| Commence breeding heifers after 24 months | 510 | 13 |
| Weigh heifers before breeding -Yes/No | 377 | 27 |

**Conclusion** The results from this survey provide an important insight into breeding and reproductive management practices on suckler beef cow herds in Ireland. Thus, based on the findings of this survey there is a requirement on all stakeholders across the beef industry to bridge the 'KT gap' if the production targets of Food Wise 2025 are to be achieved.

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**References**

Irish Cattle Breeding Federation. Beef Calving Statistics 2017.

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# *Leptotrichiaceae* identified in *post-mortem* cranial lung lobe lesions and mediastinal lymph node tissue obtained from calves with respiratory disease

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**Application** A novel bacterial species within the *Leptotrichiaceae,* family, which may play a pathogenic role in bovine respiratory disease (BRD), has been identified in *post-mortem* lesioned lung tissue and the corresponding mediastinal lymph node tissue using an amplicon sequencing assay.

**Introduction** Viral and mycoplasmal agents are responsible for the onset of BRD. They predispose the lungs to colonisation by bacterial pathogens which advance the disease (Griffin, *et al*., 2010). Current culture-based and PCR identification techniques cannot detect unknown or unculturable pathogens. Therefore, the objective of this study was to develop a single universal assay, based on phylogenetic (16S ribosomal RNA (rRNA) gene) PCR amplicon sequencing, with potential to accurately identify and differentiate bacteria in *post-mortem* lung and lymph node tissue samples from fatal BRD cases.

**Material and methods** Cranial lung lobe and corresponding mediastinal lymph node *post-mortem* tissue samples were collected from calves diagnosed as BRD cases by veterinary laboratory pathologists (Figure 1) and from clinically healthy calves. Gene amplicon libraries, targeting the V3-V4 region of the bacterial 16S rRNA gene were prepared and sequenced on an Illumina MiSeq. Quantitative insights into microbial ecology (QIIME) was used to determine operational taxonomic units (OTUs) which corresponded to the 16S rRNA gene sequences.

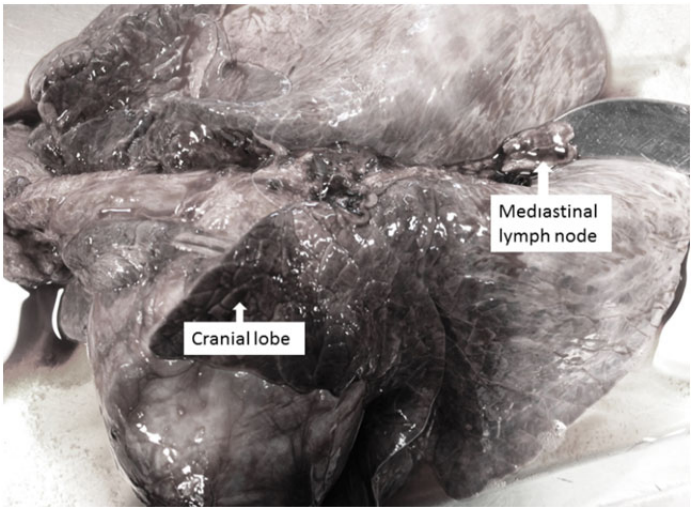


**Figure 1** Cranial lung lobe and mediastinal lymph node tissues from a fatal BRD case

**Results** *Leptotrichiaceae, Mycoplasma, Pasteurellaceae*, and *Fusobacterium* were the most abundant OTUs identified in the lungs and lymph nodes of the calves which died from BRD. *Leptotrichiaceae, Fusobacterium, Mycoplasma, Trueperella* and *Bacteroides* had greater relative abundances in *post mortem* lung samples collected from fatal cases of BRD, compared with clinically healthy calves without lung lesions. *Leptotrichiaceae, Mycoplasma* and *Pasteurellaceae* showed higher relative abundances in *post-mortem* lymph node samples collected from fatal cases of BRD, compared with clinically healthy calves without lung lesions.

**Conclusion** Species within the *Leptotrichiaceae* family are not currently associated with BRD but this family was the most abundant OTU identified and it was not present in the lesion-free lungs of the healthy calves which suggests that it may play a pathogenic role in BRD. The OTU sequence for *Leptotrichiaceae* was not identical to any known genus and appears to represent a novel species.

**Acknowledgements** The authors gratefully acknowledge funding from (DAFM) Research Stimulus Funds 11/S/131 and 11/S/322, and EU project: 311825.

**References**

Griffin, D., Chengappa, M.M., Kuszak, J., McVey, D.S. 2010. Veterinary Clinics of North America: Food Animal Practice 26, 381-394.

# Quantifying antimicrobial drug usage in calves from birth to 6 months of age on Irish suckler beef and dairy farms

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**Application** Baseline data are now available on antimicrobial drug usage in calves on commercial farms in Ireland.

**Introduction** Concern about the use of antimicrobials in food producing animals is increasing. Our aim was to quantify antimicrobial drug usage in calves using health treatment records from Irish suckler beef and dairy farms.

**Material and methods** Health treatment records for calves born between July 1, 2014 and June 30, 2015 on 79 suckler beef and 44 dairy farms were analysed. Calves were followed from birth until 6 months of age. According to standard farm protocol, calves exhibiting clinical signs of any disease were identified and antimicrobial treatment was administered. Farmers recorded the following information for each treatment administered: calf identification, age at treatment, disease event, drug name, number of treatment days, and amount of drug administered. Summary of product characteristics (SPC) were retrieved from the Health Products Regulatory Authority and drugs were classified by active substance(s). Guidelines were recently published on technical units and appropriate indicators for quantification of antimicrobial usage (Collineau *et al*., 2017). Defined daily dose for animals (DDDvet) and defined course dose for animals (DCDvet) were the technical units used to measure antimicrobial consumption. DDDvet represents the assumed average dose per kg animal per species per day. DCDvet represents the assumed average dose per kg animal per species per treatment course. Treatment incidence (TI) was the indicator used to quantify antimicrobial usage, and the calculations applied were:

$$TI_{DDDvet} = \frac{\text{total active substance administered}}{\text{DDDvet x standard BW x total calf-days}} \times 1000 \qquad TI_{DCDvet} = \frac{\text{total active substance administered}}{\text{DCDvet x standard BW x total calf-days}} \times 1000$$

Total amount of active substance administered was determined from the health treatment records. DDDvet and DCDvet for cattle, as assigned by the European Medicines Agency, were used in the calculations. Standard body weight (BW) at treatment was 135 and 108 kg for suckler beef and dairy calves, respectively. Total number of calf-days at risk was determined using movement data from the Animal Identification and Movement central database.

**Results** In total, 3,204 suckler beef calves and 5,358 dairy calves, representing 540,953 and 579,997 calf-days at risk, respectively, were included in the study. A total of 1,770 antimicrobial treatments were administered to suckler beef (n = 841) and dairy calves (n = 929) between birth and 6 months of age. Antimicrobial drug usage is summarised for suckler beef and dairy calves in Table 1. There was large variation in $TI_{DDDvet}$ and $TI_{DCDvet}$ by farm.

**Table 1** $TI_{DDDvet}$ (per 1,000 calf-days at risk) and $TI_{DCDvet}$ (per 1,000 calf-days at risk) for the ten most frequently administered active substances to suckler beef and dairy calves from birth to 6 months of age

| Suckler beef calves (n = 3,204) | | | | | Dairy calves (n = 5,358) | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Active substance | Route[1] | n[2] | $TI_{DDDvet}$ | $TI_{DCDvet}$ | Active substance | Route | n | $TI_{DDDvet}$ | $TI_{DCDvet}$ |
| Marbofloxacin | P | 145 | 0.603 | 0.244 | Florfenicol | P | 159 | 0.482 | 0.157 |
| Florfenicol | P | 128 | 0.534 | 0.174 | Enrofloxacin | P | 106 | 0.748 | 0.196 |
| Amoxicillin | P | 110 | 0.831 | 0.238 | Amoxicillin | O | 100 | 0.085 | 0.021 |
| Oxytetracycline | P | 95 | 0.572 | 0.162 | Marbofloxacin | P | 74 | 0.180 | 0.073 |
| Amoxicillin | O | 81 | 0.100 | 0.025 | Sulfadiazine, trimethoprim | O | 59 | 0.106 | 0.022 |
| Enrofloxacin | P | 57 | 0.212 | 0.056 | Neomycin | O | 57 | 0.153 | 0.038 |
| Neomycin | O | 34 | 0.073 | 0.018 | Oxytetracycline | P | 56 | 0.556 | 0.157 |
| Sulfadiazine, neomycin | O | 32 | 0.076 | 0.015 | Chlortetracycline | O | 56 | 0.019 | 0.003 |
| Sulfadoxine, trimethoprim | P | 26 | 0.098 | 0.027 | Sulfadimidine | O | 54 | 0.279 | 0.066 |
| Sulfadiazine, trimethoprim | P | 24 | 0.081 | 0.029 | Tetracycline | O | 48 | 0.040 | 0.009 |

[1]Route of administration: parenteral (P) or oral (O); [2]number of treatments administered

**Conclusion** A range of injectable and oral antimicrobial products were administered to calves on suckler beef and dairy farms. Further research is investigating the risk factors associated with antimicrobial usage in calves on Irish farms.

**Acknowledgements** This research was supported under the DAFM Stimulus Fund (11/S/131).

**Reference**

Collineau, L., Belloc, C., Stärk, K.D.C., Hémonic, A., Postma, M., Dewulf, J. and Chauvin, C. 2017. Zoonoses and Public Health 64, 165-184.

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# Evaluation of the antimicrobial and prebiotic potential of a casein-derived hydrolysate on porcine bacterial isolates *in vitro*

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**Application** Inclusion of a casein-derived hydrolysate in the diet of weaned piglets could potentially improve the composition of the gut microbiota by increasing beneficial bacteria while reducing *Salmonella enterica* ser. Typhimurium.

**Introduction** Weaning has many negative effects on the functionality of the piglet gastrointestinal tract and the population of residing microbiota. Many important functions for the host are attributed to this microbial community such as competitive exclusion of pathogens, nutrient availability and immunomodulation. Commensal *Lactobacillus spp.* and *Bifidobacterium spp.* have beneficial effects, while members of the *Enterobacteriaceae* family are linked to compromised health. Casein-derived hydrolysates had anti-inflammatory activity in *in vitro* and *ex vivo* experiments and their dietary inclusion led to shifts in the gut microbial populations of weaned piglets (Mukhopadhya *et al.*, 2015 & in press). The aim of this study was to determine if a casein-derived hydrolysate has prebiotic and/or antimicrobial potential *in vitro*.

**Material and methods** *Lactobacillus plantarum*, *L. reuteri, Bifidobacterium thermophilum*, *Escherichia coli* and *Salmonella enterica* ser. Typhimurium were revived from cryoprotective beads and subcultured following standard procedures. All bacterial strains were diluted in 10% medium [de Man, Rogosa and Sharpe (MRS) for lactobacilli and bifidobacteria and Tryptone Soya broth (TSB) for *E. coli* and *S.* Typhimurium] to obtain an inoculum of $10^6$-$10^7$colony-forming unit(CFU)/ml. The test compound was a 5 kDa retentate obtained by filtering a casein-derived hydrolysate (CH5kDaR) using a membrane filter as described in Mukhopadhya *et al*., (2015). A two-fold serial dilution (2 mg/ml-0.25 mg/ml) of CH5kDaR was made in 10% medium. In 96-well microtiter plates, duplicate wells were inoculated with 100 µl of each concentration of the compound and 100 µl inoculum. To confirm sterility, blank wells containing only 10% medium as well as a series of wells containing only the serial dilution of the compound were included. Plates were incubated aerobically for all species except for *B. thermophilum* which was incubated anaerobically at 37 °C for 18 h. Final bacterial concentrations were determined after 10-fold serial dilution, spread plating and incubation at 37 °C for 24 h aerobically or for 48 h anaerobically for *B. thermophilum*. The dilution with 5-50 colonies was selected for the calculation of CFU/ml using the formula CFU/ml=average colony number*50*dilution. All experiments were carried out with technical replicates on three independent occasions. Prior to statistical analysis, all data were logarithmically transformed. The PROC GLM (SAS 9.4) was used to analyse the data. Results are expressed as mean ± standard error.

**Results** An increase of all beneficial bacterial strains was observed as the CH5kDaR concentration increases: *L. reuteri* (≤0.7 logCFU/ml, $p<0.05$), *L. plantarum* (≤0.8 logCFU/ml, $p<0.05$) and *B. thermophilum* (≤0.9 logCFU/ml, $p<0.05$) as shown in Table 1. A reduction of at least 0.5 logCFU/ml in *S. typhimurium* counts was observed at 2 mg/ml and 1 mg/ml concentrations. CH5kDaR had no effect on *E. coli* at any of the concentrations tested.

**Table 3** Bacterial counts following exposure to a serial dilution of CH5kDaR*

| Bacterial strain | Final bacterial concentration (logCFU/ml) | | | | | SEM |
|---|---|---|---|---|---|---|
| | 0 mg/ml | 2 mg/ml | 1 mg/ml | 0.5 mg/ml | 0.25 mg/ml | |
| *L. reuteri* | 7.01$^{a}$ | 7.70$^{d}$ | 7.61$^{d}$ | 7.70$^{d}$ | 7.61$^{d}$ | 0.10 |
| *L. plantarum* | 7.66$^{a}$ | 8.45$^{e}$ | 8.18$^{d}$ | 7.85$^{a}$ | 7.90$^{b}$ | 0.08 |
| *B. thermophilum* | 6.99$^{a}$ | 7.87$^{d}$ | 7.41$^{b}$ | 7.31$^{a}$ | 7.26$^{a}$ | 0.12 |
| *S. Typhimurium* | 9.30$^{a}$ | 8.73$^{b}$ | 8.67$^{b}$ | 8.77$^{a}$ | 8.80$^{a}$ | 0.19 |
| *E. coli* | 9.38$^{a}$ | 9.27$^{a}$ | 9.11$^{a}$ | 8.89$^{a}$ | 8.71$^{a}$ | 0.68 |

* Comparisons were performed between the means of the control (0 mg/ml) vs. each treatment.
The mean significance level is defined by different lowercase letters ($b<0.05$, $d<0.001$, $e<0.0001$).

**Conclusion** The casein-derived hydrolysate displayed both prebiotic and antimicrobial activity *in vitro*. These results indicate its potential use as a dietary supplement in piglets post-weaning, although further research *in vivo* is required.

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**References**

Mukhopadhya, A., Noronha, N., Bahar, B., Ryan, M.T, Murray, B. A., Kelly, P.M., O'Loughlin, I.B., O'Doherty, J.V., and Sweeney, T. 2015. Food and Function 6, 612-21.
Mukhopadhya, A., McAlpine, P., O'Doherty, J.V. and Sweeney, T. 2017. British Journal of Nutrition (in press, under review).

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# Quantitative assessment of faecal and urinary elimination of a critically important antibiotic in dairy cows following subcutaneous administration

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**Application** Not only ceftiofur but also its metabolites should be quantified in manure to determine environmental loading of antibiotics; manure from ceftiofur treated cows should be segregated for at least a week and treated to remove antibiotic residues before releasing into slurry tanks or land application.

**Introduction** There is a growing body of evidence that antimicrobial resistance (AMR) in food producing animals can impact AMR in humans, and transfer of antimicrobial resistant bacteria (ARB) from animals to humans has been documented (Verkade and Kluytmans, 2014). Food producing animals have been considered to be a major source of AMR because of substantial amounts of antimicrobial use primarily for treatment and prevention of diseases. Following antimicrobial therapy antimicrobial residues reach the gut and make the gut environment more conducive for *de novo* emergence and/or enrichment of existing AMR. The impact of antimicrobial exposure stretches beyond the gut to the environment not only because faecal antimicrobial resistance genes (ARG) and ARB can enter the human food chain but also because antimicrobial residues excreted via faeces and urine have the potential to continue to exert selection pressure in the soil and water environment (Tello *et al*., 2012), contributing to the emergence and spread of AMR. Appropriate risk assessment and development of efficient mitigation strategies will be limited without sufficient data on the elimination of antibiotics from animals. The objective of this study was to determine temporal pattern and extent of faecal and urinary excretion of a 3$^{rd}$ generation cephalosporin, a class of antibiotics considered critically important to treat diseases in humans by WHO, in dairy cows following subcutaneous administration.

**Material and methods** Six Holstein cows in their first lactation, 110 to 200 days in milk and yielding 30.45 to 40.45 kg of milk daily, were used in this study. Cows (n = 3) assigned to the ceftiofur treatment were injected subcutaneously with 1.5 mL ceftiofur crystalline free acid sterile suspension (150 mg ceftiofur) per 45.4 kg body weight. Per manufacturer's protocol, a sequence of two injections were spaced 72 hours apart, at the base of the right ear on day 0 and at the base of the left ear on day 3. Faeces and urine were collected (accumulated over a 24 h period) immediately before antibiotic administration (day 0) and on day 1, 2, 3, 4 and 5 after administration of 1$^{st}$ dose. Additional faecal (*per rectum*) and urine (spot sampling) samples were collected on day 7, 14, 21 and 28 following 1$^{st}$ dose. Immediately following collection, faecal and urine samples were stored at -20ºC. Samples were extracted and cleaned by solid phase extraction (SPE), and subjected to UPLC-MS/MS quantification of antibiotics. We used a novel approach to ceftiofur and its derivatives. Ceftiofur is very quickly metabolised in the body, but metabolites or conjugates could be transformed back into parent compound following excretion in manure or in the environment. Therefore, any quantification approach that targets only ceftiofur will underestimate the risk of AMR associated with ceftiofur treatment and subsequent excretion in faeces and urine. We derivatised ceftiofur and its primary metabolites into a stable compound, which was then extracted and quantified (Berendsen *et al*., 2012). Daily faecal and urinary concentrations were reported as means estimated using PROC MEANS in SAS. Excretion data were analysed using the GLIMMIX procedure in SAS (SAS Inst. Inc., Cary, NC) with cow as the experimental unit. The statistical model included day as a fixed effect and cow as a random variable. Orthogonal polynomial contrasts were used to test the linear, quadratic, and cubic effects of day.

**Results** Ceftiofur was not detected in faeces and urine on day 0. Concentration of ceftiofur in faeces decreased from 23.4 ng/g on day 1 to 19.1 ng/g on day 3, but the concentration increased to 34 ng/g on day 5. Urinary concentration of ceftiofur initially decreased until day 3, then reached peak on day 4 and gradually decreased to 7.74 ng/mL on day 14. Excretion via urine was prolonged compared to faecal excretion. Daily faecal excretion (mg per day) of ceftiofur increased linearly ($P$ = 0.04) from 0.69 mg on day 1 to 1.17 mg on day 5. However, daily excretion of ceftiofur in urine tended to follow a quadratic trend ($P$ = 0.07) with a peak (3.53 mg per day) on day 4 *i.e.* day 1 post 2$^{nd}$ dose.

**Conclusion** We have successfully used a novel approach to quantitatively detect ceftiofur in bovine faeces and urine. This method allowed measurement of trace amounts of ceftiofur and its metabolites collectively in faeces and urine from treated cattle, and thus will help assess accurate environmental loading of antibiotics from the livestock industry. Faecal excretion followed a linear trend, but urinary excretion of ceftiofur was prolonged and followed a quadratic pattern. Even though only a minor proportion of total ceftiofur dose was excreted via faeces and urine, the AMR potential of this low level antimicrobial residue should not be ignored.

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**References**

Verkade, E. and Kluytmans, J. 2014. Infection, Genetics and Evolution 21, 523-530.
Tello, A., Austin, B. and Telfer, T.C. 2012. Environmental Health Perspectives 120, 1100-1106.
Berendsen, B.J., Stolker, L.A. and Nielen, M. 2012. Food Additives & Contaminants 292, 197-207.

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# Prevalence of antimicrobial resistant *Escherichia coli* in young broiler chicks

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**Application** The rapid development of a healthy gut microbiome in the growing chick reduces the prevalence of *Escherichia coli* with multiple antimicrobial resistance, although the prevalence of ampicillin resistant *E. coli* remains extremely high.

**Introduction** The growing prevalence of antimicrobial resistance by bacteria is a threat to both human and animal health. A high prevalence of antimicrobial resistant *E. coli* have been observed in day old chicks in Austria (Roth *et al*., 2017). The objective of this experiment was to determine the prevalence of ampicillin resistant *E. coli* in day-old commercial chicks in the UK, and the proportion of these *E. coli* that were also resistant to other antibiotic groups.

**Material and methods** 32 male chicks (Ross 308) were obtained from a commercial hatchery as day old chicks. A sample of the bedding (wood shavings) and feed that the birds were subsequently placed on and fed was taken. A swab from a sample of excreta in the box containing the chicks was also taken. 16 birds were sacrificed as day old chicks, being taken straight from the transport box before exposure to the bedding and feed. The remaining 16 chicks were reared in a single group fed a wheat based starter mash diet. At 15 d, these remaining birds were sacrificed. A swab was taken from the lumen of the duodenum and caecum of each chick, and placed in Amies media for transport to the laboratory. On day 1, the swabs were analysed using the streak plate method onto MacConkey agar plates containing 50 µg/ml ampicillin. On day 15, each swab was placed in nutrient broth (4 ml) and agitated to suspend bacteria. 100 µl of this suspension was then spread onto a plate containing MacConkey agar with ampicillin (50 µg/ml) for duodenal samples. Caecal samples were further diluted (10 µl in 500 µl nutrient broth; 100 µl of this diluted suspension plated out as before). Samples of feed and bedding were diluted (1 g in 10 ml distilled water), and 100 µl was then spread onto a MacConkey agar plate (containing 50 µg/ml ampicillin). Plates were incubated overnight at 37 °C. The following day, plates were assessed as having either no or low (<10 colony forming units, CFU) ampicillin resistant *E. coli* ($Amp^{50}EC$), medium (<100 CFU) or high (too numerous to count) $Amp^{50}EC$. One distinct colony of $Amp^{50}EC$ from each plate was then picked using a sterile plastic inoculating loop and transferred to an Eppendorf tube containing 500 µl MacConkey agar broth (containing 50 µg/ml ampicillin). The tube was vortexed, and then 100 µl suspension was spread evenly onto a MacConkey agar plate. The suspension was allowed to air dry, and then well-spaced antibiotic discs were placed on the plate. Plates were again incubated overnight at 37 °C, and examined the following day for resistance to the other classes of antibiotic (based on the presence or absence of growth around each disc. The antibiotic discs used were gentamicin (GEN, 10 µg, d1), streptomycin (STR, 10 µg, d15); tetracycline (TET, 10 µg, d1 and 15); ciprofloxacin (CIP, 5 µg, d1 and 15); erythromycin (ERY, 15 µg, d1), chloramphenicol (CAM, 10 µg, d15). STR and CAM replaced GEN and ERY respectively as they are more relevant to poultry production, but were unavailable on day 1.

**Results** There was no evidence of $Amp^{50}EC$ in the sample of bedding taken before the birds were placed on it. A few colonies of $Amp^{50}EC$ were detected from the sample of feed, and the colony that was then subsequently incubated was also resistant to ERY. There was a high concentration of $Amp^{50}EC$ in the sample of excreta taken from the transport box, however these were sensitive to all the other antibiotics. There was a medium or high concentration of $Amp^{50}EC$ in eight of the duodenal samples at day 1, and 15 of the 16 caecal samples. By day 15, even with dilution, all duodenal and caecal samples showed a high concentration of $Amp^{50}EC$. There was no evidence of resistance to CIP, GEN or CAM in $Amp^{50}EC$ isolated from the duodenum or caecum at either day 1 or day 15. $Amp^{50}EC$ was not resistant to STR at day 15 either. However, 57% of the duodenal samples and 80% of the caecal samples were resistant to ERY. Resistance to TET was observed in 36% of duodenal samples at day 1, and this increased to 63% at day 15. The corresponding figures for caecal samples were 67% (day 1) and 32% (day 15). At day 1, 36% of duodenal samples and 67% of caecal samples were resistant to two other unrelated classes of antibiotic in addition to ampicillin. Resistance to more than one other unrelated class of antibiotic (in addition to ampicillin) was not observed at day 15.

**Conclusion** There is a high prevalence of ampicillin resistant *E. coli* in both the duodenum and caecum of day-old and 15 d old chicks. The source of this infection appears to be the bird rather than the bedding or feed. Prevalence of cross resistance to other unrelated classes of antibiotic appears to decline as the bird ages, perhaps reflecting the development of its gut microbiome.

**References**

Roth, N., Mayrhofer, S., Gierus, M., Weingut, C., Schwarz, C., Doupovec, B., Berrios, R., Domig, K.J. 2017. Poultry Science 96, 4053-4060.

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# How should passive transfer data for dairy heifer calves be interpreted and used for disease prevention?

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**Application** Techniques to monitor passive transfer of immunity in calves are used for herd health management, but there is poor evidence to associate calf results with disease risk. More reliable interpretation could improve calf health.

**Introduction** Neonatal calves are born without maternal IgG and passive transfer of immunity from colostrum provides important protection against diseases such as neonatal septicaemia (Godden, 2008). A recent UK study showed 26 % of calves had inadequate passive transfer based on a cut-off of 56 mg/ml serum total protein (McFarlaine, 2015). Respiratory disease and diarrhoea are the most common causes of disease and death in dairy heifers and are associated with reduced performance and welfare (Godden, 2008). Farmers are recommended to ensure a serum IgG of at least 10 mg/ml or a serum total protein of at least 50 mg/ml to reduce disease risk (Godden, 2008). However, these cut off points are not empirically derived (Windeyer *et al*., 2014). The current study aims to improve understanding of the relationship between passive transfer and disease risk and provide evidence based advice on what constitutes adequate passive transfer.

**Material and methods** Three cohorts of calves were combined to give a total cohort of 726 dairy heifer calves. All calves had passive transfer assessed in the first week of life by radial-immunodiffusion to detect serum IgG and by refractometer to detect serum total protein. Calf health was recorded by weekly health scoring for 9 weeks by a veterinary surgeon or final year veterinary student. Calf growth was assessed by recording heart girth at weeks 1, 5 and 9. Statistics were completed in R with generalised linear mixed effects models used to assess odds of disease based on passive transfer, with farm and cohort used included as random effects. ROC curves were used to determine cut off points.

**Results** Of the 726 calves 16.5 % (n=120, 95% CI= 13.9-19.5 %) had serum IgG of less than 10 mg/ml, a commonly used cut off for adequate passive transfer. However, disease incidence with very high with 47.5 % diagnosed with diarrhoea (95% CI 43.8-51.2 %) and 39 % of calves diagnosed with respiratory disease (95% CI 36-43.2 %). Diarrhoea was not correlated with IgG or serum total protein. Respiratory disease was associated with both measures of passive transfer. The relationship was linear with 0.97 times the risk of BRD with each unit increase in IgG ($p=0.002$). ROC curves suggested a cut off of 22.9 mg/ml IgG or 57 mg/ml total protein created the fewest misclassification errors to predict respiratory disease from passive transfer results. These cut off values gave a sensitivity of 0.61, a specificity of 0.52 and a positive predictive value of 0.47 suggesting this is a poor method to predict respiratory disease risk. Average daily gain in the first 63 days of life was 0.6 kg/day (IQ range, 0.55-0.76 kg/day). Passive transfer measured by both IgG and total protein was strongly correlated with weight gain. Across the range of observed IgG values there was a 5.4 kg difference in calf weight by 63 days of age ($p<0.001$).

**Conclusion** There is a wide literature relating to passive transfer of IgG as an indicator of calf health (McGuirk, 2008).This data adds to the evidence that neonatal calf scours are not associated with passive transfer (Windeyer *et al*., 2014). Good passive transfer reduces the risk of respiratory disease (Godden, 2008; Windeyer *et al*., 2014) but existing classification in inadequate – calves show a linear response with disease risk reducing across the range of observed IgG or total protein. Passive transfer cut offs show poor ability to discriminate between healthy and sick calves suggesting that adequate/failure of passive transfer may not be a useful classification for calf health. Passive transfer is an important predictor of calf growth.

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**References**

Godden, S.M. 2008. Veterinary Clinics of North America Food Animal Practice 24, 19–39.
MacFarlane, J.A., Grove-White, D.H., Royal, M.D., Smith, R.F. 2015. Veterinary Record 176, 625.
McGuirk, S. 2008. Veterinary Clinics of North America Food Animal Practice 24, 139–53.
Windeyer, M.C., Leslie, K.E., Godden, S.M., Hodgins, D.C., Lissemore, K.D., Leblanc, S.J. 2014. Preventive Veterinary Medicine 113, 231–40

# Passive immunity in Irish suckler beef and dairy calves: Associations between test cut-offs for failure of passive transfer classification and calfhood morbidity, mortality and growth

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**Application** Suckler beef and dairy calves with lower passive immunity test results were more likely to experience a negative health outcome or poor growth.

**Introduction** Testing calves for failure of passive transfer (FPT) of immunity is an important step in monitoring the successfulness of on-farm colostrum management programmes and resolving on-going calf health issues. Passive immunity test results are generally categorised for FPT using test-specific cut-off values. Test cut-offs should be validated by examining their relationship with key health and performance parameters. The study objective was to evaluate associations between test cut-off values for FPT classification and morbidity, mortality and growth in Irish suckler beef and dairy calves.

**Material and methods** A total of 1,392 calves from 111 suckler beef farms and 2,090 calves from 84 dairy farms across Ireland were enrolled on the study. Calves were born between July 2014 – June 2016 and followed until 6 months of age. Blood samples were collected by jugular venepuncture and serum was harvested. Serum was analysed for total IgG by BioX K165 (ELISA), total protein by clinical analyser (TP-CA), globulin, zinc sulphate turbidity (ZST), Brix percentage, and total protein by digital refractometer (TP-DR). Farmers recorded information on disease events, health treatments, and calf mortality. Morbidity data were available for 1,192 suckler (n = 84 farms) and 1,733 dairy calves (n = 55 farms). Mortality data were available for all calves. Standardised 205-day body weight (BW) was determined for 450 suckler (n = 9 farms) and 480 dairy calves (n = 8 farms). Descriptive statistics were generated for the passive immunity test results. Subsequently, test results were incrementally categorised so that optimal test cut-offs could be established. Generalised linear mixed models were used to evaluate associations between test cut-off values and morbidity, mortality and growth. Receiver operating characteristic (ROC) curves were constructed and optimal test cut-offs for classification of health and growth outcomes were determined using maximum Youden index.

**Results** Suckler beef calves had lower mean values, compared to dairy calves, across all of the tests for passive immunity (Table 1). Optimal ELISA cut-offs for classification of morbidity and mortality in suckler beef calves were 8 and 9 mg/ml, respectively. Suckler beef calves with ELISA ≤ 8 mg/ml had greater odds of being treated for at least 1 disease event by 3 months of age (OR, 95% confidence interval: 2.0, 1.3 – 2.9, $P < 0.001$) or bovine respiratory disease (BRD) in the first 1 months of life (4.5, 1.4 – 14.5, $P = 0.01$), compared to those with ELISA > 8 mg/ml. The odds of suckler beef calves with ELISA ≤ 9 mg/ml dying by 6 months of age were almost threefold that of those with ELISA > 9 mg/ml (2.8, 1.4 – 5.8, $P < 0.01$). ELISA cut-offs that optimised classification of dairy calves for subsequent health and performance ranged from 10 to 13 mg/ml. Dairy calves with ELISA ≤ 10 mg/ml had greater odds of having 205-day BW in the lower quartile than those with ELISA > 10 mg/ml (2.2, 1.3 – 3.8, $P < 0.01$). Dairy calves with ELISA ≤ 12 mg/ml had greater odds of BRD treatment in the first 6 months of life than those with ELISA > 12 mg/ml (2.4, 1.3 – 4.4, $P < 0.01$). Conversely, dairy calves with ELISA ≤ 13 mg/ml had 40% lower odds of diarrhoea between birth to 6 months of age than dairy calves with ELISA > 13 mg/ml (0.6, 0.4 – 0.9, $P < 0.01$). Other test cut-offs that optimally classified suckler beef calves for health outcomes ranged from 56 to 61 g/l TP-CA, 26 to 40 g/l globulin, 12 to 18 ZST units, 8.4 % Brix, and 5.3 to 6.3 g/dl TP-DR. In addition, other test cut-offs that optimally classified dairy calves for health and growth outcomes ranged from 57 to 66 g/l TP-CA, 29 to 36 g/l globulin, 19 to 23 ZST units, 7.8 to 9.4 % Brix, and 5.7 to 6.8 g/dl TP-DR.

**Table 4** Descriptive statistics for passive immunity test results for suckler beef (n = 1,392) and dairy calves (n = 2,090)

| Test | Suckler beef calves | | | | Dairy calves | | | | *P*-value |
|---|---|---|---|---|---|---|---|---|---|
| | Mean ± SD | Min. | Med. | Max. | Mean ± SD | Min. | Med. | Max. | |
| ELISA, mg/ml | 12.0 ± 5.5 | 1.5 | 11.6 | 47.5 | 14.0 ± 5.9 | 1.5 | 13.9 | 55.5 | < 0.001 |
| TP-CA, g/l | 60.3 ± 8.2 | 36.7 | 59.9 | 87.7 | 62.7 ± 8.3 | 39.8 | 62.6 | 93.0 | < 0.001 |
| Globulin, g/l | 33.1 ± 9.0 | 12.4 | 32.4 | 67.1 | 35.2 ± 9.0 | 13.9 | 34.6 | 68.5 | < 0.001 |
| ZST, units | 15.9 ± 7.0 | 0.3 | 15.6 | 52.0 | 17.5 ± 6.9 | 0.5 | 17.0 | 51.4 | < 0.001 |
| Brix, % | 8.8 ± 0.9 | 6.0 | 8.8 | 13.6 | 9.0 ± 1.0 | 6.0 | 9.0 | 13.2 | < 0.001 |
| TP- DR, g/dl | 5.9 ± 0.9 | 1.5 | 5.9 | 8.7 | 6.2 ± 0.9 | 3.2 | 6.2 | 9.6 | 0.02 |

**Conclusion** These results demonstrate that relationships with health and performance need to be considered when determining cut-offs for FPT classification. Also, different test cut-offs should be applied for suckler and dairy calves because of differences in management conditions and disease risks.

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# The effect of Interleukin 8 haplotype on vitamin D status and the innate immune response to bacterial, viral and fungal ligands in calves

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**Application** Characterisation of IL8 haplotypes has important implications for the neonatal immune response and ultimately in breeding for disease resistance.

**Introduction** At >7% (DAFM/AFBI, 2014) calf mortality in Ireland is significantly higher than our European counterparts (Norway - 3.7%) (Gulliksen *et al*., 2009) and seriously limits the sustainability of the agri-food sector. It is an economic and animal welfare issue which can lead to the overuse of antibiotics in veterinary medicine. Calves rely primarily on their innate immune response during the first six months of life during which time they are susceptible to bacterial and viral pathogens. We have previously discovered two distinct IL8 promoter haplotypes (IL8-h1 and IL8- h2) in Holstein-Friesian calves and IL8-h2 calves produce significantly higher IL-8 levels in response to bacterial LPS *in vivo* (Stojkovic *et al*., 2016). Vitamin D has been shown to alter expression of innate immune cytokines to limit damage caused by excessive inflammation therefore the serum status in the IL8 haplotype calves may be altered. Our hypothesis is that IL8 haplotype will significantly affect the ability of the calf to mount an effective innate immune response and/or vitamin $D_3$ ($25(OH)D_3$) status.

**Material and methods** One month old male Holstein-Friesian calves were genotyped for IL8 haplotype: IL8-h1 (n=5), IL8-h2 (n=6) and heterozygote (n=6), and housed together outdoors. Their developing immune response to bacterial (LPS), viral (poly (I: C)) and fungal (zymosan) ligands were assessed using a whole blood *in vitro* cell stimulation system (TruCulture®, Myriad RBM) at monthly intervals. Blood samples were also taken for serum separation and haematology. Following 24 hours stimulation at 37°C using the TruCulture® system, cells were separated from the supernatant and stored at -20°C. IL-8, IL-1β and circulating $25(OH)D_3$ levels were measured in cell supernatants and serum by ELISA. Haematology analysis was performed using the ADVIA 2120 system. Statistical analysis was performed in GraphPad Prism 7 using one way ANOVA with statistical significance determined using the Holm-Sidak method.

**Results** Haematology profiles from each haplotype show age-associated changes in monocyte, lymphocyte and neutrophil number along with preliminary analysis showing between group differences. ELISA results of the TruCulture® stimulation have revealed significant inter-animal variability within each haplotype group in their response to stimulants. Basal serum IL-8 levels showed a significant difference between IL-8-h1 and IL8-h2 at months 2, 3, 4 and 6 where IL8-h2 had higher levels. Meanwhile $25(OH)D_3$ levels were significantly different at months 5 and 7 with IL8-h1 showing higher circulating levels.

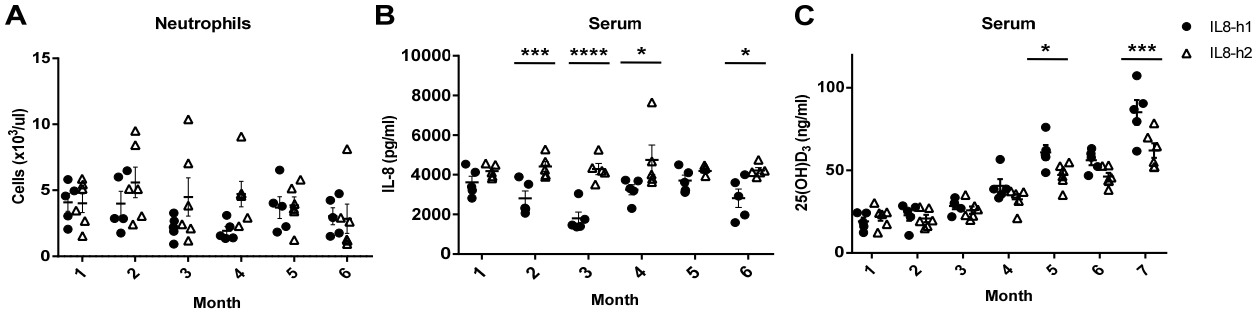


**Figure 1** Graphs show (A) neutrophil numbers (B) IL-8 and (C) $25(OH)D_3$ levels in calves of both IL8 haplotypes

**Conclusion** This is the first use of this novel immunoprofiling assay in cattle which has shown IL-8 haplotype affects the response to certain ligands and regulates vitamin $D_3$ which may have important implications for neonatal immunity and future breeding strategies.

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**References**

DAFM/AFBI. 2014. All-Ireland animal disease surveillance report [online]
Gulliksen, S.M., Lie, K.I., Løken, T. and Østerås, O. 2009. Journal of Dairy Science 92(6), 2782-2795.
Stojkovic, B., McLoughlin, R.M. and Meade, K.G. 2016. Veterinary Immunology and Immunopathology 182, 1-10.

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# Differential expression of plasma proteins at 7 days *postpartum* precede the onset of cytological endometritis in dairy cows

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**Application** This study contributes to our understanding of the pathogenesis of uterine disease and may aid the development of novel, non-invasive prognostic biomarkers to improve disease outcomes.

**Introduction** Following calving, inflammation of the uterus is common during the first week *postpartum*. However, whilst the majority of cows resolve this inflammation a significant proportion fail to do so and go on to develop uterine disease. Our previous work (Foley *et al*., 2015) identified transcriptomic differences in the endometrium of cattle that resolve inflammation and those that develop endometritis as early as 7 days *postpartum* (DPP). Although all cows were in a pro-inflammatory state at 7 DPP, those that developed endometritis were identified by a unique inflammatory signature, including upregulation of pro-inflammatory genes such as *IL1B, NLRP3* and *IL8*. The aim of this project was to investigate whether these changes could also be detected systemically by assessing the proteome of plasma samples.

**Material and methods** Plasma was collected at 7 DPP from 147 cattle. Health status was assessed at 21 DPP using endometrial cytology, where a threshold of >18% polymorphonuclear leukocytes (PMN) was used to identify cattle with endometritis (Kasimanickam *et al*., 2004). PMN analysis performed at D7 confirmed that while the majority of the herd (78%) exhibited high endometrial inflammation (>18% PMN) during the first week *postpartum*, a subset of cattle (22%) had a lower degree of inflammation. Plasma from twenty cows was selected for proteomic analysis, n=10 with high inflammation at D7 (HIGH, n=5 were subsequently healthy and n=5 which developed endometritis at 21 DPP) and n=10 with low inflammation at D7 (LOW, n=5 per group, as above). Proteins were extracted from samples using acetone precipitation. Samples were subjected to tryptic digestion followed by high performance liquid chromatography tandem mass spectrometry. Acquired spectra was processed by MaxQuant followed by protein identification using the andromeda search engine against a forward *Bos Taurus* database concatenated to a reversed decoyed fasta database and common protein contaminant. Proteins were considered differentially abundant if the ANOVA p-value <0.05 and fold change >1.5.

**Results** A total of 183 proteins were identified. Six proteins were differently abundant in HIGH inflammation cows, between those that either resolved or developed endometritis by 21 DPP (Table 1). In contrast, 12 proteins were differentially abundant in LOW inflammation cows (Table 2), where some developed endometritis and others did not.

**Table 1** Differential protein expression at 7 DPP in HIGH inflammation cows (healthy vs. endometritis at 21 DPP)

| Protein | P value |
|---|---|
| Uncharacterised protein | 0.029 |
| Uncharacterised protein | 0.031 |
| Secreted phosphoprotein-24 (Spp-24) | 0.033 |
| Serum amyloid P-component (SAP) | 0.044 |
| Protein HP-20 homolog | 0.049 |
| Glycosylation-dependent cell adhesion molecule 1 (GlyCAM-1) | 0.049 |

**Table 2** Differential protein expression at 7 DPP in LOW inflammation cows (healthy vs. endometritis at 21 DPP)

| Protein | P value |
|---|---|
| Uncharacterised protein | 0.0006 |
| Complement C5a anaphylatoxin (C5) | 0.0008 |
| Fructose-biphosphate aldose | 0.0024 |
| Uncharacterised protein | 0.0051 |
| Kininogen-2 | 0.0054 |
| Inter-alpha-trypsin inhibitor heavy chain H3 | 0.0078 |
| Coagulation factor XI | 0.0204 |
| Complement C8 beta chain | 0.0271 |
| Haptoglobin | 0.0299 |
| Uncharacterised protein | 0.0367 |
| Uncharacterised protein | 0.0430 |
| Peroxiredoxin-2 | 0.0440 |

**Conclusion** Differences in plasma proteins exist at 7 DPP between cattle that go on to develop endometritis and those that remain healthy. These differences vary depending on the level of inflammation at D7, suggesting that development of pathological inflammation that leads to endometritis involves dysregulation of different immune pathways. This work may aid the development of novel, non-invasive prognostic biomarkers to improve disease outcomes.

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**References**

Foley, C., Chapwanya, A., Callanan, J.J., Whiston, R., Miranda-CasoLuengo, R., *et al*., 2015. BMC Genomics 16, 811.
Kasimanickam, R., Duffield, T. F., Foster, R.A., Gartley, C.J., Leslie, K.E., Walton, J.S. and Johnson, W.H. 2004. Theriogenology 62, 9-23.

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# The effect of floor type and diet on the performance and locomotion scores of housed finishing dairy-origin bulls

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**Application** There is evidence from the current study that attaching rubber to concrete slats resulted in improved performance in bulls. However in order to assess the economics of each system, feed costs need to be taken into consideration.

**Introduction** Concern has been raised about the welfare of beef cattle accommodated on fully slatted concrete floors. Covering slatted floors with rubber may provide a suitable alternative to straw bedding in regions where straw availability is low. Bulls are frequently offered an intensive diet, but increased concentrate levels in the diet have been associated with increased lameness. The aim of this study was to assess the effect of attaching rubber to concrete slatted floors and to compare an intensive diet with a less intensive diet on the performance and locomotion scores of dairy-origin bulls.

**Material and methods** Forty eight dairy-origin bulls, with a mean starting live weight of 212 (s.d. 23.7) kg and mean age of 243 (s.d.26.7) days, were blocked according to live weight and breed into 12 blocks, each of four animals. Cattle within each block were randomly assigned to one of four treatments and treatments were balanced as far as possible for breed. Four animals were accommodated per pen and thus there were three pens of animals per treatment. The four treatments consisted of two floors (a fully slatted concrete floor and fully slatted concrete floor with rubber attached over the slats) and two diets; an intensive diet, whereby bulls were offered high concentrate diet or a less intensive diet, whereby the bulls were offered a grass silage based diet, supplemented with moderate levels of concentrates. All bulls were initially offered grass silage supplemented with 2kg concentrate/ head /day. Bulls allocated the intensive diet had concentrate levels increased by 1kg /week until *ad libitum* concentrate feeding was achieved. At this stage this diet was supplemented with chopped barley straw. Bulls allocated the less intensive diet were offered grass silage *ad libitum* throughout the entire experimental period and had their concentrate levels increased by 0.5 kg / week, until it was capped at 6 kg /head/day. All animals were weighed and locomotion scored (using the method of Flower and Weary, 2006) every 14 days. Four pens of bulls, one from each treatment, were slaughtered at the same time, giving three slaughter dates. All bulls were slaughtered once the oldest bull approached 16 months, balanced for treatments. The mean total duration of the experimental period was 216 days. The results were analysed by analysis of variance, in a 2 x 2 factorial arrangement, with pen used as a blocking factor.

**Results** There were no interactions between floor type and diet on animal performance and the main effects are presented in Table 1.

**Table 1** Floor type and diet effects on animals' performance

| | Floor | | | | Diet | | | |
|---|---|---|---|---|---|---|---|---|
| | Concrete slats | Rubber slats | s.e.m | P | Intensive | Less Intensive | s.e.m | P |
| Slaughter weight (kg) | 523 | 544 | 3.8 | 0.004 | 537 | 531 | 3.8 | 0.297 |
| Live weight gain† (kg / day) | 1.49 | 1.55 | 0.018 | 0.003 | 1.49 | 1.50 | 0.018 | 0.957 |
| Dressing proportion (g carcass / kg live weight) | 517 | 520 | 6.3 | 0.731 | 524 | 512 | 6.3 | 0.217 |
| Carcass weight (kg) | 270 | 283 | 3.8 | 0.043 | 282 | 272 | 3.8 | 0.109 |

† Calculated by regression

Bulls accommodated on rubber-covered slats had significantly better performance than those accommodated on concrete slats. Diet had no significant effect on animal performance. There was no significant effect of floor type or diet on locomotion scores.

**Conclusion** Finishing dairy-origin bulls in the present study accommodated on rubber covered slats had higher performance than those accommodated on concrete slats. It is interesting to note that a high level of concentrate inclusion in the diet did not significantly affect animal performance or locomotion scores of these animals.

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**References**

Flower, F.C. and Weary, D.M. 2006. Journal of Dairy Science 89, 139-146.

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# Effect of floor type and space allowance on performance and welfare of finishing beef cattle: A meta-analysis

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**Application** The results of the current meta-analysis provide a better understanding of how finishing beef cattle respond to their housing environment. Placing rubber mats (RM) on concrete slatted floors (CSF) had no effect on performance or welfare, however further research is required examining different RM types. The provision of straw bedding also had no effect on the performance or welfare variables that were evaluated, suggesting that CSF are suitable housing systems for finishing beef cattle. With regards to space allowance, the meta-analysis demonstrated that housing finishing beef cattle below 2.0 $m^2$ per head had a detrimental effect on performance and welfare. Furthermore, no benefit was evident from increasing space allowance per animal above 3.0 $m^2$ on CSF except for improved animal cleanliness.

**Introduction** Data from individual studies evaluating the effect of housing systems on performance and welfare of winter finishing beef cattle are conflicting.

**Material and methods** The objective of this study was to collate the data from previous animal housing studies and quantify, through meta-analysis, the effect of space allowance and floor type on animal performance and welfare variables. From 51 peer-reviewed articles, published between 1969 and 2017, twenty-two were determined to be eligible for meta-analysis. Papers were included in the study if they contained information on the effect of space allowance and/or floor surface on animal performance (average daily live-weight gain (ADG), feed conversion ratio (FCR) and carcass weight), lying behaviour or animal cleanliness. For the purpose of the meta-analysis the space allowances in each study were assigned to one of three classifications (i) less than 2.0 $m^2$ per animal (SA1), (ii) between 2.0 and 3.0 $m^2$ per animal (SA2) and (iii) greater than 3.0 $m^2$ per animal (SA3). The four comparisons investigated in this meta-analysis were: CSF versus (vs). RM, CSF vs. straw bedding, SA1 vs. SA2 and SA2 vs. SA3. All statistical analysis was performed using the Comprehensive Meta-Analysis statistical software (version 3, Biostat, Englewood, NJ). To account for the variation among studies, a weighted meta-regression was conducted. Factors selected for inclusion in the model, as moderator variables, were breed, age, and sex. Furthermore, for comparisons that investigated two different floor types, the difference in space allowance per animal between floor types was included as a moderator variable but was excluded from the final model if it did not affect the overall effect size.

**Results** There was no difference ($P > 0.10$) in ADG, FCR or carcass weight between concrete slatted floors (CSF) and CSF overlaid with rubber mats (RM). Using RM had no effect ($P > 0.10$) on lying duration or dirt scores of cattle. There was no difference ($P > 0.10$) in the ADG, FCR, carcass weight, lying duration or cleanliness of cattle housed on CSF or straw bedding (Table 1). Housing cattle at SA1 had a negative effect ($P < 0.05$) on ADG, FCR, lying time and carcass weight. There was no difference ($P > 0.10$) in the ADG, FCR, carcass weight or lying duration of cattle housed at SA2 or SA3. Cattle were dirtier ($P < 0.01$) when housed at SA2 compared to SA3, however there was insufficient data to make a comparison with SA1.

**Table 1** Differences in performance and welfare variables of cattle housed on CSF and on ST

| | CSF | ST | SE | P-value | n |
|---|---|---|---|---|---|
| ADG (kg) | 1.16 | 1.20 | 0.109 | 0.243 | 7 |
| FCR[1] | 9.08 | 8.60 | 0.797 | 0.168 | 6 |
| Carcass weight (kg) | 347 | 350 | 8.9 | 0.587 | 7 |
| Lying time (hrs/day) | 13.4 | 13.8 | 0.97 | 0.139 | 4 |
| Dirt scores | 42.5 | 34.1 | 4.34 | 0.426 | 5 |

ST = straw; SE = standard error; n = number of studies used in the comparison; [1]Kilograms of dry matter intake divided by kilograms of liveweight gain

**Conclusion** It was concluded that placing RM on CSF had no effect on performance or welfare variables. Likewise, using straw bedding instead of CSF had no effect on animal performance or welfare. Space allowances below 2.0 $m^2$ per animal were insufficient for finishing cattle with regard to performance; whereas increasing space allowance above 3.0 $m^2$ per animal improved animal cleanliness but had no effect on performance.

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# Gestation and early-life environment and its impact on welfare and cognition in pigs

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**Application** Litter-based factors during early life and major life events, such as pregnancy, have significant impacts upon cognition and welfare in pigs, with the potential to affect livestock production.

**Introduction** Assessing cognitive capabilities of animals is key to understanding their welfare needs (Held, *et al*., 2002). This is especially the case during major life events or following a change in conditions, when cognition and welfare needs may change or fluctuate, such as during early-life or pregnancy. A knowledge gap is present in this area, as often research is focused on human and rats or effects of the external environment. Therefore, we aimed to investigate the impact that gestation and early life environment may have in pigs and this is the first study, to our knowledge, to investigate the impact of pregnancy on the mother's cognitive bias in a non-human animal.

**Material and methods** *Study 1:* To assess the possible impact of pregnancy on cognitive bias, ten gilts were trained to differentiate between a positive (P) and negative (N) spatial location situated in opposite corners of a room, reinforced with sugar coated chocolates or coffee beans respectively. Each pig carried out training individually for 15 sessions, with an average of six trials per session, totalling 97 training trials per individual. Once trained each individual carried out cognitive bias testing on four separate occasions; before mating, then five, eight and eleven weeks post-mating. This allowed for assessment of before, early, middle and late pregnancy. The trial sequences involved two of each of the ambiguous spatial probes, situated near the negative (NN), near the positive (NP) or directly in the middle (M) of the positive and negative locations. These were interspersed with positive and negative trials, for example; P, N, *NN*, P, N, *M*, N, P, *NP*. Tests were unreinforced; all ambiguous bowl locations were empty, although P and N bowl locations were reinforced in between the ambiguous testing probes to maintain motivation to respond.
*Study 2:* We investigated the possible effects of sex ratio and litter size on future cognitive bias and injury scores of individual pigs within a litter. For this study, the sex ratio of a litter was determined by the percentage of males within the litter. After weaning pigs were housed in either a barren or enriched environment (deep straw bedding and more space) and 36 pigs (24 males and 12 females) were selected to be cognitive bias tested. As before, all individuals were trained to differentiate between reinforced positive and negative locations before being tested. As well as the cognitive bias tests, 71 pigs were individually scored depending on their injuries, according to a six-point system based on Conte *et al*., (2012) and the tail injury score and mean body score were used.
Ordinal logistic regression was used to statistically analyse tail injury score data with tail injury score denoted as the outcome variable with litter size, sex ratio, treatment, relative weight and sex included as covariates. General linear mixed models were used for all other analyses, with the final model created using stepwise deletion. For the cognitive bias analysis in relation to pregnancy, latency to approach the probe was the outcome variable with pig included as a random effect and probe location and test time were all included in the model. Cognitive bias analysis relating to litter used logged latency to approach the probe as the outcome variable and replicate and pen included as random effects. Personality score, treatment, litter size and percentage males were all included as covariates. Analysis of the body scores employed mean body score as the outcome variable and litter size, percentage males, environment type, relative weight and sex all included as covariates.

**Results** *Study 1:* Pigs before and during early gestation had more optimistic responses to the ambiguous NP, M and NN probes than during middle and late pregnancy ($F_{1,366}$= 5.0161, P= 0.026). The middle and most ambiguous location resulted in the most varied latencies across each test time ($f_{1,366}$=11.7792, P < 0.001).
*Study 2:* When analysing the effect of litter size and sex ratio on cognitive bias we found that individuals originating from a male biased litter were slower to reach each location in comparison to female biased litters ($f_{1,76}$=12.250, P<0.001). Body score analysis showed that female pigs sustained a higher body injury score in comparison to male individuals ($f_{1,598}$= 4.3496, P=0.037). Pigs housed in a more enriched environment were also more likely to have a higher body score if they originated from a litter with a high male: female ratio. This was the opposite for pigs housed in a barren environment ($f_{1,599}$= 5.6721, P=0.017). An interaction between litter size and weight (t=-2.171, P=0.029) was evident for tail injury scores, and was manifested as within the lightest and heaviest pigs, individuals originating from a very large or very small litter had higher tail injury scores

**Conclusion** There is a significant shift in the pigs' mood state between pre/early and mid/late gestational stages, suggesting that they became increasingly pessimistic as they progressed through pregnancy. Early-life factors, including sex ratio and the size of a birth litter impacted on pigs' cognitive bias responses and injury scores later in life.

**Acknowledgements** Sex ratio and litter size data was collected as part of BBSRC grant BB/K00254/1 and BB/K00254/2.

**References**

Conte, S., Lawlor, P.G., O'Connell, N. and Boyle, L.A. 2012. Journal of Animal Science 90, 373 – 380.
Held, S., Mendl, M., Laughlin, K. and Byrne, R. W. 2002. Journal of Animal Science 80, 10-17.

# Performance and welfare of steers housed on concrete slatted floors at fixed and dynamic (allometric based) space allowances

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**Application** The use of allometric equations (e.g. equation $y=0.033w^{0.667}$) to determine space requirements for winter finishing cattle is more appropriate than assigning cattle to a fixed space ($m^2$) per animal.

**Introduction** One way of possibly determining the optimum space for finishing cattle, irrespective of their weight, is through the use of allometric equations. Instead of allocating a fixed space allowance per animal, allometric equations use the progressing weight of an animal to estimate the space that they require during winter housing.

**Material and methods** The study objectives were 1), determine whether allometric equations are suitable for estimating the space requirements of finishing beef cattle housed on concrete slatted floors (CSF) over a 105 day finishing period and 2), examine the effect of fixed and dynamic space allowances on the performance and welfare of these cattle. Continental crossbred steers (n=120: mean initial live weight, 590 (S.D. 29.8) kg) were blocked by breed, weight and age and assigned to one of five space allowance treatments (three fixed and two dynamic) on CSF: i) 2.0 $m^2$ per animal, ii) 2.5 $m^2$ per animal, iii) 3.0 $m^2$ per animal, iv) Equation 1 (E1[1]); $y=0.033w^{0.667}$, where y=$m^2$ per animal and w=body weight, and v) Equation 2 (E2[2]); $y=0.048w^{0.667}$. Each treatment consisted of 6 pens, with each pen containing four steers. Steers were offered grass silage and concentrates (860 g/kg rolled barley, 60g/kg soya bean meal, 50 g/kg molasses and 30 g/kg minerals and vitamins) *ad libitum* and had free access to clean, fresh drinking water. Steers were weighed and dirt scored every 14 days. Blood samples were collected every 28 days, and analysed for complete cell counts. Behaviour was recorded using closed-circuit infrared cameras. Steers' hooves were inspected for lesions at the beginning of the study and post-slaughter. Pen was the experimental unit for all variables. All statistical analyses were performed using SAS software Version 9.3.

**Results** For treatment 4, and 5, the space allowance per animal increased from 2.3 $m^2$ at the start of the experiment to 2.6 $m^2$ at the end of the experiment, and from 3.4 $m^2$ to 3.9 $m^2$, respectively. Slaughter weight and ADG were lowest, and FCR was poorest, for steers accommodated at 2.0 $m^2$, and slaughter weight and ADG were greatest, and FCR was the best, for steers accommodated at E2 ($P < 0.05$); steers accommodated at 2.5 $m^2$ were intermediate ($P > 0.05$) to those accommodated at 2.0 $m^2$, 3.0 $m^2$ and E1, whereas steers accommodated at 3.0 $m^2$ and E1 were intermediate ($P > 0.05$) to 2.5 $m^2$ and E2 (Table 1). Carcass weight of steers housed at 2.0 $m^2$ was lower ($P < 0.05$) than all other treatments. Steers housed at 2.5 $m^2$ had lower carcass weights ($P < 0.05$) than those accommodated at E1 and E2, whereas the carcass weight of steers accommodated at 3.0 $m^2$ was intermediate. Space allowance had no effect ($P > 0.05$) on haematological variables. The number of steers lying at any one time was lower ($P < 0.05$) at 2.0 $m^2$ than any other treatment. There was no difference in lying behaviour of cattle accommodated at 2.5 $m^2$, 3.0 $m^2$, E1 or E2.

**Table 1** Effect of space allowance on intake, performance characteristics and number of hoof lesions of finishing beef steers over a 105 day study period. Values are expressed as least square means ± SEMp

| | Space allowance, $m^2$/steer | | | | | | |
|---|---|---|---|---|---|---|---|
| | 2.0 | 2.5 | 3.0 | E1[1] | E2[2] | SEMp | *P*-value |
| Grass silage dry matter intake (DMI), kg/steer/day | 1.2 | 1.2 | 1.2 | 1.2 | 1.1 | 0.02 | > 0.10 |
| Concentrate DMI, kg/steer/day | 8.8 | 9.2 | 9.6 | 9.4 | 9.8 | 0.21 | 0.07 |
| Total DMI, kg/steer/day | 10.0 | 10.4 | 10.8 | 10.6 | 10.9 | 0.2 | 0.06 |
| Initial weight, kg | 589 | 593 | 590 | 590 | 589 | 2.9 | > 0.10 |
| Slaughter weight, kg | 665[a] | 688[ab] | 705[bc] | 701[bc] | 713[c] | 8.1 | 0.038 |
| Average daily gain (ADG), kg | 0.76[a] | 0.88[ab] | 1.05[bc] | 1.09[bc] | 1.14[c] | 0.066 | 0.041 |
| Feed conversion ratio (FCR)[1] | 13.7[a] | 12.0[ab] | 10.3[bc] | 10.1[bc] | 9.4[c] | 0.69 | 0.016 |
| Carcass weight, kg | 389[a] | 401[b] | 409[bc] | 411[c] | 417[c] | 4.9 | 0.01 |
| Hide weight, kg | 49.1[a] | 50.2[ab] | 52.1[ab] | 50.5[ab] | 54.7[b] | 1.35 | 0.049 |
| Hoof lesions obtained, number per animal | 3.1 | 2.5 | 3.5 | 2.9 | 3.6 | 0.35 | > 0.10 |

[a,b,c]Least squares means within a row without a common superscript letter differ ($P < 0.05$)
[1]kilograms of dry matter intake divided by kilograms of live weight gained

**Conclusion** It was concluded that the allometric equation $y=0.033w^{0.667}$ is sufficient for estimating the space required by finishing beef cattle housed on concrete slatted floors for optimal performance and welfare, and that 2.0 $m^2$ per animal was an insufficient space allowance for housing finishing beef steers over a 105-day period.

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# When 'All-In/All-Out' is not 'All-In/All-Out': implications for pig welfare and performance

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**Application** Pigs delayed from the normal production flow were lighter and at greater risk of welfare issues suggesting that greater effort in adhering to the principles of 'All-In/All-Out' (AIAO) is required on pig farms.

**Introduction** AIAO is a management practice whereby pigs of the same age are moved together through the production stages with the ultimate aim of achieving uniformity in growth and feed efficiency and of minimising disease transmission. However, it is common to hold small and/or sick pigs back from the normal production flow to allow them to catch up; a practice which often involves remixing. The aim of this study was to investigate the possible implications of this practice for pig welfare and performance in a commercial farm with a self-declared AIAO management.

**Material and methods** A total of 1,016 pigs born within 1wk were tagged at birth and followed until slaughter on a commercial pig farm. The declared management on the farm was for pigs to spend 8 wks in the nursery stage after weaning (4 wks in each of the 1st and 2nd nursery stages), 4 wks in the grower stage and 8 wks in the finisher stage. The weekly movement of all tagged animals was tracked. Pigs were individually weighed and inspected for the presence of tail, ear and body lesions at weaning and on transfer between each production stage. Pigs were retrospectively classified into three production flows according to the time they spent in each production stage (flow 1=normal, flow 2=pigs delayed from the normal production flow by 1 wk and flow 3=pigs delayed by >1 wk). Statistical differences were detected for sow parity, birth weight and litter size between flows; thus, a nested case control design was applied whereby pigs from the three flows were matched on these three variables. The matching process yielded a final data set including 120 pigs in flow 1, 60 pigs in flow 2 and 60 pigs in flow 3. As we retrospectively followed each pig based on its own movements through the stages, each pig was considered as the experimental unit. Tail, ear and body lesions were analysed using binomial logistic regression while BW was analysed using generalised linear mixed model equations.

**Results** Growth rate differed between flows on transfer between each production stage (Table 1). Pigs in flow 1 had a higher risk of tail, ear and body lesions than pigs in flow 2 (OR=2.63; OR=4.54; OR=1.69) or 3 (OR=1.16; OR=2.94; OR=2.50). Tail lesions were 2.24 times more likely in flow 3 than in flow 2 pigs and ear lesions were 1.57 times more likely in flow 3 than in flow 2 pigs.

**Table 5** Body weight (LS means ± SE) of pigs retrospectively classified into three production flows according to the time they spent in each production stage and matched by parity, birth weight and litter size in a nested case control study

| | Weaning | | Transfer to 2nd stage | | Transfer to 3rd stage | | Transfer to finisher stage | |
|---|---|---|---|---|---|---|---|---|
| | LS means | SE | LS means | SE | LS means | SE | LS means | SE |
| Flow 1 | 7.2[a] | 0.46 | 12.5[a] | 0.46 | 30.5[a] | 0.46 | 66.8[a] | 0.46 |
| Flow 2 | 5.3[b] | 0.65 | 10.3[b] | 0.65 | 31.9[a] | 0.65 | 61.5[b] | 0.65 |
| Flow 3 | 6.2[a,b] | 0.65 | 10.3[b] | 0.71 | 25.8[b] | 0.92 | 54.4[c] | 0.66 |
| P-value | < .0001 | | < .0001 | | < .0001 | | < .0001 | |

[a,b] Within each column, different superscript letters indicate significant differences for each variable; $P < 0.05$.

**Conclusion** Production flows in which pigs were delayed were associated with poorer performance. Given the lower weaning weights in these flows it is difficult to ascertain whether this was responsible for the persistent poor performance thereafter or if it was causal. The latter is plausible given that delayed pigs were generally re-mixed which is a known stressor with a detrimental impact on growth performance. Pigs in flow 3 had more health problems at slaughter (see Calderón Díaz *et al*., 2017). Combined with our findings on welfare lesions in flow 3 pigs this suggests that being delayed from the normal production system had a negative impact on pig health and welfare. However, welfare lesions were also a problem in flow 1 pigs which were thriving. This suggests that high growth rates, heavier bodyweights and potentially lower space allowances may challenge pigs coping abilities. These results highlight the multifactorial nature of welfare problems for pigs and the negative implications of delaying pigs from an AIAO management system.

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**References**

Calderón Díaz, J.A., Diana, A., Boyle, L.A., Leonard, F.C., McElroy, M., McGettrick, S., Moriarty, J.P. and Manzanilla, E.G. 2017. Porcine Health Management 3, 13.

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# Comparing four different stocking densities on growth rate and feed efficiency of finishing pigs

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**Application** Higher stocking densities had no significant impact on pig performance, feed intake or feed conversion efficiency of finishing pigs.

**Introduction** Recent commercial performance for finishing pigs has been over 1000g/day which is 10-15% higher than previously recorded on our farms. Published research considering stocking density has been conducted where growth rate was reported at 850-950g/day. The aim of this trial was to investigate if stocking density impacted the performance of high genetic merit pigs.

**Material and methods** Upon arrival, pigs were randomly allocated to four stocking densities; T1 – 0.69m²/pig, T2 – 0.73m²/pig, T3 – 0.77m²/pig and T4 - 0.82m²/pig, with entire males and females split equally in T2 and T4 and one extra entire male in T1 and T3. Each treatment had a start weight between 38.4kg and 38.9kg. T1 (0.69m²/pig) was the maximum number of pigs allowed in the pen as per welfare guidelines (Welfare of Farmed Animals Regulations (2012) Northern Ireland), which state that pigs up to 110kgs need 0.65m² of space. There were eight pens per stocking density and all pens had fully slatted floors. Pigs acclimatised for 1 week and were fed JMW Grower, then weighed every 2 weeks until approximately 100kgs. JMW Finisher 1 was fed to approximately 70kg and JMW Finisher 2 was fed from approximately 70kgs until slaughter. Pigs judged by eye to be over 120kgs were marketed to a commercial abattoir and weight and date of slaughter recorded so analysis could be conducted over the trial period. Feed intakes from each pen were recorded. Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) was calculated. Feed remaining in hoppers was measured at the end of the trial. Mortality was recorded. The feed system used (Hotraco Orion) ensured all diets were offered *ad libitum* and the weight of feed recorded to individual pens. The feed regime was identical for each treatment. Statistical analysis was performed using a Remel Linear mixed models design for unbalanced data.

**Results Table 1**

| Start to Final Weigh | T1 | T2 | T3 | T4 | SED | P-Value |
|---|---|---|---|---|---|---|
| ADG (kg) | 1.06 | 1.06 | 1.09 | 1.07 | 0.026 | 0.302 |
| ADFI (kg) | 2.50 | 2.55 | 2.56 | 2.57 | 0.110 | 0.916 |
| FCR | 2.36 | 2.41 | 2.35 | 2.40 | 0.070 | 0.638 |
| Overall trial period | | | | | | |
| Final Weight (kg) | 125.66 | 126.03 | 125.23 | 126.11 | | |
| ADG (kg) | 1.016 | 1.025 | 1.011 | 1.036 | 0.0159 | 0.637 |
| ADFI (kg) | 2.60 | 2.67 | 2.64 | 2.74 | 0.0755 | 0.364 |
| FCR | 2.57 | 2.61 | 2.62 | 2.65 | 0.0539 | 0.473 |

Table 1 shows that there was no relationship between stocking density and pig performance. (P Value >0.05 for ADG, ADFI and FCR) This shows pigs can be stocked at higher densities with no negative impact on performance until first pigs were removed or until all pigs were slaughtered. A higher density meant more weight was gained per pen, and therefore more weight is marketed to the abattoir.

**Conclusion** The work showed that ADG, ADFI and FCR was unaffected by stocking density at the last weighing with the full pen or overall to the end of the experiment (Table 1). This result is contrary to the research of Flohr *et al*., (2016), who found that pigs offered 0.91m² had increased ADG compared to other pens at 0.65m² and Hamilton *et al*., (2003) who found a similar deterioration with decreased floor space. This variation could be caused by several variables – genetics, male castration and health status.

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**References**

Flohr, J.R., Tokach, M.D., DeRouchey, J.M., Woodworth J.C., Goodband, R.D. and Dritz, S.S. 2016. Journal of Animal Science, 94.
Hamilton, D.N., Ellis, M., Wolter, B.F., Schinkel, A.P., and Wilson, E.R., 2003. Journal of Animal Science 81, 1126-1135
Welfare of Farmed Animals Regulations. 2012. Northern Ireland, ISBN 978-0-337-98647-5

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# Attitudes of farmers towards risk factors associated with lameness occurrence in pasture based herds

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**Application** Understanding farmers' attitudes toward risk factors associated with lameness in dairy cows can be useful to design strategies to reduce these factors on farm, considering farm limitations and possibilities to adopt the changes.

**Introduction** Lameness is one of the most important multifactorial diseases in dairy cows, which decreases milk production and the welfare of these animals. The objective of the present study was to identify beliefs and attitudes of pasture based dairy farmers towards risk factors associated with lameness occurrence in pasture-based herds.

**Material and methods** In a previous part of this study, in 2015, 44 pasture based dairy farms in Santa Catarina state, Brazil were visited twice for data collection on lameness prevalence and associated risk factors. A third visit used a benchmarking approach; briefly, farmers were presented their results, in comparison with all participant farms (prevalence varied from 5 to 75% over two seasons); information about the main risk factors for lameness that had been identified at these farms (Bran *et al*., 2017) and relevant preventive measures were discussed individually with each family. In May - June 2017 a qualitative study using in-depth interviews were done with 40 of these farmers. Initially, farmers were asked to give their impressions on the prevalence of lameness in their farms, what they perceived as associated factors and their attitudes regarding the preventive and curative measures. If the associated factors identified in the initial phase of the project (cow breed; low body condition score; older cows; overgrown claws; chasing speed between pasture and milking parlour) had not been mentioned by the farmer, the interviewer proceeded to ask about them. Other questions covered farmers' attitudes and behaviours towards lame cows, to understand how this could impact the human-animal relationship. Interviews transcriptions and thematic analyses were done by the interviewer.

**Results** Although most farmers (n=33) recognised having lame cows on farm, they dismissed lameness as a problem of the free-stall system. Some comments imply a farmer's difficulty to identify lame cows [e.g., "*When it is really lame everyone knows, but when is just a little bit lame, it's not easy.*"]. The conditions of the tracks (stones, gravel, humidity and mud) was the most common cause of lameness occurrence cited by the interviewees (n=17). Participants believed that the hoofs get soft and more sensible with the mud, and that the stony tracks hurt the cow's hoof, but none connected the presence of mud with the transmission of infectious hoof diseases. The majority of the farmers expressed positive attitudes towards improving the tracks, but only five reported making any changes in the past year. The second most cited factor (n=14) was ruminal acidosis caused by feeding excess of protein and low roughage [e.g., "*Silage is a poison for that* (lameness)*, to cause acidosis. And from acidosis comes lameness*."]. This justified the identification of buffer salts as a main preventive measure for lameness. Cow breed was considered a risk factor, but only after the interviewer raised the issue, when farmers commented that Holstein cows were more susceptible to lameness than Jersey cows (the two most predominant breeds on the farms), due to their greater weight and milk production and difficulty to move. However, farmers expressed no intention to change the breed of the herd, showing a preference for the higher productive Holstein cows. None of the participants considered cow's age, body condition score, or overgrown claws a risk factor for lameness in their herds. This justified beliefs, expressed by the majority of the interviewees, that hoof trimming is not necessary for their cows (only two reported using it as a preventive measure). Hoof trimming was considered important only for free-stall farms [e.g., "*The neighbour has a free-stall, he does* (hoof trimming)*. They do it because they need to. But on the pasture system, nobody"*]. Although 18 reported having culled a cow due to lameness, this was not a main reason for culling cows. Chasing speed was not associated with the risk of increasing lameness. When asked about it, 24 farmers said they did not need to chase their cows; however, 15 recognised the need to chase the cows. Moreover, 37 talked about "lazy" or "sluggish" cows to describe lame, older cows that needed to be pushed, with many expressing negative attitudes and some describing aversive behaviours during the daily management of the herd.

**Conclusion** Results provide evidence of a prevailing culture where lameness is not a recognized problem in pasture based farms, which acts as a barrier for individual farmers to recognize this malady and associated risk factors that might trigger preventive behaviour on farm. Notion of this prevailing culture is relevant since the success of any health control programme at an individual level (i.e. control of lameness on farm) is dependent on a good understanding of the community cultural context that forms and fosters behaviours and decisions.

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**References**

Bran, J.A., Daros, R.R., von Keyserlingk, M.A.G., LeBlanc, S.J. and Hötzel, M.J. 2017. Preventive Veterinary Medicine. In press.

# A comparison of the growth performance of Holstein heifer calves fed two levels of milk replacer in combination with two levels of crude protein in the calf starter feed

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**Application** Optimising pre weaning growth rates in dairy heifers may aid in meeting weight targets at puberty and first calving and lead to increased milk production in the first and subsequent lactations.

**Introduction** Soya is a high quality protein source widely used in animal feed which combines high protein concentration with a favourable amino acid profile. However, the soya bean contains a number of anti-nutritional factors which when soya is not correctly processed can significantly decrease the digestibility and utilisation of the diet (Dei, 2011). The inclusion of Hamlet Protein ™ (HP) (a soya based feed protein ingredient where a bio-conversion process reduces anti-nutritional factors and antigens in soya to a safe level without compromising protein quality) in calf starter feed (CSF) removes the risk of anti-nutritional factors and may optimise growth rate. However, the ideal crude protein (CP) concentration of CSF including HP and the level of milk replacer (MR) that best supports HP inclusion in calf starter feed is unknown. The objective of the present study was to compare the growth performance of Holstein calves fed milk replacer at 6 litres (MR6) or 8 litres (MR8) per day together with a CP concentration in CSF of 180g/kg DM (CP18) or 220g/kg DM (CP22).

**Material and methods** From birth Holstein heifer calves (N=64) were individually housed in standard calf pens with straw as bedding. Diets (MR6 CP18), (MR8 CP18), (MR6 CP22) or (MR8 CP22) were introduced (based on date of birth and a randomised group treatment allocation) at 3 days of age together with grass hay and clean water. Calves remained in individual penning for 10 days until they were fully adapted to the bucket and teat feeding system and consuming all their daily MR allocation. Calves were then moved to larger pens where four calves born consecutively within 7 a day period and receiving the same diet were housed together, there were four pens of four calves per treatment. While housed in groups and up to weaning (day 42) MR continued to be offered individually to avoid over/under consumption by any individual calf with CSF offered on a pen basis. All calves were weaned by reducing the quantity of MR offered over a period of 6 days between 37 and 42 days of age. The consumption of MR and CSF was recorded up to weaning following which calves were housed together and fed a commercial feed pellet together with grass and maize silage. The calves were weighed at birth, when entering group housing (day 10), at weaning (day 42) at day 56 and at the end of the study (day 84). Wither heights, girth and body condition scores (BCS) were determined at birth and at 42, 56 and 84 days of age. Health and faecal scores were monitored throughout. Data were analysed by repeated measures analysis of variance using birth weight as the covariant.

**Results** Calves consuming diet MP8 CP22 achieved the CP requirement for growth (600g/d; NRC 2001) by week 2 whereas the other diets did not meet this requirement until week 4. By weaning (day 42) both MR and CP level affected calf weight with calves receiving the combination of MR8 and CP22 tending to have the highest liveweight at weaning (Table 1). There was a tendency for increased girth ($P < 0.055$) at weaning for diet CP22. No statistical differences between diets in incidence of ill-health, faecal score, physical measurements or BCS were found.

**Table 1** Liveweight (kg) recorded at 10, 42, 56 and 84 days (D) of age.

| | Treatment | | | | SEM | P<[1] | | |
|---|---|---|---|---|---|---|---|---|
| | CP18 | | CP22 | | | | | |
| | MR6 | MR8 | MR6 | MR8 | | MR | CP | MR x CP |
| D10 (kg) | 46.5 | 48.5 | 47.3 | 48.8 | 0.47 | 0.001 | 0.227 | 0.602 |
| D42 (kg) | 66.3 | 67.0 | 66.6 | 71.4 | 1.15 | 0.017 | 0.037 | 0.070 |
| D56 (kg) | 76.9 | 75.9 | 76.1 | 82.9 | 2.50 | 0.114 | 0.091 | 0.036 |
| D84 (kg) | 101.9 | 99.7 | 99.2 | 105.8 | 2.58 | 0.386 | 0.501 | 0.082 |

[1] Probability of no effect of MR, CP, or MR x CP interaction; SEM, Standard Error of the Mean

**Conclusion** A higher liveweight gain at weaning (day 42) was achieved in calves fed MP8 CP22, with no negative effect on calf health observed for this higher protein diet. This may be advantageous as pre-weaning weight gain accounts for 22 percent of the variation in first lactation milk yield (Soberon. *et al*., 2012).

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**References**

Dei, H.K. 2011. IBSN: 978-953-307-533-4, In Tech.
Soberon, F., Raffrenato, E., Everett, R.W., Van Amburgh, M.E. 2012. Journal of Dairy Science 95, 783-974.
National Research Council (NRC). 2001. Nutrient Requirements of Dairy Cattle. Seventh Revised Edition.

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# Sustainable dairy production systems: Factors affecting Holstein dairy heifer survival and lifetime productivity

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**Application** This study demonstrates that achieving target age at first calving (AFC) and live weight (LWFC) at first calving increases lifetime productivity and longevity of dairy heifers, thus reducing the number and costs of herd replacements.

**Introduction** Dairy heifers contribute significantly to farm profitability and increasing the efficiency of heifer rearing can lower the cost of milk production and improve dairy margins. The aim of this study was to assess the effect of age, live weight and estimated mature weight at first calving on subsequent lifetime milk production, fertility and longevity of dairy cattle.

**Material and methods** Lifetime records of 1,017 Holstein heifers, born between 2006 and 2017 on a UK robotic milking dairy farm, were obtained from the farm databases (Uniform-Agri Professional UK and Lely T4C), veterinary assessment notes, farm records and interviews with farm staff. Data was collated detailing mortality of calves and growing heifers according to date of birth, reason for mortality, season of death, and age at death categorised according to age (and stage of development): 0 to 48 h (perinatal), 3 to 30 d (neonatal), 1 to 6 mo., 6 to 12 mo. and 12 mo. to calving. Morbidity and treatments administered for clinical symptoms of ill health in calves and growing heifers between 03/10/14 to 08/10/16 were categorised according to animal age, disease, season of treatment and number of treatments per animal. Age at first calving (AFC) and live weight at first calving (LWFC) were recorded. Live weight at calving was calculated for all dairy cattle as the mean live weight over the first 7 d of lactation, which was determined automatically using load cells in the Lely milking robots. Proportion of mature weight (MW) was calculated as LWFC divided by mean live weight of all cows entering the ≥third lactation for each year that heifers calved and entered the study. Individual daily milk yield (MY) and calving date were used to calculate days in milk (DIM), 305 d and mean daily milk yield during first lactation. Long-term performance of animals that had exited the herd at conclusion of the study was assessed as: calving interval between first and second lactation; total lifetime days in milk (calculated as the difference between calving and drying off dates for each lactation); and lifespan in days (calculated as the difference between date of birth and date the animal left the herd). A longevity index was calculated as lifetime DIM/lifespan and was used to represent the proportion of an animal's lifetime that was spent producing milk. Data were analysed using Kaplan Meier survival analysis (Genstat 17$^{th}$ Edition), multiple regression and GLM ANOVA (Minitab 17.2.1). The population was categorised according to: AFC (d): Early; <700 d, Target; 701 to 759 d, Late; 760 to 879 d and Very late; >880 d, and LWFC (kg): <500 kg, 501 to 550 kg, 551 to 600 kg, 601 to 650 kg and >651 kg, which were applied as fixed effects, while animal was applied as a random effect in the models.

**Results** Percent of heifers born alive that did not enter the milking herd, due to mortality or culling, was 18.8%. Estimated probability of perinatal and neonatal mortality was greater for calves born in winter. AFC was 49 d greater, while LWFC was 41 kg less for heifers that received five or more treatments for pneumonia and/or enteric disease compared with those that received no treatment. Mean AFC was 759 (± 4.3) d and LWFC was 571 (± 5.3) kg. Only 12.1% of animals achieved both the national target AFC (22 to 24 mo.) and/or LWFC (550 to 650 kg) and only 4.9% achieved both the target AFC and MW (85% pp MW). Lower MY was produced by animals that calved with AFC <700 d and LWFC <550 kg, however, above these values there was no effect of AFC and LWFC on milk yield. Analysis of AFC and LWFC in combination showed that LWFC was more closely associated with first lactation MY than AFC. Heifers that calved for the first time at <700 d of age and <500 kg had greater lifetime DIM and spent a greater proportion of their life in production, spending 54% of their life in a productive state, compared with 33% achieved by heifers that calved at LWFC >651 kg. Optimum lifetime productivity in terms of milk yield, longevity and proportion of life spent productive was achieved at an AFC between 701 and 759 d and LWFC between 551 to 600 kg, which is equivalent to LWFC between 79 and 81% of mature weight.

**Conclusion** In this herd only one in eight heifers calving by the national target AFC of 22 to 24 mo., one in twenty five calved at the LWFC of 550 to 600 kg for this breed and only one in twenty achieved both targets. Failure to achieve industry targets was potentially due to lack of measuring live weight of growing heifers, along with effects of pneumonia and/or enteric disease. These findings demonstrate the importance of farm specific data in achieving a balance between MY and life span, replacement rate and lifetime productivity.

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# Preference of Holstein calves for potential enrichment items in combination or on a rotating schedule

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**Application** Giving dairy calves several items with which they can interact occupies more of their time than a single, changing item, and may improve their welfare. Calves preferred scented hay from hay nets to brushes, teats and ropes.

**Introduction** Environmental enrichment, meaning the addition of stimuli or increased complexity to the environment for the benefit of the animals, is a common strategy to improve welfare and reduce undesirable behaviour in captive animals, and sometimes has benefits for productivity as well (Leone and Estevez 2008; Meagher *et al.*, 2014). The environment in early life is likely of particular importance for both welfare and productivity in the long-term (e.g. Hultgren and Svensson 2009; Costa *et al*., 2016). Very little research has been done to identify effective physical environment enrichment for young cattle to date. We therefore investigated dairy calves' relative usage of various items added to the pen as a measure of preference, and compared this between two different enrichment schedules (static or fixed, multi-item vs. rotating enrichments). The goal was to identify items and enrichment protocols that have potential to improve welfare and promote normal behaviour.

**Material and methods** Twenty-seven Holstein heifer calves were housed from 3 ± 3 days of age in individual pens and fed milk replacer twice per day (4 L/feeding for 28 d then 3 L/feeding). On day 21 of the study, they were assigned alternately to one of three treatments: control (C), fixed enrichment (FE), which received all four items for the full treatment period, and rotating enrichment (RE), which received one item each week. The items provided were a blind rubber teat, a brush, a hanging rope or spring, and a net filled with scented hay. Observations were conducted using scan sampling three days per week for 4 weeks, from 12:00-14:30. Interactions with the enrichment as well as general time budgets were assessed including play and time sucking on neighbouring calves or parts of the pen. Welch's t-tests for unequal variances were used for planned comparisons of usage between enrichment items and overall usage between enrichment protocols. A chi square test was used to compare enrichment use as a binary variable between weeks within treatments FE and RE.

**Results** Calves spent the most time interacting with the hay net compared to the non-food items (mean 2.0±0.5 vs. 0.2±0.05 % of all scans including both FE and RE calves; $F_{1,17}$=11.5, $P$=0.003). The calves in FE spent more time interacting with enrichments than those in RE did (mean 4.0±0.9 vs. 1.7±0.04 % of scans; $F_{1,10}$=5.66, $P$=0.038).
Overall, use of enrichments did not change significantly over time ($P$>0.05). Numerically, the highest proportion of calves using enrichment was in the first week, with 78% of calves in enriched pens being seen interacting with enrichment at least once.

**Conclusion** A net containing forage with added scent occupied more of calves' time than the other items; teats attracted surprisingly little attention in this group of calves. More research may be needed to identify appropriate non-food stimuli to improve welfare in milk-fed calves. Providing several stable enrichment items is likely to be preferred and to better promote normal behaviour in dairy calves than a frequently rotating schedule, and interest in these items was maintained for more than three weeks. Future analysis will investigate the effects of these enrichments on behaviour, including undesirable sucking, and health.

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**References**

Costa, J.H.C., von Keyserlingk, M.A.G. and Weary, D.M. 2016. Journal of Dairy Science 99, 2453-2467.
Hultgren, J. and Svensson, C. 2009. Preventative Veterinary Medicine 89, 255-264.
Leone, E.H. and Estevez, I. 2008. Poultry Science 87, 14–21.
Meagher, R.K., Ahloy Dallaire, J., Campbell, D.L.M., Ross, M., Møller, S., Hansen, S.W., Díez-León, M., Palme, R. and Mason, G. 2014. PLoS One. 9, e110589.

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# Impact of weaning method on feeding behaviour of group housed dairy calves

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**Application** Weaning method had a significant impact on calves' use of automatic concentrate and milk feeders. Gradual weaning encouraged increased concentrate consumption at an earlier age, however, at 8 weeks of age calves were consuming greater than 2 kgDM concentrate per day regardless of weaning treatment.

**Introduction** Weaning is regarded as one of the most significant stressors in early life and has the potential to impact on calf behaviour, performance and welfare (Weary *et al*., 2008). Data derived from automatic feeders has the potential to highlight feeding behaviours which could indicate a behavioural response to stress as a result of feeding regime (De Paula Viera *et al*., 2008). The aim of this study was to examine the impact of abrupt or gradual weaning of group housed dairy calves on usage of automatic milk and concentrate feeders.

**Material and methods** Ninety Holstein calves balanced for sex and birth weight were allocated at 5 days of age to either GRADUAL (n=45) or ABRUPT (n=45) weaning treatments. Calves were group housed in one of 6 replicate straw bedded pens serviced by automatic milk and concentrate feeders and had free access to fresh drinking water upon entrance to the group pen. ABRUPT calves were offered 6L/day milk replacer (MR) between days 5-49 with complete withdrawal of MR at d50. GRADUAL calves were offered 6L/day MR between days 5-35, following this MR volume was reduced gradually between days 36-56 from 6L/day to 2L/day with complete withdrawal of MR at d57. Milk replacer was offered at a rate of 150g/L with both treatments offered the same total quantity of milk powder (39.3 kg). Milk feeding behaviour, including frequency and duration of visits and drinking speed, was recorded via automatic feeder. Duration of visits to the concentrate feeder were also recorded automatically. Weights were recorded on a daily basis via half bodyweight scales linked to the automatic milk feeder. Feeding behaviour data were split into 4 periods; d5-35 (Period 1), d36-49 (Period 2), d50-56 (Period 3) and d57-62 (Period 4). Data was analysed using GenStat® (version 16.2, VSN International Ltd). Data, excluding number of visits, were fitted to a repeated measures REML model with fixed effects of Sex, Weaning Plan and Age and associated interactions with Pen included as a random factor. For Periods 2-4, a covariate obtained by averaging data from the 5 days immediately prior to the beginning of the next period, was included in the model. Number of visits to the feeder were fitted to a GLMM using the same criteria as described above.

**Results** GRADUAL calves displayed an increased number ($P<0.001$) of shorter visits ($P<0.001$) to the milk feeder than ABRUPT calves during Period 2 (d36-49). This resulted in GRADUAL calves occupying the milk feeder for over 2 minutes per day per calf longer than ABRUPT calves ($P<0.001$). Weaning of ABRUPT calves at d50 resulted in an initial steep increase in number of unrewarded visits to the milk feeder ($P<0.001$) compared with GRADUAL calves, however this had abated by d52, with number of unrewarded milk feeder visits comparable between weaning treatments. GRADUAL weaned calves displayed an increased number of unrewarded visits to the milk feeder following complete withdrawal of milk during Period 4 (d57-62) when compared to ABRUPT calves ($P<0.001$). GRADUAL calves consumed almost twice as much concentrate per day ($P<0.001$) during Period 2 (d36-49) when compared with ABRUPT calves, spending ~ 8 minutes per calf per day longer in the concentrate feeder than ABRUPT calves ($P<0.001$). There was no significant difference in concentrate intake during Period 4 (d57-62) with calves from both treatments consuming greater than 2kg concentrate DM per day, however, GRADUAL calves spent approximately 11 minutes per day longer in the concentrate feeder when compared with ABRUPT calves ($P<0.001$).

**Conclusion** Gradual weaning of calves encouraged earlier intake of starter concentrate when compared with abruptly weaned calves, however this advantage did not exist past 8 weeks of age. Abrupt weaning of calves at 50 days of age resulted in a rapid increase of unrewarded visits to the milk feeder, however, throughout the experimental period, gradually weaned calves displayed an increased number of unrewarded visits to the milk feeder, this potentially indicating an underlying behavioural response to hunger as a result of milk feeding regime.

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**References**

De Paula Vieira, A., Guesdon, V., de Passillé, A.M., von Keyserlingk, M.A.G. and Weary, D.M. 2008. Applied Animal Behaviour Science 109(2-4), 180-189
Weary, D. M., Jasper, J. and Hötzel, M.J. 2008. Applied Animal Behaviour Science 110(1-2), 24-41

# An investigation of pasture allowance and its effect on the performance of growing dairy heifers

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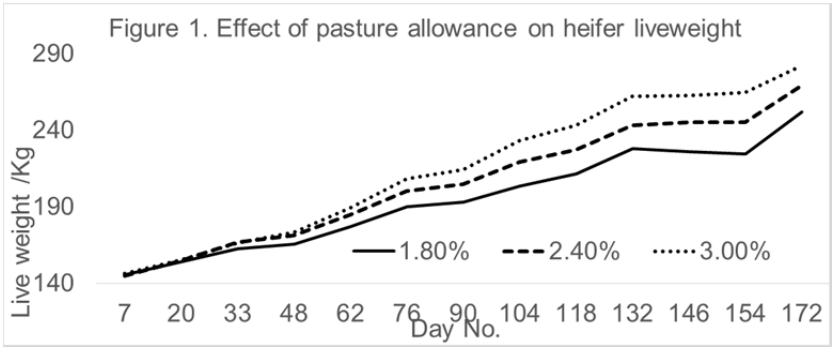
**Application** Pasture allowance (PA) of 3.0% live weight (LW) per Kg dry matter (DM) of grazed grass per animal per day produced a significant improvement in the growth of Holstein heifers compared to reduced PA. Pasture utilisation (PU) was decreased as a result of higher PA, whilst pasture growth was not significantly affected.

**Introduction** Grazed grass provides a high protein/low fat diet, and remains the cheapest source of high quality feed in ruminant livestock systems. In pasture based animal production systems, profitability is directly related to the efficiency of grass production and utilisation (Dillon *et al*., 2008). Industry guidelines for PA vary from 2.0% to 3.0% for growing cattle. The aim of this study was to investigate the effect of pasture allowance on heifer performance, in addition to pasture production and utilisation.

**Material and methods** Holstein heifers (n=72) were assigned to three PA treatments at 1.8%, 2.4% and 3.0% LW allowance of grazed grass per day. Paddock area was allocated based on Kg DM available on a rotational basis. Treatment groups were setup in triplicate (n=8) comprising heifers aged average (Av) 5 months (s.e. 3.88) with Av weight 155 kg (s.e. 3.63). The study commenced on 6 April 2017 with a 14 day rotation length (2 day paddock residency) until 1 June 2017, with a rotation length of 24.5 days thereafter (3.5 day paddock residency). Three separate areas were available for grazing, each comprising 3.68 ha. These were split into seven blocks (A to G) and further subdivided into paddocks specific to treatment with 1215 $m^2$, 1742 $m^2$ and 2310 $m^2$ available for 1.8%, 2.4% and 3.0% PA, respectively. A proportion of all paddock areas were used for silage production initially and extra grazers were incorporated to manage the surplus pasture available in the paddocks. Herbage production and quality was only focused on in the core grazing areas. Av pre-grazing herbage cover across treatments was 3330Kg DM/ha. Extra grazers' numbers were managed daily in order to achieve similar post grazing heights. Heifer live weight and body condition score (BCS) were recorded on a fortnightly basis using a manually operated weighbridge (Tru Test Ltd, UK). Compressed sward heights were measured with a rising plate meter (Jenquip, New Zealand) and recorded on animal entry and exit to each paddock. Herbage mass was determined by taking grass clippings (Bosch, UK) of 0.1 m x 1 m across 5 random plots within each paddock (total 1.0 $m^2$) cut above 4 cm.. Samples collected were weighed fresh and submitted for laboratory analysis to determine the oven dry matter (dried at 60$^o$C for 72 h); water soluble carbohydrate (WSC), crude protein (CP), acid detergent fibre (ADF) and metabolisable energy (ME) via near infrared spectrometry (0.2 m2 above 4 cm). Additional samples were retained frozen for assessment of pasture composition. Data were analysed using repeated measures and by analysis of variance (ANOVA).

**Results** Heifers allocated a greater PA exhibited a significant ($p$<0.001) increase in LW performance as shown in Fig 1, with an average daily LW gain of 0.82 kg/day in 3.0% treatment group compared to 0.64 kg/day in 1.8% group. Average BCS was higher for heifers in the 3.0% group by the end of the treatment period than the PA 1.8% heifers. Animals within the 3.0% group demonstrated an increase in average daily intake of 1.2 Kg DM more than 1.8% PA. A significant difference ($p$<0.001) in pasture utilisation (PU) was evident across the three treatments. The 1.8% PA demonstrated higher PU of 81.5%, than 3.0% PA at 67.3%. PA treatment had no significant effect on pasture production or pasture quality.




**Conclusion** High profitable lifetime index (PLI) Holstein Dairy heifers' can perform to target LW gain on a solely pasture based grazed diet, across a 160 day grazing season. Whilst higher PA improves LW performance, PU and stocking rate are compromised. 2.4% PA provided satisfactory LW gains and PU at 0.75kg/day and 72% respectively. All treatments had high pasture production and quality, justifying the time spent managing heifer grazing.

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**References**

Dillon, P., Hennessy, T., Shalloo, L., Thorne, F. and Horan, B. 2008. Dairy Technology 61, 16-29.
Virkajarvi, P., Nousiainen, J., Khalili, H. 2002. Animal Feed Science and Technology 97, 23-40.

# The effect of calf jackets on the health and performance of dairy origin beef calves

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**Application** The use of calf jackets at an average ambient temperature of 16°C did not have any significant effect on the health and performance of dairy-origin beef calves.

**Introduction** At birth a calf's thermoregulatory system is metabolically immature, due to its inability to generate heat through rumen fermentation (Collier *et al*., 1982). This is of particular concern during variations and extremities in climatic conditions (Bateman *et al*., 2016). New born calves have a lower critical temperature range of 9-15°C (Scanes, 2011). However, this may be influenced by wind speed, humidity, age, bodyweight, nutrition and the quality of bedding (Camiloti *et al*., 2012). Thermoregulatory responses to cold stress come at a cost; calves housed at -4°C compared to 10°C have on average a 32% higher maintenance requirement (Scibilia *et al*., 1987). Furthermore, in many dairy origin beef systems, calves are bought at a young age, and often re-grouped and co-mingled with calves from different farms, each with a differing previous pathogenic exposure, thus creating an increasingly stressful environment (Hulbert & Moisa, 2016). Calf jackets present the opportunity to provide calves with a barrier to environmental conditions without restricting air flow through the house.

**Material and methods** This study was an on-farm trial which began in May 2017 and will continue until April 2018 to take into account the effects of calf jackets at differing ambient temperatures over a one year period. Two batches of dairy bred beef calves have been through this trial from May to August 2017 (n=88, n=75 respectively). Calves were sourced from 10 and 14 dairy farms respectively and moved to a single commercial calf rearing facility. Calves were weighed and vaccinated for viral pneumonia on day 1 (day after arrival) and assigned to one of four treatment groups: (i) no calf jacket (Control), (ii) calf jacket for 2 weeks post arrival (Arrival) (iii) calf jacket for a minimum of 2 weeks and until 65kg live weight (Weight) (iv) calf jacket until 5 days post weaning (Weaning). Treatment groups were balanced for breed, sex, weight and source farm and all calves were under 75kg on arrival. Calves were housed in a naturally ventilated, straw bedded shed and fed milk via an automatic feeder, with ad lib access to concentrates ( 16g/kg DM CP). Milk replacer CP was 20g/kg DM at an inclusion rate of 128g/L. Calves were weighed automatically daily via a partial weigh platform linked to the milk feeder. Disease incidence and antibiotic treatments were recorded daily. Skin temperature of 20 jacket calves and 20 non jacket calves was monitored using a DS1922L iButton (Maximum Integrated) during week 2 and 3 of each batch. Ambient temperature and humidity within the shed were monitored continually throughout the trial period using 3 DS1923 iButtons. Each calf was scored for rub marks around the withers, shoulder and legs on removal of the calf jacket. D50 weight was analysed by ANCOVA using start weight as a covariate, while the remainder of the data was analysed by ANOVA using Gentsat.

**Results** Mean initial live weight was 53.5±1.5kg. There was no significant difference in D50 weight, DLWG or disease incidence across the four treatment groups as shown in Table 1. Ambient temperature remained high throughout batch one and two with a range of 2.6-28.6 °C and 8.3-27.8°C respectively. Calves wearing a jacket had on average a 3.91 °C higher skin surface temperature ($p$<0.001) during week 2 and 3. No rub marks occurred as a result of the calf jacket in either batch.

**Table1** Mean performance and disease incidence of calves in batch one and two

| | Control | Arrival | Weight | Weaning | SEM | Sig. |
|---|---|---|---|---|---|---|
| D50 weight (kg) | 99.3 | 101.8 | 93.5 | 97.6 | 2.4 | ns |
| DLWG (kg/d) | 0.93 | 0.95 | 0.80 | 0.88 | 0.49 | ns |
| Jacket days | $0^{a}$ | $14^{b}$ | $21^{c}$ | $52^{d}$ | 0.75 | <0.001 |
| Disease incidence (%) | 36.6 | 31.0 | 48.8 | 38.5 | 0.41 | ns |

**Conclusion** The use of calf jackets during the summer months at an average ambient temperature of 16 °C did not present any significant benefit in terms of calf health or performance.

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**References**

Bateman, H. G., Hill, T.M., Aldrich, J., Schlotterbeck, R. and Firkins, J. 2012. Journal of Dairy Science 95, 363-369.
Camiloti, T.V., Fregonesi, J.A., Keyserlingk, M.A.V. and Weary, D. 2012. Journal of Dairy Science 95, 3380-3383
Collier, R.J., Beede, D.K., Thatcher, W.W., Israel, L.A. and Wilcox, C.J. 1982. Journal of Dairy Science 65, 2213-2227.
Hulbert, L. E. and S. J. Moisa (2016). Journal of Dairy Science 99(4), 3199-3216.
Scanes, C. 2011. Fundamentals of Animal Science, Delmar, USA.
Scibilia, L.S., Muller, L.D., Kensinger, R.S., Sweeney, T.F. and Shellenberger, P.R. 1987. Journal of Dairy Science 70, 1426 1433.

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# The effects of milk quantity, forage type and age at forage introduction on pre weaned dairy calf performance

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**Application** Concentrate dry matter intake (DMI) of conventionally milk fed calves was greater than accelerated milk fed calves. Accelerated milk fed calves consumed greater dietary DMI than conventionally milk fed calves. No interaction was found between concentrate DMI and forage treatment, however there was a significant MR x forage interaction.

**Introduction** Feeding accelerated (accel.) milk replacer (MR) quantities to dairy calves has been associated with reduced concentrate (conc.) DMI, compared with conventional (conv.) MR feeding. Providing forage to pre weaned calves has been reported to encourage conc. DMI, however, little information exists concerning the effects of forage type combined with animal age at forage introduction on conc. DMI. This study investigated the outcomes of offering calves on accel. and conv. milk feeding regimen, various forage types and ages at dietary introduction, on conc. DMI and total dietary DMI. This study hypothesised that offering calves on Accel. MR regimen a forage source earlier in life would encourage conc. DMI.

**Material and methods** 75 dairy origin calves were introduced to dietary treatments (Table 1) at 5 d of age. Treatment groups were balanced for sex and birth weight. Water, and conc. provision was available on an *ad libitum* basis to calves throughout the study. Daily recording of water, forage (if applicable), and conc. intakes was conducted from 5 d to 70 d of age. All animals weaned on d 70. Data were analysed using a REML variance components analysis with Genstat v16 software.

**Table 1** Milk feeding level and forage treatment

| Forage treatment | CS14 | CS56 | GS56 | NF* | | |
|---|---|---|---|---|---|---|
| Milk feeding regimen | Accelerated | | | | Conventional | |
| MR feeding (days) | 5-42 | 43-56 | 57-67 | 68 -70 | 5-67 | 68-70 |
| MR powder (g/day) | 1350 | 900 | 450 | 300 | 600 | 300 |
| MR quantity (L/day)** | 9 | 6 | 3 | 2 | 4 | 2 |
| Frequency (meals/day) | 3 | 2 | 1 | | 2 | 1 |

*CS = chopped straw, GS = grass silage, NF = no forage provison, number = age at introduction **MR powder 150g/L

**Results** (Table 2) Daily conc. DMI was higher (P=0.021) for conv. milk fed animals (0.69kg/d) compared to animals receiving accel. milk feeding (0.64kg/d). Forage did not affect conc. DMI, however there was a significant interaction (P=0.019) from MR x forage on conc. DMI. Calves from Accel.NF and Accel.CS14 treatments consumed less conc. dry matter (DM) than animals in Conv.NF and Conv.CS14 treatments. Animals from treatment group Conv.CS14, consumed more conc. DM than animals in Accel.GS56, Accel.CS56, Conv.GS56 and Conv.CS56. Calves fed accel. MR quantities consumed greater total DMI/d (P=<0.001) than conv. milk fed calves at 1.61kg/d and 1.28 kg/d respectively. Calves provided with grass silage from 56 d, and calves with no forage provision consumed less (P=<0.001) water than calves provided with chopped straw from 56 d of age. Calves from Conv.GS56 consumed less water (P=0.047) than calves from Accel.GS56, Accel.CS56, Conv.CS14, and Conv.NF treatments at 1.25 L/day vs.1.49, 1.71, 1.56, and 1.30L/day respectively.

**Table 2** Predicted means for dietary intake of dry matter and water

| | | Conc. DMI (kg/d) | SED | Sig. | Total DMI (kg/d) | SED | Sig. | Water intake (L/d) | SED | Sig. |
|---|---|---|---|---|---|---|---|---|---|---|
| MR level | Accelerated | 0.64[a] | 0.021 | 0.021 | 1.61[b] | 0.023 | <0.001 | 1.49 | 0.059 | NS |
| | Conventional | 0.69[b] | | | 1.28[a] | | | 1.46 | | |
| Forage | No Forage | 0.66 | 0.03 | NS | 1.42 | 0.032 | NS | 1.33[a] | 0.083 | <0.001 |
| | Grass silage 56 days | 0.67 | | | 1.45 | | | 1.37[a] | | |
| | Chopped straw 14 d | 0.69 | | | 1.48 | | | 1.48[ab] | | |
| | Chopped Straw 56 d | 0.64 | | | 1.42 | | | 1.63[b] | | |
| MR x Forage | Accel.NF | 0.62[a] | 0.043 | 0.019 | 1.57 | 0.047 | NS | 1.36[abc] | 0.117 | 0.047 |
| | Accel.GS56 | 0.67[ab] | | | 1.65 | | | 1.49[bcd] | | |
| | Accel.CS14 | 0.62[a] | | | 1.61 | | | 1.40[abc] | | |
| | Accel.CS56 | 0.65[ab] | | | 1.61 | | | 1.55[cd] | | |
| | Conv.NF | 0.71[bc] | | | 1.28 | | | 1.30[ab] | | |
| | Conv.GS56 | 0.67[ab] | | | 1.26 | | | 1.25[a] | | |
| | Conv.CS14 | 0.76[c] | | | 1.35 | | | 1.56[cd] | | |
| | Conv.CS56 | 0.64[ab] | | | 1.24 | | | 1.71[d] | | |

**Conclusion** Conv. milk fed calves consume more concentrate when offered chopped straw from 14 days of age. Significant interactions were found between forage and MR x forage on water intake, with calves fed conv. MR and provided with grass silage consuming less water than conv. Calves offered chopped straw from 56 d of age.

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## References

Castells, L., Bach, A., Aris, A., Terré, M. 2013. Journal of Dairy Science 96(8), 5226-5236

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# An investigation of serum IgG concentrations and rates of passive transfer among calves on commercial Irish dairy farms

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**Application** High serum IgG concentrations and rates of passive transfer, found among calves on Irish dairy farms, are reflective of good colostrum management, and reduce the risk of morbidity and mortality. Colostrum management standards, however, could be improvement among larger herds, and in the second half of the calving season.

**Introduction** Neonatal calves are immunologically naïve, and must acquire passive immunity immediately after birth (Barrington and Parish, 2001). Colostrum, the first mammary secretion following parturition, can provide such immunity, once it contains an immunoglobulin G (IgG) concentration ≥ 50 mg/ml, and a sufficient quantity is received by the calf in a timely manner (Godden, 2009). Failure to achieve adequate passive transfer of immunity is identified as a serum IgG concentration < 10 mg/ml after 24 hours of life, and predisposes calves to an increased risk of morbidity and mortality (Weaver *et al*., 2000). The aim of this study was to evaluate serum IgG concentrations, and rates of passive transfer among calves on commercial Irish dairy farms.

**Material and methods** Spring calving (February to April) commercial Irish dairy herds (n=47), with a herd size of ≥ 70 cows, were enrolled in the study. These were located in the Munster region of Ireland, and each farm was visited on two separate occasions. The first visit occurred early in the calving season, while the second visit was conducted in the latter half of the season. During each visit calves >24hrs and < 6 days old were identified. Blood samples were taken from these animals via the jugular vein using a plain serum tube (10ml BD Vacutainer®, BD, Belliver Industrial Estate, Plymouth PL6 7BP. UK). Samples were then kept on ice until placed in refrigeration at 4°C for 24 hours prior to serum separation by centrifugation (3000 g x 30 mins). Following centrifugation, serum samples were collected and frozen at -20°C until IgG concentration determination.Serum samples were later analysed in duplicate, at 1:2 dilutions, using radial immunodiffusion kits (Triple J Farms, Bellingham, Washington, USA). Variables associated with serum IgG concentration were investigated using a mixed model in PROC MIXED (SAS Institute Inc., Cary, NC, Version 9.1). Independent variables included in the model were herd size category, calving commencement week of year, and visit number. Visit number was included as a repeated measure. Herd sizes were categorised from 1 to 4; category 1 = 74-107 cows, category 2 = 110-138 cows, category 3 = 140-189 cows, and category 4 = 210-385 cows.

**Results** Mean serum IgG concentration was 30.31 (SD 14.19) mg/ml and ranged from 0 to 86.38 mg/ml. The proportion of samples with a concentration < 10 mg/ml IgG was 8.48 %. Herd size category ($P<0.05$) had an effect on serum IgG concentration. For category one, serum IgG concentration was higher (31.73 mg/ml; SE= 1.74; $P=0.25$) than category two (28.84 mg/ml; SE=2.04), three (28.05 mg/ml; SE=2.22; $P=0.17$) and four (23.94 mg/ml; SE= 2.39; $P<0.01$). No difference existed between category two and category three ($P=0.7731$), and four ($P=0.0841$), and between category three ($P=0.1543$) and category four. Farm visit ($P<0.05$) had an effect on serum IgG concentration, which was higher in visit one (29.74 mg/ml; SE=1.36; $P<0.05$) than in visit two (26.54 mg/ml; SE=1.54). Maintaining high standards over a prolonged period of time (10-12 weeks), in labour intensive spring calving systems, can prove difficult. This could potentially be the cause of differences between visit one and two. Calving commencement week of year ($P=0.097$) had no effect on serum IgG concentration.

**Conclusion** High levels of passive transfer of immunity (91.52 %) are being achieved within Irish dairy herds; however calf serum IgG concentrations are influenced by the size of the herd within which they are born, and if born early or late in the calving season. This indicates that improved colostrum management is required among larger herds (>107 cows), and irrespective of herd size, standards achieved early in the calving season must be maintained throughout.

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**References**

Barrington, G.M., and Parish, S.M. 2001. Veterinary Clinics of North America: Food Animal Practice 17(3), 463-476.
Godden, S.M., Haines, D.M., Konkol, K., and Peterson, J. 2009. Journal of Dairy Science 92(4), 1758-1764.
Weaver, D.M., Tyler, J.W., VanMetre, D.C., Hostetler, D.E. and Barrington, G.M. 2000. Journal of Veterinary Internal Medicine 14(6), 569-577.

# The effects of supplementing drinking water with molasses on the performance of UK Holstein-Friesian heifer calves

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**Application** Highlighting the importance of the provision of water within dairy heifer rearing systems, supplementation with molasses offers potential growth and health benefits in pre-ruminant calves possibly by promoting earlier rumen development

**Introduction** Growth and health have great importance within dairy calf management (Huuskonen *et al.,* 2011). Although positive correlations between water and concentrate intakes have been reported (Kertz *et al.,* 1984), the focus for improvement within heifer rearing practices is often based around colostrum, liquid feed and concentrate intakes, with no regard for the fundamental importance of water. Thomas *et al.* (2007) identified a correlation between the palatability of drinking water and calves intakes; however, there is limited information on the associative effects on concentrate intakes and health traits. Therefore, the aim of this research project was to examine the effects of 0%, 5% and 10% inclusion of molasses in drinking water on: water and concentrate intakes, weight gain and disease incidence.

**Material and methods** Holstein-Friesian heifer calves (n=27) were recruited to the trial at 7 days of age. They were allocated (n=7 per group) to one of three drinking water treatments - 0% molasses (0Mo), 5% molasses (5Mo) or 10% molasses (10Mo) using stratified randomisation to take account of birth weight and dam parity. 0Mo calves were offered tap water at ambient temperature, 5Mo calves were offered tap water containing molasses (CP 6.5%, ME 12.7 MJ/kg DM; Economol, ED&F Man, London) at 5% (v/v), 10Mo (v/v), calves were offered water containing 10% molasses. Calves were offered 2-5 litres of their water treatment each day, dependent upon individual intake. Any water remaining was measured the following morning. The calves were housed indoors in individual metal pens (1.0X1.8m) bedded on wheat straw. Calf coats were provided to all calves when ambient temperature dropped below 15 $^{o}$C. Calves were fed calf milk replacer (CP 26%, oil 16%; 150g CMR per litre; Heiferlac, Volac Ltd.) on a rising plane of nutrition from 5 l/day at birth to 7 l/day by day 45. All calves had *ad libitum* access to a coarse calf starter (CP 18%, ME 11 MJ/kg DM; Heritage, H.J. Lea Oakes) and wheat straw offered in racks. Measurements included daily intakes of water (Kg), starter feed (Kg) and CMR (l) and the incidence of respiratory disease and diarrhoea. Liveweight (Kg) was determined at 7 days of age and then weekly using a Volac weigh-band. Daily liveweight gain (DLWG) was calculated from weight change each week. Data were analysed using repeated measures ANOVA, correlation and regression analysis, and Chi$^2$.

**Results** DLWGs were higher in calves offered molassed water (Table 1). However, there were no effects on liveweight, daily concentrate and water intakes or total concentrate and water intakes. There were significant effects of time on all parameters. There were no significant interactions between time and treatment for DLWG (P=0.702). The linear regression analysis of DLWG (Kg/day) against daily concentrate intake (Kg) in week seven indicated a significant (P=0.012) effect of concentrate intake (Kg) on DLWG (Kg/day) with a modest positive correlation ($R^2$=0.33). The incidence of scours was higher (P<0.05) in 0Mo (66.7%) and 5Mo (33.3%) than 10Mo (0%). Pneumonia incidence was also higher (P<0.05) in 0Mo (55.6%) and 5Mo (11.1%) than 10Mo (0%).

**Table 1** Effects of supplementing water with 10% (10Mo), 5% (5Mo) or 0% (0Mo) molasses on performance of Holstein-Friesian heifer calves (n=9 per treatment).

| Parameter | Treatment | | | S.E.M. | Probability | | |
|---|---|---|---|---|---|---|---|
| | 0Mo | 5Mo | 10Mo | | Treatment | Time | Interaction |
| DLWG (Kg/day) | 0.49 | 0.64 | 0.70 | 0.152 | 0.011 | <0.001 | 0.702 |
| Liveweight (Kg) | 55.8 | 56.7 | 60.3 | 0.65 | 0.260 | <0.001 | 0.317 |
| Daily Concentrate Intake (Kg/day) | 0.20 | 0.27 | 0.31 | 0.040 | 0.141 | <0.001 | 0.238 |
| Daily Water Intake (Kg/day) | 0.40 | 0.31 | 0.52 | 0.137 | 0.160 | <0.001 | 0.317 |
| Total Concentrate Intake (Kg) | 2.85 | 3.54 | 4.40 | 0.261 | 0.157 | <0.001 | 0.137 |
| Total Water Intake (Kg) | 7.13 | 5.21 | 7.67 | 0.553 | 0.383 | <0.001 | 0.108 |

**Conclusion** These results suggest inclusion of molasses in the drinking water of pre-weaned Holstein-Friesian heifers increased DLWG (Kg/day). The results suggest positive benefits to health from supplementing water with molasses, as calves on treatments 10Mo and 5Mo reported less scour and respiratory disease than those on 0Mo.

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**References**

Huuskonen, A., Tuomisto, L. and Kauppinen, R. 2011. Journal of Dairy Science, 94 (5), 2475-2480.
Kertz, A.F., Reutzel, L.F. and Mahooney, J.H. 1984. Journal of Dairy Science, 67 (12), 2964-2969.
Thomas, LC, Wright, TC, Formusaik, A, Cant, JP and Osbourne, VR. 2007. Journal of Dairy Science, 90 (8), 3831-3837.

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# Blood immune transcriptome analysis of artificially fed dairy calves and naturally suckled beef calves from birth to 7 days of age

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**Application** This study investigated the molecular mechanisms involved in the developing immune response of neonatal beef and dairy calves. Identified genes of interest have potential as biomarkers of immunocompetence in future studies.

**Introduction** Neonatal calves possess an immature and naïve immune system and are reliant on the intake of maternal colostrum. Variation in colostrum management in beef and dairy calves is thought to affect early immune development. Therefore, the objective of this study was to examine changes in gene expression and investigate molecular pathways and biological functions involved in the immune-competence development of neonatal Holstein dairy calves and naturally suckled beef calves using next generation RNA-sequencing during the first week of life.

**Material and methods** Jugular whole blood samples were collected into RNA tempus tubes from Holstein dairy calves (n= 8) artificially fed 5% B.W. colostrum, and from naturally suckled Charolais-Limousin (CL; n = 7) and Limousin-Friesian beef calves (LF; n = 7), for subsequent RNA isolation. Blood samples were harvested at 0, 48, 72 and 168 hours (h) post-birth. mRNA was isolated from the whole blood, cDNA libraries prepared from mRNA and subsequently sequenced with single-end reads. RNAseq processing was carried out as described by Johnston *et al*., 2016. Quality assessment of the filtered data was performed by FastQC and reads were UMD3.1 *Bos taurus* genome. Read alignment was performed using STAR. Analysis of the count data was performed using DESeq2, and functional analysis of DEG was carried out using IPA.

**Results** Dairy calves were first compared to LF beef calves (Figure 1A). At 48h post-birth, comparison of LF to dairy calves highlighted a number of biologically interesting networks as enriched, including haematological system development and function. Top canonical pathways enriched included *IL-1* and *IL-8* signalling were both upregulated in dairy calves. At 72h post-birth, a significant number of pro-inflammatory cytokines, including *IL-2* complex, were upregulated in dairy calves. At 168h post-birth, comparison of dairy to LF calves highlighted enrichment of a number of networks including humoral immunity. A number of upstream regulators such as *TNF* and *LPS* were present at lower levels in LF calves. Dairy calves were also compared to CL beef calves (Figure 1B). Enriched networks included cell mediated immune response. Upstream regulators activated in beef CL calves after 48h included *IG,* with a significant number of pro-inflammatory cytokines upregulated in dairy calves. Comparison of beef CL to dairy at 72h highlighted *IL-6* and *IL-8* signalling as upregulated canonical pathways in dairy calves. At 168h post-birth, comparison of CL to dairy calves resulted in the enrichment of cell maintenance and structure, and also inflammatory response, which were upregulated in dairy calves at 168h. Upstream regulators identified during the comparison included a number of cytokines which were upregulated in dairy calves and immunoglobulin which was upregulated in CL at 168h post-birth.

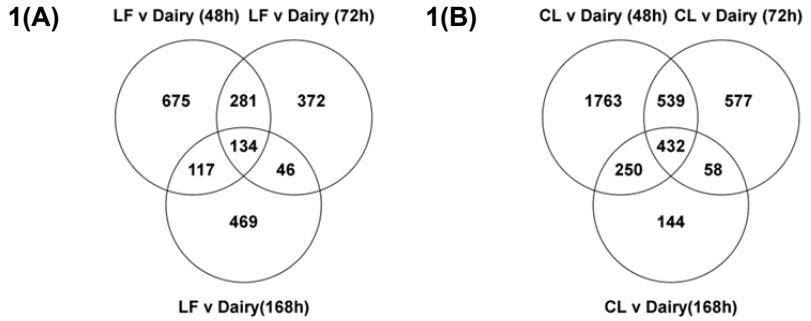


**Figure 1** DEG identified in the comparison of LF beef calves (Figure 1A) and CL beef calves (Figure 1B) to dairy calves at 48h, 72h and 168 timepoints.

Pathway and functional analysis were performed using ingenuity pathway analysis (IPA).

**Conclusion** These data provide a greater understanding of the molecular control of the early development of the neonatal immune system of dairy and beef calves, highlighting some of the molecular mechanisms regulating the immune response, likely due to variations in colostrum ingestion. Dairy calves initially demonstrate a surge in pro-inflammatory cytokines with major differences observed between beef and dairy calves at 168h post-birth including increased abundance of *IG* in beef CL calves.

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**Reference**

Johnston, D., Earley, B., Cormican, P., Kenny, D.A., McCabe, M.S., Kelly, A.K., *et al*., 2016. PLoS ONE11(8).

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# An investigation into the unique attributes of a premium beef scheme competing in an international market

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**Application** Premium beef producers can differentiate their produce through the emphasis of a number of unique characteristics. This will enable beef producers to successfully compete in global markets.

**Introduction** This paper uses the resource-based view (RBV) theory to examine how a premium beef producer can obtain a competitive advantage in an international market. Seminal research in this area has shown that small agri-food producers can compete in global markets by pursuing a product differentiation strategy and targeting a niche market of consumers (Humphrey, 2006; Dentoni and Reardon, 2010). When a firm uses a differentiation strategy, it can uniquely position itself from its competitors and receive a price premium. Therefore, through the provision of high quality premium food products, producers can differentiate their products and meet consumers' needs in a global marketplace.
The objectives of this research were to identify the unique attributes of successful agri-food international supply chains; and to determine how a premium beef supply chain can achieve a competitive advantage in an international market.

**Material and methods** To meet the aims of the study a qualitative method approach was used. More specifically, this research used case studies to explain how rural agri-food entrepreneurs could build global premium brands through the use of their unique resources. Case study research involves studying one or more situations in depth, and it is the recommended methodology to use in order to gain a rich and comprehensive understanding of the phenomena being investigated (Yin, 2003). Seven case studies of best practice within Northern Ireland (N.I.) and the Republic of Ireland (R.O.I.) were identified through 'Expert Interviews' with DAERA (N.I.) and Bord Bia (R.O.I). A benchmarking analysis was then completed using NVivo to identify the key attributes for optimal performance.

**Results** Premium agri-food producers that have achieved success in international markets exhibit the VRIN (valuable, rare, inimitable and non-substitutable) criteria. Initial analysis on the unique attributes/resources using Nvivo has identified a number of key attributes (including Selective Distribution, Nutritional Benefits, Heritage, Certifications, Social Image and Endorsements). These attributes are prevalent in successful premium international food supply chains. In this pre-Brexit era this could have significant implications for agri-food producers and supporting government agencies.

**Conclusion** As BREXIT approaches for the UK economy, rural agri-food producers face uncertainty in exporting markets. However, rural entrepreneurs with premium agri-food products can take practical steps such as using selective distribution channels and highlighting quality certifications, this will ensure their products are capable of meeting consumers' needs in international markets.

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**References**

Barney, J. 1991 Journal of Managemen, 17 (1), 99.
Dentoni, D. and Reardon, T. 2010. Journal on Chain and Network Science 10 (3), 159-171.
Humphrey, J. 2006. The European Journal of Development Research 18 (4), 572.
Yin, R.K. 2003. Case Study Research: Design and Methods. Thousand Oak, CA: Sage.

# Diet and animal factors affect methane and carbon dioxide emissions from beef cattle in temperate grassland system

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**Application** In beef cattle, enteric emissions are driven by feed and animal factors. Manipulating feed type (e.g., forage type) and animal factors (e.g. body weight) may mitigate greenhouse gas emissions from beef cattle in UK grassland systems.

**Introduction** The livestock sector contributes c. 14.5% of all anthropogenic greenhouse gas (GHG) emissions (Ripple *et al*., 2014). Cattle emit 4.6 $GtCO_2eq$/year with beef and dairy cattle accounting for 2.5 $GtCO_2eq$/year and 2.1 $GtCO_2eq$/year, respectively (Herrero *et al*., 2016). Sustainable intensification relies on increasing yields while simultaneously decreasing the environmental impacts (Eisler *et al*., 2014) by adopting diet and animal based mitigation strategies (Misselbrook *et al*., 2013). Life cycle impact assessment at North Wyke Farm Platform (NWFP, Rothamsted Research) has demonstrated that the uncertainty in emission measurements can be reduced by assessing emissions at an individual animal level (McAuliffe *et al*., 2017). Here, we studied how dietary composition influences enteric emissions and emission intensities, and how it is influenced by animal factors including breed at individual animal level in beef cattle in grazing systems.

**Material and methods** Ninety beef cattle (n=90) were used at the NWFP to assess the influence of the forage characteristics and animal factors on enteric emissions. The NWFP consists of three 'farmlets' each of approximately 22 ha with three contrasting pasture forages: 1) permanent pasture (PP); 2) reseeded pasture with a high sugar grass variety (HS); 3) reseeded pasture with a mixture of high sugar grass and white clover (WC, see McAuliffe *et al*., 2017). Silage made from these swards was supplied to 30 growing beef cattle on each farmlet (balanced for breed and sex) during winter housing (average live weight = 336.7 ± 62.21 kg). Enteric $CH_4$ and $CO_2$ was measured on the three farmlets for 5 weeks using the GreenFeed individual animal measurement technique (GEM). Animals were given individual free access to the GEM with an ear-tag identification and were trained to the GEM for 3 weeks before starting the trial. Drinking water and silage was given ad lib. A small quantity of concentrates (210 ± 7 g/visit) was fed to the cattle via the GEM, to encourage its use. The $CH_4$ and $CO_2$ emissions (g/day) were determined from $CH_4$ and $CO_2$ concentrations, corrected by ambient air concentrations determined when no animal was using GEM, air flow in the collection pipe, rate of air capture and air temperature. The daily $CH_4$ and $CO_2$ measurements based on each visit were averaged across all values in a two-week period to give a single value for each animal estimated by averaging multiple emission measurements made over all visits per day to the GEM. The effects of diet and animal factors on the two response variables -$CH_4$ and $CO_2$ emissions were tested using separate linear mixed models with animal as the random term and all the other included variables as fixed terms. The statistical tests were performed using Genstat 18 (VSNI).

**Results** The results show significant effect of diet (forage treatment) and animal factor (body weight) on daily $CH_4$ and $CO_2$ emissions measured at individual animal level (Table 1).

**Table 1** Results of the linear mixed- effect models showing influence of forage treatment and birth weight on daily $CH_4$ and $CO_2$ production in beef cattle. Significant p-values ($p<0.05$) are given in bold.

| | Forage treatment | | | | Body weight | | | |
|---|---|---|---|---|---|---|---|---|
| | n.df | F-value | d.d.f | P value | n.df | F-value | d.d.f | P value |
| $CH_4$ production (g $d^{-1}$ $animal^{-1}$) | 2 | 7.65 | 71 | **<0.001** | 1 | 53.91 | 78 | **<0.001** |
| $CO_2$ production (g $d^{-1}$ $animal^{-1}$) | 2 | 3.38 | 71 | **0.039** | 1 | 142.41 | 77 | **<0.001** |

**Conclusion** The findings offer evidence regarding animal parameters and forage characteristics as key drivers of enteric emissions in the UK grassland systems.

**Acknowledgements** This work was supported as part of Rothamsted Research's Institute Strategic Programme – Soil to Nutrition (BB/PO1268X/1), using the North Wyke Farm Platform (BB/J004308/1), both funded by the UK BBSRC.

**References**

Eisler, M. C., Lee, M. R., Tarlton, J. F., Martin, G. B., Beddington, J., Dungait, J. A and Misselbrook, T. 2014. Nature 507, 32-34.
Herrero, M., Henderson, B., Havlík, P., Thornton, P. K., Conant, R. T., Smith, P and Butterbach-Bahl, K. 2016. Nature Climate Change 6, 452-461.
McAuliffe, G. A., Takahashi, T., Orr, R. J., Harris, P., and Lee, M. R. F. 2017. Journal of cleaner production 30, 1-9.
Misselbrook, T., Del Prado, A., and Chadwick, D. 2013. Agricultural and Food Science 22, 93-107.
Ripple, W. J., Smith, P., Haberl, H., Montzka, S. A., McAlpine, C and Boucher, D.H. 2014. Nature Climate Change 4, 2-5.

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# Genome-wide association study of economically important traits in Irish Charolais and Limousin cows

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**Application** Genetic variants associated with economically important traits could be incorporated into the genomically assisted beef cattle breeding programme in Ireland and ultimately improve the sustainability of the national herd.

**Introduction** Reproductive efficiency and longevity are key drivers of profitability in beef cow herds (Diskin and Kenny, 2016). Incorporation of genetic variants associated with such traits into the Irish genomically assisted beef cattle breeding programme would improve the sustainability of the national herd. We conducted a genome wide association study (GWAS) to identify genetic variants associated with fertility, longevity and other economically important traits in Charolais and Limousin cows, the two most predominant beef cattle breeds in Ireland.

**Material and methods** Records from 12,548 purebred Charolais and Limousin cows, genotyped using the custom built International Dairy and Beef SNP chip (IDB; version 2; Mullen *et al.*, 2013) were available for this study. Genotypes were subsequently imputed to IDBv3 density using FImpute version 2.2. All genotypes that reached the IDBv3 density of 50K SNPs were uploaded to the Golden Helix (version 8.7.1) platform. Quality control (QC; Laurie *et al.*, 2010) analysis was carried out and genotypes merged with cow fertility related phenotypes, collated from the Irish Cattle Breeding Federation database. The traits included carcass weight, live weight, estimated feed intake, gestation length, age at first calving, direct calving difficulty, maternal calving difficulty (MCD), docility, calving interval, cow survival and mortality. Following the merging of genotypic and phenotypic data files, a final cohort of 12,439 animals, consisting of 7,239 Limousin and 5,200 Charolais cows were used for a genome wide association study (GWAS). Analysis was carried out using Golden Helix software, employing a mixed linear model for each breed (Kang *et al.*, 2010). This resulted in the generation of a breed specific dataset (n=2) for each of the traits of interest. Following on from GWAS, data for each of the two breeds were subsequently subjected to a meta-analysis (METAL software; Willer *et al.*, 2010) in order to identify SNPs associated with the traits of interest across both cow breeds. All SNPs that reached a nominal statistical significance of 0.05 were subjected to Ingenuity pathway analysis

**Results** Genetic variants with an associated meta-analysis based P-value > $5x10^{-5}$, consistent with the statistical threshold employed by the Wellcome Trust (Burton *et al.*, 2007) were determined to have genome-wide significance (Table 1). Enriched pathways associated with the fertility related traits included the STAT3 pathway and IL-3 signalling.

**Table 2** Association between statistically significant SNPs and selected cow traits

| rsID | P-value | Trait | Chromosome_Megabase | Nearest gene |
|---|---|---|---|---|
| rs133745645 | 4.61E-05 | Age at first calving | 3_113 | *GIGYF2* |
| rs41592228 | 2.17E-06 | Calving interval | 8_84 | *HSD17B3/SLC35D2* |
| rs111030667 | 1.78E-05 | Carcass weight | 20_23 | *ANKRD55* |
| rs109195906 | 1.84E-06 | Docility | 13_6 | *BTBD3* |
| rs109195766 | 8.68E-06 | Feed intake | 21_41 | *TTYH3* |
| rs109842672 | 3.21E-06 | Gestation length | 28_17 | *RHOBTB1* |
| rs43509004 | 3.49E-05 | Gestation length | 7_23 | *ACSL6* |
| rs109521286 | 2.64E-05 | MCD | 1_18 | *TMPRSS15* |
| rs110708213 | 2.82E-05 | Mortality | 10_26 | *OR6S1* |

**Conclusion** Following validation, SNPs identified as statistically significant could be included into the Irish national genomic selection programme as markers for cow fertility and longevity. Further investigation into the genes and pathways highlighted in this study may improve our understanding of the biology underpinning fertility and longevity in cattle.

**Acknowledgements** This study was funded by the Irish Department of Agriculture, Food and the Marine (Ref: 11/S/104)

**References**

Burton, P.R., Clayton, D.G., Cardon, L.R. *et al.* 2007. Nature 447, 661-678.
Diskin, M. and Kenny, D. 2016. Theriogenology 86, 379-387.
Kang, H.M., Sul, J.H., Zaitlen, N.A., Kong,.S-y., Freimer, N.B., Sabatti, C. and Eskin, E. 2010.
Laurie, C.C., Doheny, K.F., Mirel, D.B., *et al.* 2010. Genetic Epidemiology 34, 591-602.
Mullen, M.P., McClure, M.C., Kearney, J.F., *et al.* 2013. Interbull Bulletin.
Willer, C.J., Li, Y. and Abecasis, G.R. 2010. Bioinformatics 26, 2190-2191.

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# Evaluation of production efficiencies among beef and dairy crossbred suckler cows in early lactation

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**Application** Availability of production efficiency evaluations of lactating beef and dairy x beef cows at pasture should enable breeders to select for improved performance of the beef suckler cow herd through replacement strategy implemented.

**Introduction** Efficient and profitable suckler beef production must be the core elements of technical efficiency; cost reduction through efficient utilisation of pasture and increased output through improved animal performance. Where feed costs account for over 75% of direct costs in beef production (Finneran *et al*., 2010), utilising grass, the cheapest feed, source is vital. In a pasture based system, suckler cows can consume 85% of total herd feed requirements (McGee, 2009), with two-thirds of this energy required for maintenance. Selecting cows that efficiently convert herbage intake to beef output is therefore critical to sustainable beef production. Consequently, the objective of this study, carried out at Teagasc Grange, Dunsany, Co. Meath, Ireland, was to determine individual estimates of grass dry matter intake (GDMI) and compare the production efficiency of beef and dairy x beef (DBX) crossbred suckler cows during early lactation at pasture.

**Material and methods** Data were available from 91 cows during early lactation in 2016; 36 beef and 55 DBX cows. Animals were selected from Aberdeen Angus and Limousin sires. Animals were managed under a rotational grazing system with an average pre- and post- grazing height of 10.5 (s.d. 1.4) cm and 4.3 (s.d. 0.3) cm, respectively. Cow live weight (BW) and body condition score (BCS; 0 to 5) were recorded every three weeks. The weigh-suckle-weigh technique was used at 52 ± 5.6 days in milk (DIM) in 2015 to establish cow milk yield during early lactation at pasture. Individual estimates of GDMI were established using the n-alkane technique (Dillon, 1993) at 58 ± 5.6 DIM. The measures of gross efficiency calculated were milk yield per unit intake and intake capacity which was expressed as GDMI per 100 kg BW. Energetic efficiency of residual feed intake (RFI) was also estimated by regressing energy intake on its assumed components of metabolisable BW, BW change, milk yield and BCS. Data were analysed using mixed models in PROC HPMIXED (SAS Inst. Inc., Cary, NC). Fixed effects included in all models were cow type (beef or DBX), cow parity (1 and 2), sire breed (AA or LM) and DIM.

**Results** Beef cows were 62 kg ($P<0.001$) heavier and had 0.23 greater BCS ($P<0.05$) than DBX in early lactation. The DBX produced 1.1 kg more milk ($P<0.05$) and had a greater milk yield per unit intake (0.12 kg; $P<0.01$) than beef cows. No difference was observed in overall GDMI between cow types, however DBX had 0.23 kg greater intake per unit BW ($P<0.01$) than beef cows (Table 1).

**Table 1** Effect of cow type (with the associated weighted standard error of the mean and P-value) on BW, BCS, GDMI, milk yield, gross efficiency measures and RFI

| | Beef | Dairy x Beef | s.e. | P-value |
|---|---|---|---|---|
| BW (kg) | 592 | 530 | 8.9 | <0.001 |
| BCS (0 to 5) | 2.81 | 2.58 | 0.054 | <0.01 |
| Milk yield (kg) | 6.3 | 7.4 | 0.33 | <0.05 |
| GDMI (kg/d) | 13.1 | 13.2 | 0.37 | NS |
| GDMI/100kg BW (kg) | 2.16 | 2.39 | 0.056 | <0.01 |
| Milk Yield/GDMI (kg) | 0.48 | 0.60 | 0.029 | <0.01 |
| RFI (UFL) | -0.40 | 0.26 | 0.310 | 0.1483 |

**Conclusion** Results from the current study showed that beef cows were heavier and had a better BCS during early lactation. However there was no difference observed in estimated individual GDMI between beef and DBX during early lactation. Production efficiency measures showed DBX had a greater milk yield, milk yield per unit intake and intake per unit BW than beef cows.

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**References**

Dillon, P. 1993. PhD, National University of Ireland, Dublin.
Finneran, E., Crosson, P., O'Kiely, P., Shalloo, L., Forristal, D. and Wallace, M. 2010. Journal of Farm Management 14, 95-116.
McGee, M. 2009. Irish Grassland Association Journal 43, 125-131.

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# Weaned calf output in relation to suckler cow liveweight in purebred Luing, and crossbred Aberdeen Angus x Limousin and Limousin x Aberdeen Angus beef cows

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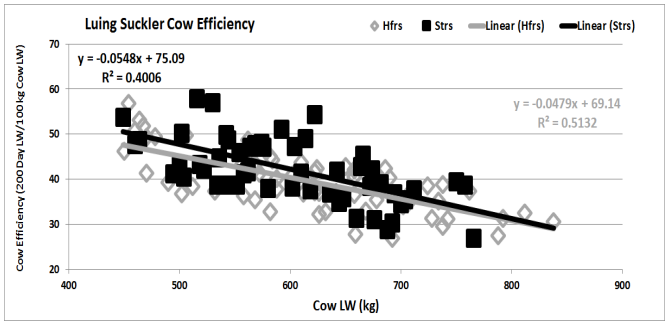
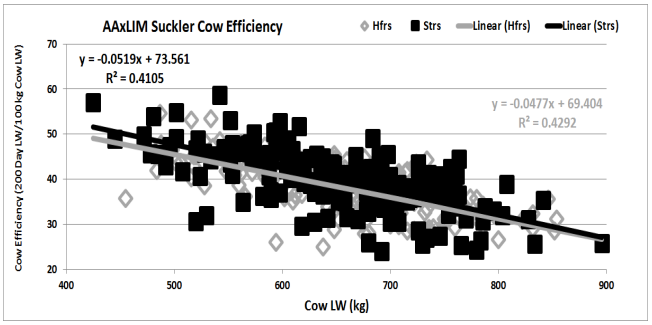
**Application** Farmers should be encouraged to evaluate the efficiency of their own suckler herd weaned calf output using this routine "suckler cow efficiency" measure as an annual aid to both technical and financial management of their business.

**Introduction** Increasing cow size is recognised as one of the determinants of suckler system input costs whilst the weight of calf output at weaning is an important measure of output value. The weight of suckled calf output in relation to the size of the cow (often termed suckler cow efficiency) is one of the major comparative efficiency measures of suckler beef production systems. The objective of the current analysis was to examine the relationships between weaned calf weight and cow size across three distinct suckler cow breed types.

**Material and methods** As part of the long term policy at the SRUC Beef and Sheep Research Centre (BSRC), south of Edinburgh, three distinct herds of spring calving cows were maintained under common management for the years 2012-2016. A purebred herd of Luing cows and two sub-herds of Aberdeen Angus x Limousin (AAxLIM) and Limousin x Aberdeen Angus (LIMxAA) based on a 2-breed reciprocal crossbreeding programme were maintained on the upland farm at BSRC. Routine farm management practices included weighing cows and calves at weaning in autumn along with body condition score measurements. The "suckler cow efficiency" measure was calculated by scaling the individual 200 day calf liveweight (200D LW) measurement of each calf to its dam's liveweight (LW) measured at the same time. REML and linear regression analysis of this routine "suckler cow efficiency" measure (n=772) was undertaken using Genstat 16.

**Results** Average (s.e.) cow LWs (kg) and calf 200D LWs (kg) were 609 (8.1), 653 (4.60), 623 (6.23) and 244 (2.81), 251 (1.83), 230 (2.13) for Luing, AAxLIM and LIMxAA cows and calves respectively. Suckler cow efficiency values (200D calf LW/100 kg cow LW) declined as cow LW increased as expected (Figure 1) and for both steers and heifers were significantly (P<0.05) higher in Luing compared to either AAxLIM or LIMxAA cows (Table 1).



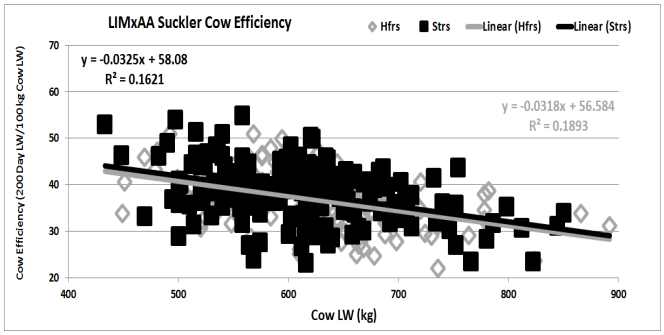




**Table 1** Average suckler cow efficiency (200D calf LW/100 kg cow LW)

| | Luing | AAxLIM | LIMxAA | s.e.d. | Sig. |
|---|---|---|---|---|---|
| Steers | 42.9$^{a}$ | 40.2$^{bc}$ | 38.4$^{cd}$ | 0.962 | * |
| Heifers | 40.9$^{b}$ | 38.9$^{cd}$ | 37.1$^{d}$ | | |
| Average | 41.9$^{a}$ | 39.6$^{b}$ | 37.8$^{c}$ | 0.683 | ** |

Values not sharing common superscripts differ (P<0.05)

**Figure 1** Suckler cow efficiency relationships in Luing, AAxLIM & LIMxAA suckler cows

**Conclusion** Cows producing calves at above average weights (the regression lines on the graphs) may be suitable as dams of replacement heifers whilst those with lower than average values may not. Luings had higher suckler cow efficiency values than either AAxLIM or LIMxAA cows and AAxLIM cows had higher values than LIMxAA cows.

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# Effect of production system on the performance of growing Holstein bulls

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**Application** *Ad libitum* concentrate feeding led to significantly greater weight gains, however there was no difference in feeding *ad libitum* concentrates at grass or while housed. Supplementing 2kg concentrates at grass resulted in significantly greater live weight at housing compared to those with no concentrate supplementation.

**Introduction** Holstein bull beef production is commonly an intensive, *ad libitum* concentrate indoor system. However, the economic viability of such a system is often contested. Consequently, with an increasing focus on gaining more from grass; the inclusion of a grazing period in bull beef production systems may offer the opportunity to greatly reduce production costs. The objective of this trial was to investigate the effects of four production systems on the performance of Holstein bulls during their first summer.

**Material and methods** A 2 (autumn or spring born (A)) x 4 (production system (PS)) factorial design experiment involving 112 Holstein bulls was undertaken at AFBI, Hillsborough. The four production systems differed during the summer with treatments including (i.) housed with *ad libitum* access to concentrates and silage (HA) (ii.) grazed with *ad libitum* access to concentrates (GA) (iii.) grazed with 2 kg/day concentrate supplementation (G2) and (iv.) grazed with no concentrate supplementation (G). A total of 56 autumn born (AB) bulls had a mean initial age of 191 days and 196kg live weight; while the 56 spring born (SB) bulls had a mean initial age of 106 days and live weight 106kg. Each treatment group consisted of 14 animals which were balanced for weight and age. All HA cattle were housed on slatted accommodation with access to cubicles, while SB HA also had access to a straw bedded creep area. GA, G2 and G were rotationally grazed in 7 day paddocks from 26th May 2017. Bulls were offered fresh feed daily, concentrates were fed either in a trough (HA, G2) or creep feeder (GA) and grass silage (HA) was fed through a diet feeder. HA and GA were gradually built up to *ad libitum* concentrate feeding, at the same rate over a period of 8 weeks for AB cattle and 4 weeks for SB cattle. G2 were fed 2kg from the beginning of the trial. AB cattle were housed on 24$^{th}$ August 2017, after 90d at grass, while SB cattle were housed on 11$^{th}$ October 2017, following 138d at grass. Bulls were weighed fortnightly to monitor performance. Grass and silage samples were analysed weekly using NIRS, while concentrates were analysed via a wet chemistry analysis. Pre- and post-grazing heights were measured using a Jenquip rising plate meter, and the difference used to estimate intake. Concentrate and silage intakes were monitored daily.

**Results** The concentrate consisted primarily of maize, rolled barley, US distillers and corn gluten at 20, 19.9, 19.8, 14.9 g/kg respectively. Concentrate dry matter (DM), crude protein (CP) and metabolisable energy (ME) were 87.1%, 150g/kgDM, 11.1 MJ/kgDM respectively. Grazed grass had a DM of 14%, CP of 190g/kgDM and ME of 11MJ/kgDM. Likewise for the grass silage DM, CP and ME were 26%, 124g/kgDM and 10.6MJ/kgDM respectively. Bulls had the greatest live weight gain when receiving *ad libitum* access to concentrates (HA & GA). There was no significant difference in performance between HA & GA. Bulls on G2 were significantly heavier at housing than the G bulls. SB bulls had lower DLWG than AB bulls on the same production system. There was a significant interaction between age and production system for concentrate and estimated forage DMI.

**Table 1** Performance and intakes of Holstein bulls during the first summer in four production systems

| Age (A) | Autumn born | | | | | | Spring born | | | | | | A x PS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Production system (PS) | G | G2 | GA | HA | SEM | Sig. | G | G2 | GA | HA | SEM | Sig. | Sig. |
| Housing weight (kg) | 279.1$^{a}$ | 299.2$^{b}$ | 345.8$^{c}$ | 338.8$^{c}$ | 4.890 | <0.001 | 196.2$^{a}$ | 222.9$^{b}$ | 278.0$^{c}$ | 289.4$^{c}$ | 7.510 | <0.001 | ns |
| Housing age* (d) | 283 | 274 | 281 | 285 | 10.35 | ns | 246 | 242 | 238 | 249 | 6.040 | ns | ns |
| DLWG (kg/d) | 0.90$^{a}$ | 1.15$^{b}$ | 1.67$^{c}$ | 1.59$^{c}$ | 0.066 | <0.001 | 0.64$^{a}$ | 0.84$^{b}$ | 1.24$^{c}$ | 1.33$^{c}$ | 0.056 | <0.001 | ns |
| Concentrate DMI (kgDM/d) | 0.00$^{a}$ | 1.57$^{b}$ | 6.81$^{d}$ | 5.20$^{c}$ | 0.253 | <0.001 | 0.00$^{a}$ | 1.63$^{b}$ | 5.13$^{d}$ | 4.27$^{c}$ | 0.150 | <0.001 | <0.001 |
| Estimated forage DMI (kgDM/d) | 6.21$^{d}$ | 4.96$^{c}$ | 3.69$^{b}$ | 1.90$^{a}$ | 0.077 | <0.001 | 4.32$^{c}$ | 4.40$^{d}$ | 2.87$^{b}$ | 0.70$^{a}$ | 0.019 | <0.001 | <0.001 |

*Autumn born bulls were housed after 90 days at grass, and spring born bulls were housed after 138 days at grass

**Conclusion** *Ad libitum* concentrate feeding resulted in superior liveweight, however GA had a significantly greater concentrate DMI than HA without a significantly greater weight gain. Supplementing 2kg concentrates at grass led to greater live weight compared to no concentrate supplementation.

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# The effect of grazing system and trace element supplementation method on the performance of beef heifers

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**Application** The study shows that the implementation of an intensive grazing system can increase output per hectare and grass utilisation on beef farms.

**Introduction** There is significant scope to increase grass utilisation on UK beef farms. The average grass utilised on a beef farm is 4.1tDM/ha while the potential yield is up to 15tDM/ha. Current estimates suggest that by improving grass utilisation by 1tDM/ha combined with improvements in grass quality could increase margin over feed costs by £239/ha (Lively and Aubry, 2017). However, there is limited information available on the optimum grazing management strategies for beef cattle. Within grazing beef cattle trace element deficiencies have been identified. In addition there is limited information on the efficacy of contrasting methods of supplementing trace elements on the performance of grazing beef cattle. The aim of this study was to examine the impact of grazing system and trace element supplementation method on growing beef heifer performance and herbage utilisation**.**

**Material and methods** A 2 (grazing system) X 4 (trace element supplementation method) factorial design study using 72 Stabiliser and Limousin X Holstein Friesian heifers was carried out at AFBI Hillsborough from April to September 2017 (initial live weight 341kg and age 368 days). The swards were composed of perennial ryegrass and white clover and received the same fertiliser treatment throughout the study. There were two replicates of each grazing treatment. Treatment groups were balanced for breed and live weight whilst grazing areas were blocked according to in-field location.
*Grazing System Treatment*:

1. Traditional System: 3.5 day paddock (0.47ha), stocking rate of 5.5 animals per hectare on a 24.5 day rotation
2. Intensive System: daily area allocations which provided 2kg DM/100kg bodyweight (BW).

*Trace Element Supplementation Method*: 4 heifers from each grazing treatment were randomly allocated to 4 trace element supplementation methods; Control (no supplement), Bolus A, Bolus B, and a Drench. Boluses were given at the start of the experiment and the drench was given every six weeks also beginning at the start of the experiment at a rate of 1ml/10kgBW (i.e. 34ml for a 340kg heifer). Pre and post grazing sward heights were measured by taking 30 samples per 0.1ha in a 'W' shape across the paddock with a 'Jenquip EC10' rising platemeter. Intake and grass utilisation rate were estimated by calculating the difference in the pre and post grazing sward heights. Animal live weight was monitored weekly and live weight gain calculated by linear regression. Genstat was used to statistically analyse sward performance and also animal performance data which included initial live weight and age as co-variates.

**Results** There was a significant (P>0.001) difference between grazing treatments in post graze sward height, animal intake, grass utilisation rate and daily grazing area required (Table 1). Grass utilisation rate was 19% higher in the intensive grazing system relative to the traditional system. The intensive grazing system had a lower (P<0.001) post grazing sward height of 5.4cm vs the Traditional systems 7cm; (P<0.001), minimising the requirement for corrective sward management e.g. topping. The intensive grazing system supported an additional 33% live weight gain per hectare relative to the traditional system. This equates to an additional 328kg live weight per hectare valued at £590. There was no significant (P>0.05) difference in cattle performance due to trace element supplementation method or grazing system, nor were there any significant (p>0.05) interactions between them. The Control, Bolus A, Bolus B, and Drench treatment methods had live weight gains of 0.77, 0.87, 0.83 and 0.82kg/d respectively. Animals involved with the Traditional grazing system and Intensive grazing system had live weight gains of 0.84 and 0.80kg/d respectively.

**Table 1** The effect of grazing system on sward performance, animal intake and grass utilisation

| | Grazing System | | | |
|---|---|---|---|---|
| | Traditional | Intensive | SED | Sig. |
| Average pre graze sward height (cm) | 11.0 | 10.9 | 0.29 | NS |
| Average post graze sward height (cm) | 7.0 | 5.4 | 0.14 | P<0.001 |
| Average estimated animal intake (kgDM/hd/d) | 7.4 | 5.7 | 0.18 | P<0.001 |
| Average grass utilisation rate (%) | 54 | 73 | 1.5 | P<0.001 |
| Average area grazed per day (ha) | 0.13 | 0.09 | 0.004 | P<0.001 |

**Conclusion** The intensive grazing system produced 33% more live weight gain per hectare relative to the traditional grazing system. Neither grazing system nor trace element supplementation method had a significant effect on animal performance.

**Acknowledgements** The authors acknowledge AHDB Beef and Lamb, DAERA and Agrisearch for funding this study.

**References**

Lively, F. and Aubry, A. 2017. Proceedings of "More From Grass" Open Day, 20th June 2017, AFBI Loughgall, 21-25.

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# Comparison of ration characteristics for beef finisher cattle in north-eastern Scotland and preliminary estimates of their relationships with indicators of reticulo-ruminal health

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**Application** A high level of variation in diets among farms presents a challenge to replication in on-farm observational studies. A risk classification based primarily on the absolute amount of barley and wheat in the diet was closely linked with rumen damage. The use of reticuloruminal pH buffers can help to improve faecal consistency and overall condition of cows.

**Introduction** Sub-acute ruminal acidosis (SARA) is a digestive disease of intensively fed cattle. It is known that diets high in rapidly fermented carbohydrates lead to a number of physiological and pathological changes. Whilst many factors, both dietary and management practices adopted on farm, are believed to contribute to the onset of SARA, as yet there has been no large study of intensely fed beef cattle to examine the relationships among dietary inputs and disease risk. The primary aim of this work was to identify collaborating farms for a larger study on rumen function under diverse dietary regimes, with secondary objectives being to develop a method of scoring rumen damage that may be used and determine preliminary associations that could be tested in future studies.

**Material and methods** Each of 10 large beef producing farms in Aberdeenshire, North East Scotland was administered a questionnaire regarding the feeding management of finishing cattle, including the composition of the finishing ration, the approximate intake of each dietary component per head per day, together with the approximate ages and weights when introduced to the finishing ration and when sent for slaughter. Farms were then preliminarily classed as being at a high or low risk of acidosis based on the responses to the questionnaire. Each of the farms was visited and samples of the whole finishing ration and of the barley component were taken for further analysis of physical and chemical structure. Cattle which were due to be slaughtered were observed on each farm and a score was assigned for the general condition and contentedness of the animals, ranging from 0 to 5; 0 being poor condition (restless behaviour, poor coat condition with evidence of soiling, high number of lame beasts); 5 being very content animals (high proportion of clean animals lying down and ruminating with no evidence of lameness). The dung was assessed and scored from 0 to 5; 0 being solid dung consistency with no signs of scouring; 5 being grey, foamy scour. The chew-count per rumination was counted for 10 cattle per farm and in each pen, the percentage of cattle that were ruminating was estimated. The finishing ration from each farm was analysed chemically and physically. Percentage in dry matter (DM) of neutral detergent fibre (NDF), starch and moisture were measured using NIR. The whole ration and the barley samples were analysed using the Penn State Separator Sieve Test (PSSST). 20 continental crossbreed cattle between 12 and 24 months of age from each farm were followed through to slaughter. The rumen from each animal was examined immediately after slaughter. Individual subjective scores were assigned to each rumen based on colour of the rumen/papillae, length and width of papillae and degree of damage to the rumen wall. Samples from each rumen were collected for microscopic examination and scoring after staining with haematoxylin and eosin to reveal tissue structure. Each rumen was re-assessed after blanching and rumens were assigned a score of 0 to 4 based on the proportion of the rumen surface that assumed a black appearance; 0 being no black tissue and 4 being mainly black.

**Results** Each farm had a unique combination of dietary components. Rumen damage was significantly and positively correlated with the percentage of barley or wheat in the ration. Dung score was significantly and positively correlated with barley and wheat in the ration and with the percentage of barley particles > 19 mm. Cow condition (contentment index) was significantly and positively higher on the two farms that fed potatoes. Chew count was not significantly associated with any of the input variables. Cow condition (contentment) was higher on the 5 farms that used a buffer in their ration than the 5 farms that did not.

**Conclusion** There was a very high level of variation in diets among farms. The absolute amount of barley and wheat in the diet was closely associated with rumen damage. Reticuloruminal pH buffers were associated with improved faecal consistency and overall condition of cows.

**Acknowledgements** This project was funded by BBSRC and industry partners Harbro, AB Vista, Chr Hansen and QMS. We are very grateful to the farmers and the farm workers who collaborated in this project.

# Intake, growth, carcass and selected meat quality traits of steers offered grass silage and supplementary concentrates with increasing levels of dried corn gluten feed

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**Application** Under the conditions of this experiment, dried corn gluten feed (CGF) had a feeding value comparable to that of rolled barley/soya bean meal.

**Introduction** A variety of feedstuffs of varying carbohydrate composition are available to rectify deficiencies in nutrient supply for finishing cattle fed grass silage-based diets. There has been relatively little evaluation of dried CGF as a feed ingredient in beef rations (Erickson *et al*., 2012), and especially when offered as a supplement to grass silage. The objective of this study was to determine the effects of replacing rolled barley and soya bean meal with dried CGF in a supplement on intake, growth, carcass and selected meat quality traits of beef cattle offered grass silage.

**Material and methods** Late-maturing breed steers (initial live weight, 445 kg, SD 38.7) were blocked by sire breed and weight and from within block randomly assigned to one of four (n=12) concentrate treatments. The control concentrate comprised of 865 g rolled barley, 60 g soya bean meal, 50 g molasses, and 25 g minerals and vitamins/kg fresh weight. In the other three concentrates, barley and all the soya bean meal was replaced with 250, 500 and 750 g dried CGF (crude protein (CP), 204 g/kg dry matter (DM)) per kg fresh weight. Concentrates were prepared as coarse mixtures. Steers were individually offered 4.0 kg DM of the respective concentrates, in two feeds daily, as a supplement to grass silage (DM digestibility, 713 g/kg; CP 102 g/kg DM) offered *ad libitum* during a 134 day finishing study. Animals were weighed at the beginning and end of the study, and every 14 days throughout. Ultrasonic fat and muscle depth were measured at the beginning and end of the study. Post-slaughter, carcass weight and, carcass conformation and fat score were determined. At 48 h *post-mortem*, the lightness (L*), redness (a*) and yellowness (b*) of subcutaneous fat were recorded. After 72 h *post-mortem* pH, drip loss and L*, a*, b* colour (after 1 h bloom) of the *longissimus thoracis* muscle were recorded. Data were statistically analysed using ANOVA with terms for treatment and block in the model.

**Results** Inclusion of dried CGF in the concentrate supplement had no effect (P>0.05) on average daily live weight gain, final live weight, silage DM intake, feed conversion ratio, carcass traits (Table 1) or ultrasonic measures of body fat and muscle. Muscle colour 'L*', 'a*', 'b*', saturation or hue and subcutaneous fat 'a*', 'b*' values and saturation did not differ (P>0.05) between treatments. However, steers offered 750g CGF/kg had a higher (P<0.01) fat 'L*' value compared to those offered 250 g CGF/kg. Hue angle for subcutaneous fat colour was greater (P<0.05) for steers offered 750g CGF/kg compared to the other treatments. There was no difference (P>0.05) between treatments in muscle pH and drip loss.

**Table 1** Effect of dried CGF inclusion level in a barley-based concentrate on dry matter (DM) intake, daily live weight gain (ADG), feed conversion ratio (FCR) and carcass traits of finishing steers offered grass silage

| | Corn gluten feed inclusion level (g/kg) | | | | s.e.m. | P-value |
|---|---|---|---|---|---|---|
| | 0 | 250 | 500 | 750 | | |
| ADG (kg) | 1.04 | 1.02 | 1.06 | 0.99 | 0.043 | n.s. |
| Silage DM intake (kg/day) | 5.3 | 5.2 | 5.3 | 4.9 | 0.17 | n.s. |
| Total DM intake (kg/day) | 9.3 | 9.2 | 9.3 | 8.9 | 0.17 | n.s. |
| FCR (kg DM/ kg ADG) | 9.0 | 9.1 | 9.0 | 9.2 | 0.44 | n.s. |
| Carcass weight (kg) | 330 | 330 | 331 | 328 | 6.1 | n.s. |
| Kill-out proportion (g/kg) | 561 | 572 | 568 | 573 | 6.3 | n.s. |
| Carcass conformation score (1-15) | 8.9 | 9.3 | 9.3 | 9.4 | 0.46 | n.s. |
| Carcass fat score (1-15) | 7.7 | 6.9 | 7.5 | 7.1 | 0.37 | n.s. |

**Conclusion** Dried CGF can replace rolled barley/soya bean meal at inclusion rates up to 750g/kg in a concentrate supplement to high-digestibility grass silage without negatively affecting performance or selected meat quality traits.

**Acknowledgements** Department of Agriculture, Food and the Marine, Research Stimulus Fund - Project FEFAN-11/S/122

**References**

Erickson, G. E., Klopfenstein, T. J. and Watson, A. K. 2012. In: Makkar, Harinder P.S. Biofuel co-products as livestock feed - Opportunities and challenges, Food and Agriculture Organization of the United Nations, Rome, Italy, 77-100.

# The effect of nutritional strategy on methane emissions, milk yield and fatty acid profiles in Holstein Friesian dairy cows

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**Application** Predicting methane emissions from dairy cows using a laser methane detector and fatty acid profiles.

**Introduction** Methane ($CH_4$) emissions are a product of the enteric fermentation of ruminants. Individual dairy cows vary in the quantity of $CH_4$ they produce with estimates ranging from 278 to 456g $CH_4$/d (Garnsworthy *et al*., 2012) and this can be linked to feed intake and diet digestibility. Quantifying $CH_4$ production from individual animals can be time consuming and costly using methods such as respiration chambers or $SF_6$. $CH_4$ emissions have been associated with milk composition (Dehareng *et al*., 2012) and enteric emissions are linked to milk mid-infrared spectrometry based on the synthesis of $CH_4$ having a relationship to butterfat, and lactose production (Vlaeminck and Fievez, 2005). Non-intrusive estimates of enteric $CH_4$ can be obtained using a laser methane detector (LMD) and results can be approximated with closed chamber measurements. The aim of this study was to compare the effect of two diets (50% and 100% grazed grass) on $CH_4$ emissions from dairy cows and to test associations between LMD measurements and fatty acid profiles.

**Material and methods** Dairy cows in early to mid lactation were randomly allocated to one of two dietary treatments (14 cows per group) as part of a 12 week continuous design feeding experiment. Groups were balanced for live-weight, parity, and lactation stage, with cows milked 3 times per day. Dietary treatments consisted of 100% grazing with no housing, and 50% grazing with housing for one period of approximately 8 hours between evening and morning milking. While housed, cows were fed an ad lib total mixed ration (TMR) consisting of silage, alkalage, beans, rape meal and whey permeate with both groups of cows being fed 900g of parlour concentrate per day. Milk yield (MY) was recorded at every milking and samples were taken once per week at each of the three milkings and then combined for fatty acid profile analysis using a Milkoscan minor spectrophotometer. $CH_4$ emissions were estimated using a LMD taking two $CH_4$ readings per second, for approximately 4 minutes for each animal after midday milking three days per week using the procedure described by Chagunda *et al*. (2013). $CH_4$ readings per animal, grouped by week of experiment and reading day, were log transformed to attain normality of the data, with zero readings excluded. Thrice weekly $CH_4$ estimates were averaged per cow, and mixed models were used to test whether $CH_4$ emissions and milk production differed between treatment groups. Linear mixed effects models were fitted in R using lmer and Anova packages at a 5% significance level. Weekly means for milk concentrations of fat, protein, urea, lactose, total unsaturated FA, polyunsaturated FA, stearic acid (C18:0), short chain FA, medium chain FA and long chain FA were tested for normality, and correlated with mean weekly methane measurements per cow.

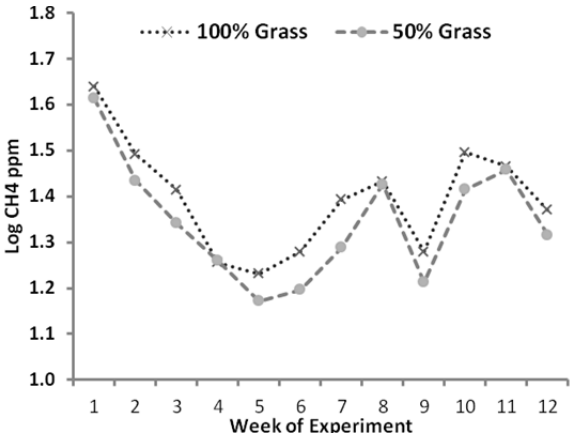


**Figure 1** Weekly group average $CH_4$ readings

**Results** Average daily MY of 28.1kg, and 30.8kg was recorded for 100%and 50% grazed groups respectively over the 12 week experiment. $CH_4$ measurements averaged 55.0 and 46.5 ppm per sample for the 100% and 50% grazed groups respectively. Mean methane emissions were significantly higher [$\chi^2$ (1) =3.416, $p<0.05$] in the 100% grazed treatment group by approximately 7.97 ppm per sample +/- 1.03 standard errors. Daily MY did not significantly differ between treatment groups across the whole experiment, however a significant difference was found from week 8 onwards; there was a weak (but ns) relationship with $CH_4$ estimates.

**Conclusion** Replacing a proportion of grazed grass in the diet with a TMR significantly lowered $CH_4$ emissions. Fatty acid profiles showed weak correlations with transformed methane measurements, nevertheless, there were differences in milk fatty acids between feed groups and further analysis will be carried out.

**Acknowledgements** This research was funded by the Scottish Government RESAS programme 2016-2021

**References**

Chagunda, M. G. G., Ross, D., Rooke, J., Yan, T., Douglas, J. L., Poret, L., McEwan, N. R., Teeranavattanakul, P. and Roberts, D. J. 2013. Acta Agriculturæ Scandinavica 63, 68–75.
Dehareng, F., Delfosse, C., Froidmont, E., Soyeurt, H., Martin, C., Gengler, N., Vanlierde, A. and Dardenne P. 2012. Animal 6:10, 1694-1701.
Garnsworthy, P., Craigon, J., Hernandez-Medrano, J. H. and Saunders, N. 2012. Journal of Dairy Science 95, 3181-3189.
Vlaeminck, B. and Fievez, V. 2005. Communications in Agricultural and Applied Biological Sciences 70, 43-47.

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# The effect of concentrate supplementation strategy on dry matter intake, substitution rate and nitrogen excretion in late lactation spring calving grazing dairy cows

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**Application** Offering concentrate supplementation to dairy cows in late lactation is important to improve cow performance and reduce nitrogen excretion when grass quality and quantity are poorer in the autumn.

**Introduction** The Irish dairy industry produces high quality dairy products from sustainable grass based systems of production (O'Brien *et al*., 1996). In the autumn when cows are in late lactation grass quantity and quality are poorer (Burke *et al*., 2008) and subsequently milk quality is poorer (O'Brien *et al*., 1996). Grass is limiting in energy and concentrate supplementation can be offered to increase energy intake, improve milk quality (Kolver *et al*., 1998), increase dry matter intake (DMI) and improve cow performance (Stockdale, 2000). However, important parameters such as DMI and substitution rate [SR; 'the decrease in pasture intake per kilogram of supplemented feed' (Kellaway and Porta, 1993)] must be considered as grass is the cheapest feed source for dairy cows and should be maximised in their diet (Finneran *et al*., 2010). Furthermore, concentrate supplementation may also reduce nitrogen (N) excretion through reducing overall N intake and possibly altering N excretion from urine to faeces (Van Vuuren *et al*., 1987). The Hypothesis of this experiment was that offering supplementation and altering carbohydrate type would improve cow performance and reduce N excretion. Therefore, the objective of this research was to investigate the effect of concentrate supplementation strategy on DMI, SR and N excretion in late lactation dairy cows.

**Material and methods** Thirty six Holsten Friesian dairy cows were blocked on days in milk (+185DIM) and balanced for parity, pre-experimental milk yield and milk composition, predicted 305day milk yield and BCS. Cows were randomly assigned to one of three dietary treatments in a randomised complete block design (n=12). The dietary treatments (T) were: grass only (T1); grass + 2.6kg DM barley based concentrate (T2): grass + 2.6kg DM maize based concentrate (T3). The diets were fed for a 14day acclimatization period and then for a further 63days. Pasture DMI was determined during week five of the study (mid-September - average 220DIM or 31 weeks into lactation) by extracting n-Alkanes from pasture, concentrate and faeces samples according to the method of Dove and Mayes (2006). Data was checked for normality and analysed using repeated measures in PROC MIXED of SAS where treatment, covariate, parity and their interactions were included as fixed effects.

**Results** Cows offered T1 had a lower milk yield (14.54kg) than T2 (17.15kg, $P<0.001$) and T3 (16.73kg, $P<0.001$). Similarly, T1 had lower fat and protein kgs (1.47kgs) than T2 (1.51kgs; $P<0.001$). Cows offered T2 (1.51kgs) had higher fat and protein kgs than T3 (1.48kgs, $P<0.001$). Cows offered T1 had a higher pasture DMI (17.29kg DM/d) than T2 (13.68kg DM/d, $P<0.001$) and T3 (13.34kg DM/d, $P<0.001$). Substitution rates were 1.36kg and 1.49kg for T2 and T3 respectively. The N intakes of cows offered pasture only (0.821kg/day) was significantly higher than both supplemented treatments (T2:0.735kg/day, $P=0.008$; T3:0.737kg/day, $P<0.01$). Supplemented cows had significantly higher N in milk (T2: 15.7%, $P<0.001$; T3: 15.5%, $P<0.001$) than cows offered pasture only (11.9%). Cows offered concentrate supplementation had significantly lower N in urine (T2: 56.6%, $P<0.001$; T3: 57.3%, $P=0.003$) than cows offered pasture only (62.3%). Cows offered T2 proportioned significantly more N to faeces (27.7%) in comparison to cows offered grass only (25.9%, $P=0.04$).

**Conclusion** In conclusion, concentrate supplementation in late lactation increased milk yield and kgs of milk solids but reduced pasture intake. Supplemented cows also excreted less N in the urine and more in milk than unsupplemented cows. Of the two supplementation types, cows offered the barley based supplement had higher milk solids and excreted N in urine.

**Acknowledgements** The authors gratefully acknowledge funding from Enterprise Ireland and Glanbia under the DairyTech project.

**References**

Burke F., O'Donovan M.A., Murphy J.J., O'Mara F.P., Mulligan F.J. 2008. Livestock Science 114, 325–335
Dove, H. and Mayes, R. W. 2006. Nature Protocols 1, 1680-1697
Finneran, E., Crosson, P., O'Kiely, P., Shalloo, L., Forristal, D. and Wallace, M. 2010. Journal of Farm Management 14, 95–116.
Kellaway and Porta. 1993. Dairy Research and Development Corporation Australia
Kolver, E., Muller, L.D., Varga, G.A. and Cassidy, T.J.1998. Journal of Dairy Science 81 (7), 2017-2028.
O'Brien, B., Crosse, S. and Dillon, P. 1996. Irish. Journal of Agricultural and Food Research 35, 113–125.
Stockdale, 2000. Animal Production Science 40 (7), 913-921.
Van Vuuren, A.M., Meijs J.A.C. 1987. Animal Manure on Grassland and Fodder Crops. Fertilizer or Waste? Developments in Plant and Soil Sciences, vol 30. Springer, Dordrecht

# The effect of calcareous marine algae, with or without marine magnesium oxide, and sodium bicarbonate on milk production in mid-lactation dairy cows

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**Application** The inclusion of rumen buffers, in a TMR fed to Holstein dairy cows, improved milk components. Calcareous marine algae increased milk fat and protein yield, offering dairy producers a solution to improve milk production.

**Introduction** High producing dairy cows consuming highly fermentable diets often experience low rumen pH (Plaizier *et al*., 2008), initiated by the accumulation of volatile fatty acids in rumen fluid (Whelan *et al*., 2013). Rumen buffers are commonly added to lactating cow diets to avoid prolonged episodes of low rumen pH and the associated production losses. The addition of sodium bicarbonate (SB) to the diets of high producing dairy cows, as a rumen buffer, has become a regular practice in many parts of the world (Rauch *et al*., 2012). In recent years' calcareous marine algae (*Lithothamnion calcareum*) has been used to stabilise rumen pH and improve fermentation (Cruywagen *et al*., 2015). The objective of this experiment was to evaluate different dietary buffers on the milk production, milk composition, dry matter intake and feed efficiency of mid-lactation dairy cows.

**Material and methods** The supplements included were: calcareous marine algae (*Lithothamnion calcareum*), with or without marine magnesium oxide (precipitated magnesia derived from seawater), and SB. Fifty-two multiparous and four primiparous cows (62.7 ± 3.4 DIM) were assigned to four experimental treatments based on calving BCS (3.1 ± 0.03; scale 1 to 5), pre-experimental milk yield (34.7 ± 0.79 kg/d) and previous 305-day yield (7073 ± 198 kg). Cows were housed in a free stall barn and had *ad-libitum* access to total mixed ration (TMR) and water. The diets were based on a forage: concentrate ratio of 46:54. Dietary treatments consisted of the control (283 g starch and sugar, and 230 g neutral detergent fibre (NDF) from forage per kg dry matter (DM)) including no dietary buffer (CON); the CON plus 3.5 g/kg DM calcareous marine algae (CMA); the CON plus 3.5 g/kg DM calcareous marine algae and 0.9 g/kg DM marine magnesium oxide (CMA+MM); the CON plus 7 g/kg DM SB. The experiment lasted for 80 days (d), which included 7 d acclimatisation to the diet and 73 d of data collection. Milk production data were analysed using the MIXED procedure (SAS, version 9.4). The model included fixed effects of treatment, week and parity as well as treatment by week interaction with cow considered as the random effect. Week was considered as a repeated measure. Pre-experimental milk yield and calving BCS were considered as covariates.

**Results** The DMI of cows consuming SB tended to be higher than cows on the CON diet (+ 1.9 kg, $P<0.10$). CMA increased the production of milk solids (fat and protein kg/d) compared to CON (+ 0.16 kg, $P<0.01$), CMA+MM (+ 0.09 kg, $P<0.05$) and SB (+ 0.10 kg, $P<0.05$). Both CMA (+ 0.09 kg, $P<0.01$) and CMA+MM (+ 0.06 kg, $P<0.01$) increased milk fat yield compared to CON but were not different to each other and SB. Cows supplemented with CMA (+ 0.19 %, $P<0.01$), CMA+MM (+ 0.25 %, $P<0.01$) and SB (+ 0.19 %, $P<0.01$) increased milk fat concentration compared to CON but were not different from each other. The CMA treatment increased milk protein yield compared to CON (+ 0.04 kg, $P<0.01$), CMA+MM (+ 0.06 kg, $P<0.01$) and SB (+ 0.04 kg, $P<0.01$). The SB treatment reduced the efficiency of milk production, energy-corrected milk (ECM) per kg of DMI, compared to CON (- 0.11 kg, $P<0.01$), CMA (- 0.12 kg, $P<0.01$) and CMA+MM (- 0.13 kg, $P<0.01$).

**Conclusion** Results indicate that the addition of rumen buffering products can increase milk fat concentration when included in lactating dairy cow diets. The use of CMA when compared to sodium bicarbonate, in such diets, can increase milk production efficiency and combined fat and protein yield (kg/d). This offers dairy producers, with milk pricing based on milk solids, the opportunity to increase the value of their milk.

**Acknowledgements** The authors gratefully acknowledge funding from Celtic Sea Minerals, Cork, Ireland.

**References**

Cruywagen, C. W., S. Taylor, M. M. Beya, and T. Calitz. 2015. The effect of buffering dairy cow diets with limestone, calcareous marine algae, or sodium bicarbonate on ruminal pH profiles, production responses, and rumen fermentation. Journal of Dairy Science 98, 5506-5514.

Plaizier, J. C., D. O. Krause, G. N. Gozho, and B. W. McBride. 2008. Subacute ruminal acidosis in dairy cows: The physiological causes, incidence and consequences. Veterinary Journal 176, 21-31.

Rauch, R. E., P. H. Robinson, and L. J. Erasmus. 2012. Animal Feed Science and Technology. 177, 180-193.

Whelan, S. J., F. J. Mulligan, J. J. Callan, B. Flynn, and K. M. Pierce. 2013. Effect of forage source and a supplementary methionine hydroxyl analogue on rumen fermentation parameters in lactating dairy cows offered a low crude protein diet. Animal Feed Science and Technology 183, 62-66.

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# Rumen "inertness" of a novel whey protein gel of fish oil feed supplement for dairy cows

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**Application** Lipid composite gels may be an effective way of preventing rumen exposure to unsaturated oils, and so could be used to improve milk fatty acid profile, and also enhance transfer of beneficial fatty acids to the cow.

**Introduction** There is renewed interest in dairy cow supplements containing very long chain n-3 polyunsaturated fatty acids (PUFA), such as those rich in eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), to improve cow health and fertility. Feeding such oil sources can have a negative impact on rumen digestion and subsequent milk quality and production. In addition, highly unsaturated oils undergo extensive biohydrogenation in the rumen. Therefore, rumen inertness is key to optimising absorption of the beneficial fatty acids (FA) by the cow, and preventing adverse effects. Rumen inertness technologies vary widely in their effectiveness (especially for oils high in PUFA), with most resulting in inconsistent effects *in vivo*, but lipid composite gels may offer a practical solution (Gadeyne *et al*., 2016) The main objectives of this study were (i) to successfully make a whey protein gel of fish oil (WPG-FO), and (ii) to test the rumen inertness when fed to lactating dairy cows.

**Material and methods** The WPG-FO was manufactured using the method of Carroll *et al*., (2006) and Kliem *et al*. (2017). Briefly, whey protein isolate (UltraWhey 90; Volac International Ltd., Royston, UK) was hydrated then mixed with salmon oil (Inovitec, Tarporley, UK; EPA and DHA content of 6 and 7 g/100 g total fatty acids, respectively), before being homogenised, transferred to food cans and then heated to 120°C for 138 min. Four late-lactation cows (mean 286 d in milk ± 11.8 s.e.m.) were randomly allocated to one of two treatment groups, where a standard lactating cow diet was hand-mixed with either WPG-FO or unprotected fish oil (FO). The trial ran for 5 days and each supplement was included incrementally over days 1 and 2, so that from day 3-5 each cow was consuming 200 g fish oil equivalent per day. Cows on the FO diet were also fed whey protein isolate. Cows were fed and milked twice daily. Milk samples were taken at each milking on days 0 and 5, and analysed for milk FA profile and composition. Milk fat *trans*-11 18:1 and 18:0 concentrations were used as indicators of rumen protection, as EPA and DHA inhibit the final step of biohydrogenation (*trans*-11 18:1 to 18:0). Results were analysed using the Mixed procedure in SAS with a model which included effects of treatment and day.

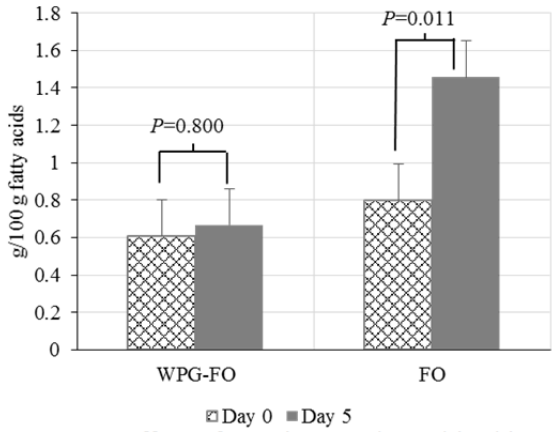


**Figure 1** Effect of supplementation with either whey protein gel of fish oil (WPG-FO) or fish oil (FO) on milk fat *trans*-11 18:1 concentration

**Acknowledgements** This study was supported by the University of Reading. The authors gratefully acknowledge staff at the Centre for Dairy Research for care of the animals, and Volac International Ltd., for donating the whey protein isolate.

**References**

Carroll, S. M., DePeters, E. J. and Rosenberg, M. 2006. Journal of Dairy Science 89, 640-650.
Gadeyne, F., De Neve, N., Vlaeminck, B. and Fievez, V. 2016. European Journal of Lipid Science and Technology 119, 1600345
Kliem, K. E., Humphries, D. J., Grandison, A. G., Morgan, R., Givens, D. I., Reynolds, C. K. 2017. Advances in Animal Biosciences 8, 71

# Supplementation of dairy cow diets with docosahexaenoic acid enriched microalgae and its effect on milk fatty acid profile over time

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**Application** Microalgae can be fed to cows to improve the healthiness of milk by increasing the milk content of docosahexaenoic acid (DHA), but rumen adaptation may occur with extended feeding time.

**Introduction** The benefits of long chain fatty acids (FA) on human health have long been recognised, particularly the very long chain *n*-3 polyunsaturated fatty acids (LC *n*-3 PUFA) such as DHA - C22:6n-3, (Marventano *et al*., 2015). The primary producer of LC *n*-3 PUFA at the bottom of the food chain is algae (ALG). The addition of dried ALG to the diet of dairy cows has previously been shown to increase the content of LC *n*-3 PUFA in milk (Till *et al*., 2016). However, ruminal adaption to high levels of dietary PUFA can occur, altering the formation of specific biohydrogenation intermediates and potentially decreasing the amount of DHA available for secretion in milk (Shingfield *et al*., 2006), although little information is available for ALG. The objective of the study was to determine the effect of feeding DHA enriched ALG over time on the milk FA profile of Holstein-Friesian dairy cows.

**Material and methods** Sixty Holstein-Friesian dairy cows were randomly allocated to one of two diets at 3 weeks post calving based on parity, calving date, milk yield at 14-21 days in milk, and live weight measured 1 week prior to the start of the study (week 0). The basal ration contained maize and grass silages and straight feeds, and was supplemented with one of two levels of ALG (*Schizochytrium limacinum;* Alltech UK Ltd.,) to provide two treatments; 0 (Control) or 100 g algae/cow/d (Algae). The ALG contained 13.5 g/kg DM crude protein, 58 g/kg oil, 0.28 g/100 g FA as C20:5*n*-3 and 25.7 g/100g FA as C22:6*n*-3. The diets were fed for a total of 14 weeks. Cows were milked twice daily and samples taken at consecutive am and pm milkings at wks 0, 1, 2, 4, 8 and 14 for subsequent analysis of fatty acids, with milk fat content measured weekly. Individual intake was recorded daily. Data were analysed as repeated measurements analysis of variance using Genstat (version 16.1), using data recorded in week 0 as a covariate where appropriate.

**Results** Supplementation with ALG had no effect ($P > 0.05$; Table 1) on DMI or milk yield, which averaged 22.1 and 40.4 kg/d respectively, but there was an effect of time ($P < 0.001$), with both peaking at week 4 and 5 then declining. Similarly, milk fat content and yield were not affected by diet ($P > 0.05$), but there was an effect of time ($P<0.05$). Feeding ALG decreased ($P<0.05$) milk fat content of C18:0 and increased milk fat content of C18:1 *trans*-11, C18:2 *cis*-9 *trans*-11 conjugated linoleic acid (CLA). Milk fat content of DHA increased ($P < 0.001$; Figure 1) from week 2 onwards, peaking at week 14 with an increase of 0.35 g/100 g in ALG fed cows compared to those fed control diet at wk 14.

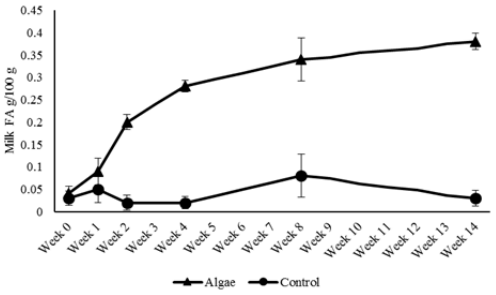


**Figure 1** Change in milk fat DHA content g/100 g over 14 weeks in cows fed algae or control

**Table 1** Intake, milk yield, milk fat content and selected fatty acids in cows fed algae or control

| | Treatment | | | P value[1] | | |
|---|---|---|---|---|---|---|
| | Algae | Control | s.e.d | D | T | D x T |
| DM intake, kg/d | 22.0 | 22.1 | 0.861 | 0.905 | <.001 | 0.791 |
| Milk yield, kg/d | 40.4 | 40.4 | 1.023 | 0.980 | <.001 | 0.729 |
| Milk fat, g/kg | 36.9 | 37.5 | 2.09 | 0.702 | 0.048 | 0.912 |
| Milk fat yield, kg/d | 1.46 | 1.52 | 0.2 | 0.401 | 0.013 | 0.738 |
| Milk fatty acids, g/100g | | | | | | |
| C16:0 | 30.3 | 31.1 | 0.828 | 0.507 | 0.124 | 0.250 |
| C18:0 | 7.89 | 8.41 | 0.439 | 0.019 | 0.039 | 0.345 |
| C18:1 trans 11 | 1.21 | 0.831 | 0.163 | 0.002 | 0.109 | 0.356 |
| C18:2 cis-9 trans-11 CLA | 0.796 | 0.532 | 0.069 | 0.038 | 0.003 | 0.052 |
| C20:5n-3 (EPA) | 0.129 | 0.077 | 0.019 | 0.376 | <.001 | 0.242 |

[1]Probability of significant effects attributable to the diet (D), time (T), and their interaction (D x T)

**Conclusion** Algae can be added to the diet of dairy cows from 3 weeks post calving at a rate of 100 g/cow/d to improve milk content of DHA, with no negative impact on cow performance or rumen adaptation over time.

**Acknowledgements** The authors gratefully acknowledge Alltech for funding this study.

**References**

Marventano, S, Kolacz, P, Castellano S, Galvano F, Buscemi S, Mistretta A and Grosso G, 2015. Int J Food Sci Nutr, 1–12.
Shingfield, KJ, Reynolds, CK, Hervas, G, Griinari, JM, Grandison, AS and Beever, DE, 2006. J. Dairy Sci, 89, 714-732.
Till, B.E., Huntington, J.A., Taylor-Pickard, J. and Sinclair, L.A. 2017. Advances in Animal Biosciences 8 (1), 79.

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# Comparison of the effects of pasture and total mixed ration diets on dairy cow milk and milk solids production

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**Application** Incorporating white clover into grass swards permits greater milk production compared to grass only swards, and similar herbage production at a lower N input. A TMR diet results in greater milk production compared to a pasture based diet.

**Introduction** Feed quality has a major influence on dairy cow milk production. Total mixed ration (TMR) systems are usually considered to supply a more consistent diet quality compared to pasture based systems (Kolver *et al*., 1998). Irish milk production systems are predominantly spring calving and grass based. The inclusion of white clover (*Trifolium repens* L.; clover) into perennial ryegrass (*Lolium perenne* L.) swards can potentially increase dairy cow milk production (Phillips & James, 1998; Egan *et al*., 2017). The objective of this experiment was to compare milk and milk solids production and composition from three feeding systems: total mixed rations (TMR), grass only sward receiving 250 kg N/ha (GO) and grass and clover sward receiving 150 kg N/ha (GC). The hypothesis is that milk production will be greatest on TMR and least on GO.

**Material and methods** An experiment was undertaken at Teagasc, Animal and Grassland Research and Innovation Centre, Moorepark, Fermoy, Co. Cork, Ireland from February to October 2017. Fifty-one dairy spring calving cows from the Moorepark dairy herd were blocked according to breed (15 Friesians and 2 Jersey × Friesians per treatment), calving date (mean = 17 February +/-3days), parity (2.53; 5 primiparous and 12 multiparous) and pre-experimental milk and milk solids yield and randomly allocated to one of the three treatments (n=17) The three treatments were: TMR (grass silage (4.5 kg DM/cow), maize silage (9 kg DM/cow) and concentrates (8.5 kg DM/cow)), GO and GC. The TMR treatment were housed indoors and fed using electronic controlled Roughage Intake Control system feed bins (Hokofarm Group B.V., Voorsterweg 28, 8316PT Marknesse, Netherlands. TMR cows were fed at 08:30 am daily using a Keenan diet feeder wagon. Grazing or TMR feeding commenced in February, and cows were added to their respective treatment groups approximately 2 weeks post calving. The GO and GC treatments were stocked at 2.75 cows ha in a closed farm system. Swards were rotationally grazed to achieve a target post-grazing sward height of 4 cm. Measurements included pre-grazing herbage mass, sward clover content, milk yield and milk solids yield (sum of fat and protein yield). Data were analysed using PROC Mixed in SAS.

**Results** Pre-grazing herbage mass was similar on GO and GC treatments (1653 kg DM/ha). Average sward clover content was 18.8%, ranging from 4.6% in February to a peak of 44.2% in September. Average daily milk yield and milk solids yield, and cumulative milk solids yield were significantly greater on TMR compared to GO and GC. The GC treatment had greater milk and milk solids yield compared to GO but this was not significant.

**Table 1** Average daily milk and milk solids yield and cumulative milk solids yield for cows on grass-only swards receiving 250 kg N/ha (GO), and grass-clover swards receiving 150 kg N/ha (GC) and total mixed ration (TMR) treatments.

| | GO | GC | TMR | S.E. | P Value |
|---|---|---|---|---|---|
| Milk yield (kg/cow/day) | 22.0[a] | 23.0[a] | 29.6[b] | 1.04 | <0.001 |
| Milk solids yield (kg/cow/day) | 1.77[a] | 1.88[a] | 2.31[b] | 0.078 | <0.01 |
| Cumulative milk solids yield (kg MS/cow) | 448[a] | 476[a] | 580[b] | 22.9 | <0.01 |

**Conclusion** Milk production was greater on the TMR treatment compared to GO and GC. Including clover in a perennial ryegrass sward resulted in similar herbage production to grass only, despite reducing N fertiliser input by 100 kg N/ha.

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**References**

Egan, M., Lynch, B.M., Hennessy, D. 2017 Journal of Agricultural Science 155, 657-668.
Kolver, E.S. and Muller, L.D. 1998. Journal of Dairy Science 81, 1403-1411.
Phillips, C. J. C., and James, N.L. 1998. Animal Science 67, 195-202.

# The effect of post-harvest treatment of field beans on dairy cow performance

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**Application** Post-harvest treatment of field beans (either dry coarsely milled, dry finely milled or moist propionic acid treatment) had no effect on dairy cow performance.

**Introduction** As a result of price volatility and instability in supply, the UK livestock sector is seeking to reduce its reliance on imported protein feedstuffs. Consequently, there is increasing interest in the use of locally-grown forage and grain legume crops. Field beans (*Vicia Faba*) are a grain legume of particular interest as they can be grown successfully in many areas across the UK, with recent AFBI research demonstrating that beans can be included in dairy cow diets at up to 5.0 kg/cow/day with no loss in performance (Johnston *et al*., 2016). However, in the more northerly parts of the UK beans are frequently harvested with a moisture content in excess of 16%, and as such must be treated to prevent mould growth. While drying is the practice most commonly used, there is little information available on the impact of the degree of milling of dried beans on subsequent performance, and important issue given the high starch content of beans. In addition, acid treatment of moist beans has been conducted on some farms, although the impact of this on subsequent performance, is unknown. This study was designed to examine the impact of moist preservation of field beans using propionic acid, and the extent of physical treatment of dried field beans, on dairy cow performance and nutrient utilisation.

**Material and methods** Eighteen mid-lactation Holstein-Friesian dairy cows were used in a three period (each of four weeks duration) change over design experiment. The field bean crop used in the experiment (*Var. Boxer*) was harvested at a moisture content of approximately 25%. Three treatments, each comprising a different post-harvest treatment of field beans, were examined. Following harvest, approximately 2/3 of the bean crop was dried at 80$^{o}$C for four hours to achieve a moisture content of 16%, before being left to cool. Beans were then either coarsely rolled (CR) or finely milled (FM). The remaining 1/3 of the bean crop was coarsely rolled and the beans then treated with propionic acid at a rate of 20 litres/ton fresh beans (ACID). Cows on all three treatments were offered a mixed ration comprising grass silage and concentrates (forage: concentrate ratio of 60: 40 on a dry matter basis). The concentrate component of the diet with treatments CR and FM comprised a common 'pre-mix' (600 g/1000 g concentrate) with the remaining 400 g per 1000 g concentrate comprising field beans (CR or FM treated beans, respectively). With treatment ACID the same concentrate pre-mix was used (600 g/1117 g concentrate), while the moist field beans were incorporated at 517 g/1117 g concentrate, the higher inclusion reflecting their lower DM content. The experimental concentrates were designed to achieve an intake of field beans of approximately 3.5 kg per day with each treatment. Data recorded during the final week of each period was analysed using ANOVA, with the analysis taking account of the change-over design nature of the study.

**Results** Neither degree of processing of dried field bean (either CR or FM), or treatment of moist rolled field bean with propionic acid (ACID), had any effect on any of the cow performance parameters measured.

**Table 1** Effects of post harvest treatment of field beans on cow performance

| | CR | FM | ACID | S.E.M | P-Value |
|---|---|---|---|---|---|
| Silage DMI (kg/day) | 13.0 | 13.2 | 13.4 | 0.34 | 0.786 |
| Total DMI (kg/day) | 22.6 | 22.9 | 22.3 | 0.55 | 0.759 |
| Milk yield (kg/day) | 33.5 | 33.3 | 32.0 | 1.35 | 0.690 |
| Milk fat (g/kg) | 41.9 | 41.5 | 42.2 | 1.27 | 0.932 |
| Milk protein (g/kg) | 33.7 | 33.9 | 33.9 | 0.63 | 0.968 |
| Milk fat + protein yield (kg/day) | 2.50 | 2.47 | 2.33 | 0.111 | 0.511 |
| Final body condition score | 2.58 | 2.58 | 2.55 | 0.065 | 0.917 |
| Final live weight (kg) | 658 | 658 | 653 | 18.5 | 0.977 |

**Conclusion** Milk yields are reflective of stage of lactation and intakes achieved. Cows offered propionic acid treated field beans had similar performance to those offered dried field beans. Degree of processing of dry field beans (coarsely rolled or finely milled) had no effect on animal performance. The results of this study highlight that a number preservation options exist for moist field beans.

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**Reference**

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# Dry matter intake of pasture fed dairy cows – the effect of pasture allowance and duration on dry matter intake and production throughout early lactation

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**Application** Short term reductions in pasture allowance (PA) in early lactation influence immediate dry matter intake (DMI) and milk production; however, there was little effect on bodyweight (BW) and body condition score (BCS) which demonstrates the ability of the cows to adapt to short term reductions in PA.

**Introduction** Grass growth can be highly variable in spring with many farmers experiencing short term deficits during the first grazing rotation. Ganche *et al*. (2013) demonstrated that post grazing sward height can be used as a short term strategy to manage pasture deficits. The experimental aim was to determine the effect of PA (60% vs. 100% of intake capacity (IC)) offered for 2 or 6 weeks on immediate and carryover DMI, milk production, BW and BCS of dairy cows in early lactation.

**Material and methods** On March 14, 2016, 105 dairy cows (30 primiparous) were randomly assigned to one of seven grazing treatments for 16 weeks. Cows were randomised on parity, breed, calving date, pre-experimental production. The control (CTL) treatment were allocated 100% of their IC which is based on parity, days in milk, potential milk production and BW (Faverdin *et al*. (2011). The remaining grazing treatments received 60% of the CTL treatments PA for either two (x2) or six (x6) weeks from week one (E), week three (M) or week five (L) of the experiment. Milk yield was recorded daily, while milk composition, BW and BCS were measured on a weekly basis. Fresh grass was allocated after each milking and PA was offered above 3.5cm. Post grazing sward height was measured daily. All cows grazed together from week eleven when the final 60% treatment had ceased. Dry matter intake was estimated using the n-alkane technique (Mayes *et al*., 1986). The estimations were carried out during week two for the E60%x2 and CTL, in week four for the M60%x2 and CTL, in week six for the L60%x2, E60%x6 and CTL, in week eight for the M60%x6 and CTL and in week 10 for the L60%x6 and the CTL treatment. Each intake estimation period coincided with each treatments respective final week of 60% PA. During week 16 DMI was also estimated to determine any carryover effect of early lactation treatments. Variables associated with DMI were analysed using PROC MIXED in SAS version 9.4 (SAS Institute Inc., Cary, NC, USA). The model contained terms for treatment, breed, parity, days in milk and pre-experimental milk yield and BCS.

**Results** The E60%x2, M60%x2 and L60%x6 had significantly lower DMI than CTL during their respective measurement periods (see Figure 1). All other treatments had similar DMI to CTL during their respective intake estimation period. Dry matter intakes achieved by the restricted cows were at least 80% of CTL demonstrating that cows are able to adapt and graze below their allocation to increase DMI when PA was reduced. All treatment groups had a similar DMI in week 16 (15.4 kg DMI/cow/day). All treatment groups had significantly lower milk (see Figure 2) and milk solids yield in comparison to CTL at the time of their respective DMI estimation but there was no carryover effect of treatment on milk production variables (17.5 kg milk/cow/day, 1.41 kg MS/cow/day). The M60%x2 and M60%x6 had significantly lower BW than CTL group (-15kg, -27kg respectively) during restriction but this may be due to differences in gut fill. All treatment groups had a similar BW in week 16 (468 kg). There was no effect of treatment on BCS during the 60% restriction period (2.86) or in the carryover period (3.02).

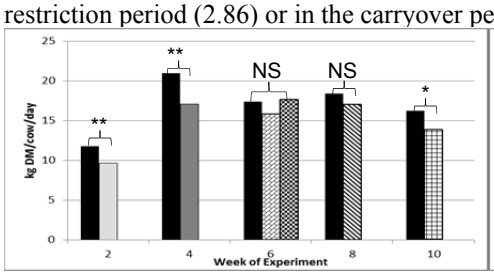


**Figure 1** Dry matter intake estimations

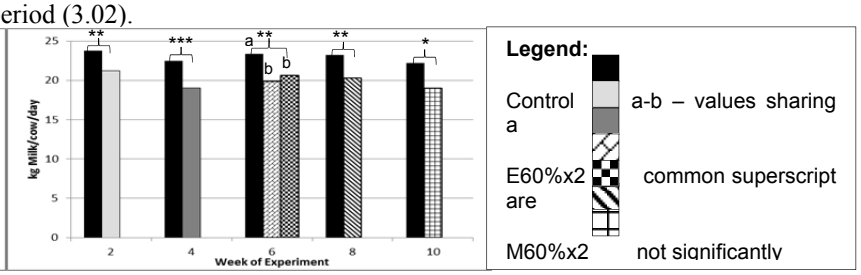


**Figure 2** Daily milk yield at respective intake estimation period

**Conclusion** While allocating 60% IC can reduce DMI and milk production in the short term, once cows are allocated 100% of their IC DMI and milk production returns to the same level as the CTL treatment. This suggests that PA can be used as a tool to overcome short term grass deficits during the first grazing rotation. However, further investigation is required to examine the effects on dairy cows' health, fertility and welfare.

**References**

Faverdin, P., Baratte, C., Delagarde, R. and Peyraud, J. 2011. Grass and Forage Science 66**,** 29-44.
Ganche, E., Delaby, L., O'Donovan, M., Boland, T. and Kennedy, E. 2013. Animal 7, 1390.
Mayes, R., Lamb, C. and Colgrove, P. M. 1986. The Journal Of Agricultural Science 107, 161-170.

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# Grass silage particle length and grass to maize silage ratio effects on production and reticulo-rumen pH in dairy cows

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**Application** Feeding dairy cows a short compared to a long chop length grass silage will increase milk yield and tend to reduce milk fat content, but there is no effect on reticulo-rumen pH. Feeding dairy cows mixtures of maize and grass silages will increase intake, milk production and milk protein content, and result in a lower rumen pH compared to grass silage as the only forage.

**Introduction** Mixed rations (MR) fed to UK dairy herds have a higher proportion of long (>19 mm) particles, less shorter particles (<4 mm) and a considerably higher mean particle size (PS) compared to those reported for North American diets (Tayyab *et al*., 2017). These differences are mainly due to the long PS of the grass silage (GS) fed in the UK (Tayyab *et al*., 2017). Most studies that have investigated the effect of forage particle length on rumen function and dairy cow performance have however, fed North American type diets based on lucerne and maize silage, and are therefore less applicable to UK feeding systems. This study was conducted to evaluate the effect of the PS of GS when fed with two ratios of maize silage (MS) on animal performance and reticulo-rumen pH in high yielding dairy cows in the UK.

**Material and methods** Sixteen early lactation (60 ± 10.6 days in milk) multiparous Holstein-Friesian cows (balanced for previous lactation and parity) were group housed and individually fed in a 4×4 Latin square design with four periods each of 28-d duration, with measurements undertaken during the final week of each period. Perennial ryegrass was harvested at 2 theoretical chop lengths (10 and 44 mm; short and long chop length, respectively), and included at two ratios with maize silage (GS:MS) of 100:0 or 40:60 (DM basis). The silages were supplemented with concentrates to produce four isonitrogenous and isoenergetic diets with a forage to concentrate ratio of 0.54:0.46 (DM basis). A rumen protected soybean meal was used to balance the undegradeable protein content of diets. The four diets were: long chop GS (100:0 **LG**); short chop GS (100:0, **SG**); 40:60 long chop GS:MS (**LM**) and 40:60 short chop GS:MS (**SM**). During the final week of each period milk yield was recorded at each milking and four milk samples (2 am and 2 pm milkings) collected for subsequent analysis of fat, protein and lactose using a Milkoscan Minor analyser (Foss, Denmark). Feed intake was recorded using Hokofarm roughage intake feeders (Marknesse, Netherlands). A pH bolus (e-Cow®, eCow Devon Ltd, Exeter Devon, UK) was administered orally to each cow two weeks prior to the collection week during periods 1 and 3, and data recorded every 15 min (Mottram *et al*., 2008). Data were analysed as a 2×2 factorial design with main effects of chop length (C), forage ratio (F) and their interaction (C × F) using GenStat 18.1.

**Results** Dry matter (DM) intake was 0.9 kg/d higher in cows when fed the short compared to the long chop grass silage, and 3.2 kg/d higher (P<0.001) in cows when offered the GS:MS diets compared to GS alone (Table 1). An interaction (P=0.011) was observed between C and F on milk yield, with cows having a lower milk yield when offered long chop GS alone, but not when fed the LM diet. Milk fat content tended (P<0.1) to be higher in cows when fed the long chop GS, whereas milk protein content was higher in cows when fed GS:MS diet. The minimum reticulo-rumen pH and mean pH were lower (P<0.001) in cows when fed GS:MS than GS alone, but there was no effect (P>0.05) of GS chop length.

**Table 1** Intake, milk production, milk composition and reticulo-rumen pH in high producing dairy cows

| | Treatments | | | | | P-value | | |
|---|---|---|---|---|---|---|---|---|
| | LG | SG | LM | SM | SED | C | F | C × F |
| DM intake (kg/day) | 20.0 | 20.5 | 22.8 | 24.0 | 0.56 | 0.035 | <0.001 | 0.335 |
| Milk yield (kg/day) | 37.3 | 39.1 | 41.1 | 40.5 | 0.63 | 0.179 | <0.001 | 0.011 |
| Milk fat (g/kg) | 40.1 | 38.5 | 39.5 | 38.6 | 0.93 | 0.090 | 0.560 | 0.418 |
| Milk protein (g/kg) | 30.9 | 30.7 | 32.3 | 32.4 | 0.28 | 0.738 | <0.001 | 0.461 |
| Daily minimum pH | 5.99 | 5.98 | 5.90 | 5.87 | 0.039 | 0.421 | 0.001 | 0.594 |
| Mean pH | 6.42 | 6.41 | 6.33 | 6.34 | 0.035 | 0.998 | 0.001 | 0.775 |

**Conclusion** Grass silage chop length had no effect on reticulo-rumen pH but a longer particle size reduced DM intake and milk yield, but had little effect on milk fat yield. Partial replacement of grass with maize silage reduced reticulo-rumen pH but increased DM intake and milk yield irrespective of the chop length of the grass silage.

**Acknowledgements** This study was funded by AHDB Dairy.

**References**

Mottram, T., Lowe, J., McGowan, M. and Phillips, N. 2008. Computers and Electronics in Agriculture 64, 45-48.
Tayyab, U., Wilkinson, R.B., Reynolds, C.K. and Sinclair, L.A. 2017. Advances in Animal Biosciences 8, 73.

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# Investigation of the use of faecal fibre (NDF) as an alternative to diet samples for determination of N isotopic fractionation in dairy cattle

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**Application** Faeces cannot be used as an alternative to feed samples when estimating N isotopic fractionation in dairy cows.

**Introduction** Previous work has demonstrated the use of N isotopic fractionation as a simple proxy for Feed Conversion Efficiency (FCE). The increase in $^{15}N$ between diet and plasma protein (termed $\Delta^{15}N$) was negatively related to FCE (e.g. Wheadon *et al*., 2014). This provides a useful, low-cost, approach for estimation of FCE for genetic or other evaluations, but depends on the availability of feed samples. There are many situations, including when animals are grazing heterogeneous swards or when samples are obtained at the abattoir, where feed samples are not available and so this work explored the use of faecal samples as an alternative.

**Material and methods** Samples of faeces were obtained from five cows offered each of six different diets based on a range of forages: four total mixed rations (TMR) offered to lactating dairy cows and two TMR offered to dry cows. The main ingredients of the milking TMR were: (1) wheat straw, wet distillery by-product and a concentrate blend; (2) grass silage, maize silage, field beans and wheat; (3) grass silage, maize silage and a concentrate blend; and (4) grass silage, maize silage and a concentrate blend. The main ingredients of the dry cow TMR were: (1) wheat straw and one-third (DM basis) of a lactation diet based on wet distillery by-product and a concentrate blend; and (2) wheat straw and one-third (DM basis) of a lactation diet based on grass silage, maize silage and a concentrate blend. Samples of the diets and faeces were dried (60$^{o}$C), milled and analysed for NDF content. Duplicates of the TMR and isolated NDF were analysed for $^{15}N$ content by isotope-ratio mass spectrometry (Iso-Analytical Ltd., Crewe). Nitrogen-15 results are expressed in delta units (‰) relative to the international standard (air).

**Results** The NDF concentrations in the lactation TMRs were 344, 329, 352 and 378 g/kg DM respectively, whilst the dry period TMRs had 464 and 620 g NDF/kg DM. Corresponding faecal NDF concentrations (with s.d. in parentheses) were 553 (39.6), 546 (21.4), 480 (15.9), 492 (27.9), 649 (11.1) and 632 (21.7) g/kg DM. There was no significant relationship between the $^{15}N$ content of diets and NDF prepared from faeces produced by cows consuming those diets ($r^2$=0.09; n=30; Not significant; Figure 1). The discrepancy between feed and faecal NDF $^{15}N$ was most pronounced for diets that contained a higher proportion of grass and maize silages.

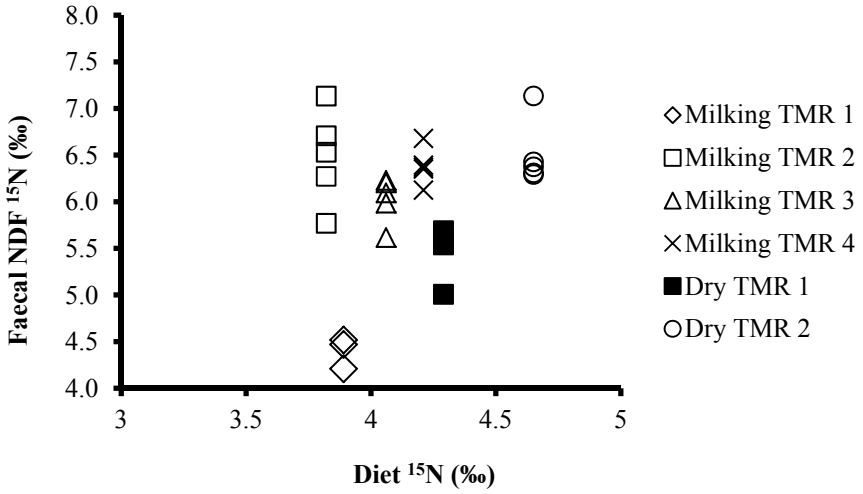


**Figure 1** Relationship between the $^{15}N$ content of faecal NDF and diets for six dairy cattle diets

**Conclusion** There was no significant relationship between the $^{15}N$ content of feed samples and NDF prepared from faeces of cattle consuming the same diets.

**Acknowledgements** This work is supported by the Scottish Government Rural Affairs, Food and Environment Strategic Research Programme

**Reference**

Wheadon, N.M., McGee, M, Edwards, G.R. and Dewhurst, R.J. 2014. British Journal of Nutrition 111, 1705-1711.

# Is there a relationship between the age at which training the National Hunt Thoroughbred Racehorse commences and chiropractic assessment of skeletal symmetry?

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**Application** Commencing training of National Hunt (NH) racehorses at 3yrs (rather than 4) had, on average, less misalignments of the spine and pelvis. Training at an earlier age may be beneficial for skeletal symmetry.

**Introduction** Injury in the thoroughbred (TB) racehorse is the greatest source of lost training days and time off from the racetrack. Previous research has highlighted that Repetitive Strain Injury (RSI) is the most common cause of musculoskeletal failure in the horse (Reed *et al* 2013). The age at which training commences and the appropriateness of this training are key determinants in affecting RSI. Joint dysfunction and asymmetry are strong indicators for uneven loading of bones and joints and consequent RSI. Chiropractic method's aim to improve joint function (restoring optimal nerve and muscle function) and the symmetry balance of the musculoskeletal system through application of a combination of stimulating, mobilising and manipulative techniques. Limited research has shown that symmetrical horses are less likely to develop RSI. The aim of this study is to investigate if the age at which training commences, is related to the number of misalignments (MA's) observed in the racehorse training to run in Point to Points (P2P).

**Material and methods** 16 sound TB racehorses from a single racing stable in a similar training routine were assessed for MA's of the neck, spine and pelvis by a qualified McTimoney Animal Practitioner. All chiropractic assessments were carried out on the same day and in the same location. Appropriate written informed consents were obtained prior to the study. A pilot study confirmed the reliability of the method at the venue. Two groups of horses were assessed, 8 four year olds (4yo) who began training at 3 and 8 five year olds (5yo) who began training at 4. Each group contained 6 geldings and 2 females. Each randomly selected animal was manually assessed on a level concrete floor and MA's at specific anatomical locations noted. The mean numbers of MA's were computed by group according to age, anatomical location and sex. Data was analysed using JMP Statistical Discovery Software with Standard Deviation and Standard Error of Mean calculated using a 95% confidence interval. Students t-test's assuming unequal variances were used to compare data sets. Also calculated were O'Brien, Levene and 2-sided F-tests.

**Results** Age had a significant effect ($p<0.05$) on the number of MA's observed in the cervical spine and pelvis.

**Table 1** Percentage of MA from possible MA by Anatomical Area and Age

| Anatomical Area | % MA from possible MA | | Mean | SD | Range | Mean | SD | Range |
|---|---|---|---|---|---|---|---|---|
| | 4yo | 5yo | 4yo | | | 5yo | | |
| Cervical | 32.14 | 39.29 | 2.25 | 0.71 | 1–3 | 2.75 | 1.58 | 0–5 |
| Thoracic | 35.00 | 38.33 | 5.25 | 2.87 | 0–8 | 5.75 | 3.11 | 1–10 |
| Lumbar | 14.58 | 31.25 | 0.88 | 0.99 | 0–2 | 1.88 | 1.89 | 0–5 |
| Pelvis | 12.50 | 25.00 | 0.38 | 0.52 | 0–1 | 0.75 | 1.04 | 0–2 |

The mean number of MA's observed in both groups were compared to the total number of MA's possible in each animal and expressed as a percentage. There were statistically significant differences ($p=0.049$) between the number of MA's in the cervical spine of the 4yo ($n=2.25$) group and the 5yo's ($n=2.75$) when tested using a 2-sided F-Test.
There was a statistical significance ($p=0.0004$) observed in the number of MA's detected in the Pelvis area of the different groups when tested using Levene's test. A mean of 0.38 was present in the 4yo's compared to 0.75 in the 5yo's.

**Conclusion** This study provides positive support for the commencement of training of NH racehorses at 3, rather than 4 years of age. The appropriate training of horses to run in 4yo P2P's has been shown to be beneficial in terms of MA observed, particularly in the pelvis and neck. Further study based on a larger cohort of participants is warranted and recommended.

**Acknowledgements** There was no external funding for this project

**References**

Reed, S., Jackson, B., Wood, J., Price, J. and Verheyen, K. 2013. The Veterinary Journal 196, 339 – 344.

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# Risk factors for horse falls in the cross-country phase of British Eventing competitions: A comprehensive data analysis

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**Application** The number of competition starts a horse has may influence its ability to jump fences, potentially increasing the risk of error and a horse fall.

**Introduction** Equestrian eventing is one of the most dangerous Olympic sports, otherwise known as the 'Equestrian Triathlon'. Eventing consists of three phases: dressage, show-jumping and cross-country, which are completed by a single horse and rider combination. The dressage is a series of predetermined movements that need to be completed accurately by the horse and rider. The show-jumping comprises of a course of coloured jump poles that if touched by the horse, will fall and incur penalties. The cross-country is a sequence of solid jumps and obstacles including water and ditches that are jumped at speed by the horse and rider over undulating grassland. The biggest risk for both horse and rider is a horse fall in the cross-country phase of eventing in which the horse somersaults over the jump, potentially crushing the rider.
The aim of the study was to investigate risk factors associated with horse falls on the cross-country phase of British Eventing competitions.

**Materials and methods** Data collected by British Eventing from all affiliated competitions between dates 2005 and 2015 were analysed using Generalised Linear Mixed Effects Model (GLMM) with a logit-link binomial error structure in R. This process enabled the analysis of factors recorded in relation to the occurrence of a horse fall to be determined.
There was an average of 85,000 sets of data per year, which equated to a sample size of 850,000. Each set of data has information about the performance of one horse and one rider at one event. Consequently, the dataset contains both independent and related data, as a rider may compete more than one horse at the same event, they may ride at a number of events in the year and may have competed every year since 2005.
The factors recorded included event, date, class, section, prize money, dressage penalties, show-jumping penalties (jump and time), cross-country penalties (jump and time) - this included if there is an elimination code, e.g. HF, U2, R3, total penalties, horse name, horse gender, horse age, horse height, rider name, rider gender, rider age, rider points (including foundation points, horse points (including foundation points), number of starts in the last 30, 60 and 90 days for both horse and rider, and number of horse falls and unseated rider falls experienced in career, 6 months and 1 year, for both horse and rider.

**Results** Variables retained in the final model include: number of horse falls the horse has had in its career, number of horse starts in 30-60 days, rider gender, number of horse starts in 60-90 days, number of horse starts in 0-30 days, horse grade, rider age, dressage penalties, horse age, show-jumping penalties, days since last start for the rider, number of rider starts 30-60 days and horse gender.
All variables consisting of how many starts the horse has had (0-30 days, 30-60 days, 60-90 days) were retained in the final model. For every start the horse had in the previous 30-60 days, the likelihood of a horse fall increased by 25% (Odds Ratio 1.25, $P<0.001$)

**Conclusions** Results suggest that number of starts accumulated in the last 30, 60 and 90 days for the horse are associated with increased odds of a horse fall occurring. It has been reported that race (jump) horses in Australia are predisposed to catastrophic limb injury due to physiological damage sustained from loading incurred during high speed exercise, prior to race starts (Boden *et al.,* 2007). Subclinical limb injury incurred from prior competition starts may reduce the horse's ability to jump fences with the appropriate level of precision and strength, putting the horse at an increased risk of error and therefore a horse fall.

**References**
Boden, L.A., Anderson, G.A., Charles, J.A., Morgan, K.L, Morton, J.M., Parkin, T.D.H., Clarke, A.F., and Slocombe, R.F. Risk factors for thoroughbred racehorse fatality in jump starts in Victoria, Australia (1989-2004). 2007 Equine Veterinary Journal 39 (5), 422-428.

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# Yeast supplementation may have a positive effect on faecal microbiome in ponies fed high starch and high fibre diets

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**Application** VistaEQ supplementation to equine diets can be potentially used to prevent acidosis and increase fibre digestibility in equids. It may help to meet energy requirements of performance horses while maintaining gut health.

**Introduction** High-starch diets are known to have detrimental effects on the hindgut microbiota of the horse, while diets high in fibre are known to maintain a healthy gut and reduce disease incidence (Harris *et al*., 2016). Fibrous feedstuffs tend to have lower energy density and digestibility compared to starch-rich processed feeds, hence, the reason why many performance horses are fed the latter. Thus, there is a need to develop alternative feeding strategies in order to meet the energy requirements of high-performance horses, whilst maintaining healthy digestive microbiota and welfare of the horse. Yeast supplements have been shown to improve fibre digestibility and thus increase feed efficiency as well as negating the deleterious effects of starch on the hindgut environment. The aim of this study was to determine the effect of VistaEQ, a product containing 4% live yeast *Saccharomyces cerevisiae* (*SC*) strain Y1242, on faecal microbial populations of ponies fed high-starch or high-fibre diets.

**Material and methods** Illumina next generation sequencing of the V3-V4 region of the 16S rRNA gene was performed. Seven mature Welsh ponies were used in an incomplete 4x4 Latin square design consisting of 4 x 19-day periods. Following DNA extractions from faeces and library preparation, α-diversity and LEfSe analysis were performed using 16S metagenomics pipeline in QIIME. The differences in microbial relative abundance were considered significant when LDA score (log10) >2 (Segata *et al.,* 2011).

**Results** Supplementation with VistaEQ increased the relative abundance of Lachnospiraceae and Dehalobacteriaceae families in the faeces of horses fed the high-fibre diet. These bacteria are associated with healthy core microbiome in the large intestine of the horse (Dougal *et al.,* 2013). Moreover, supplementation increased the counts of fibrolytic bacteria (*Ruminococcus*) when fed with high-fibre diet and reduced counts of lactate producing bacteria (*Streptococcus*) when high-concentrate diets were fed. In addition, VistaEQ increased the relative abundance of some acetic, succinic acid producing bacterial genera (*Succinivibrionaceae*) and butyrate (*Roseburia*) producing bacteria when fed with high starch and high fibre diets, respectively.

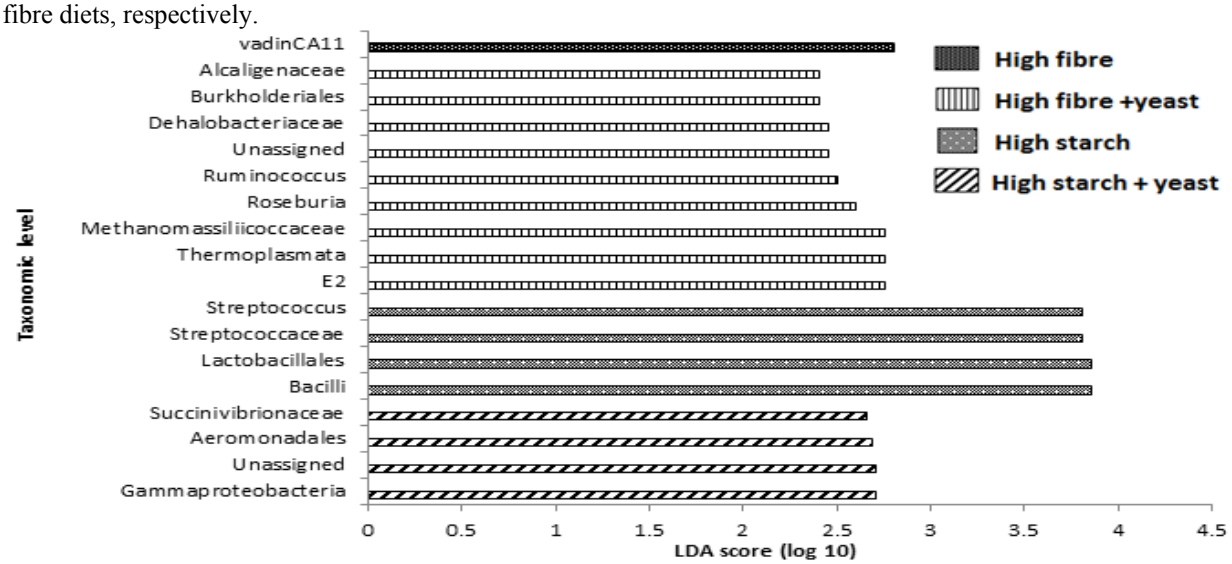


**Figure 1** Linear discriminant analysis effect size (LEfSe) showing significant taxa for each diet

**Conclusion** VistaEQ may have positive effects on the utilization of fibre in ponies as well as reducing risk of acidosis development when diets rich in starch are fed.

**Acknowledgements** The authors gratefully acknowledge the funding received from AB Vista.

**References**

Dougal, K., de la Fuente, G., Harris, P. A., Girdwood, S. E., Pinloche, E., Newbold, J. C. 2013. PLoS ONE 8.
Harris, P. A., Ellis, A. D., Fradinho, M. J., Jansson, A., Julliand, V., Luthersson, N., Santos, A. S., Vervuert, I. 2016. Animal 10, 1-10.
Segata, N., Izard, J., Waldron,.L., Gevers, D., Miropolsky, L., Garrett, W. S. and Huttenhower, C. 2011. Genome Biology 12, R60.

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# Crib biting and equine Gastric Ulceration Syndrome: is there a physiological link?

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**Application** Equine Gastric Ulceration Syndrome and crib biting, are conditions that have previously been linked together. A greater understanding of any link between these conditions would allow improved management and equine welfare.

**Introduction** Equine Gastric Ulceration syndrome **(**EGUS) is highly prevalent within performance horses but it has also been identified in horses in light work, broodmares and semi-feral animals. However, domesticated horses in work appear to suffer with EGUS in greater severity (Ward *et al.,* 2015). Crib biting (CB) is a stereotypical behaviour, a coping mechanism to deal with environmental stress (McBride and Hemmings, 2009). Previously these two conditions have been linked (Nichol *et al.,* 2002), however the exact mechanism linking both conditions is unclear. A recent report by Scott *et al.* (2017) identified that CB horses did not display anatomical differences in G Cell concentrations in cadaver stomachs compared to non-CB controls. Thus CB horses are not anatomically predisposed in the stomach to EGUS. Therefore any link between CB and EGUS may be physiological upregulation of gastrin through nervous innervation during crib biting, making the stomach more acidic.

The aim of this study was to identify if upregulation of stomach acid led to a lower stomach pH in CB horse compared to non-CB controls using cadavers.

**Material and methods** A total of 18 stomachs were collected from horses slaughtered for human consumption, CB (n=9) non-CB (n=9). CB behaviour was either identified in lairage or from dental pathology. Data of breed, sex and age were collected from passports. Age mean 17 ± 5 years, some ages were unknown. Sex mares, (n=9) and geldings (n=9), breeds were Thoroughbreds, sport horses or unrecorded but of sport horse type.
Stomachs were collected post slaughter, part of the selection criteria were stomachs with limited distention to prevent too much acid buffering from the feed content. Stomachs were transported to the laboratory in an insulated box. On return stomachs were placed in an incubator at 37 °C to ensure the temperature of the content would represent a live horse and reflect a true pH. Incisions were made in the fundic and pyloric regions and pH of the lining was measured in each region with a hand held pH probe. The contents, collected from the pyloric region, was passed through a muslin bag and the pH of the acid was measured using a pH probe. Following this stomachs were opened by an incision along the greater curvature to check for ulceration. Data were analysed by ANOVA in Genstat.

**Results** Of the 18 horses, ulceration matching grade 1 criteria were identified in 2 CB and 2 non-CB stomachs. There was no difference in pH between tissue regions or pyloric digesta between CB and non-CB horses ($P>0.05$).

**Table 1** Mean pH of regions and digesta for CB and non-CB stomachs

| CB Fundic | NCB Fundic | CB Pyloric | NCB Pyloric | CB Digesta | NCB Digesta |
|---|---|---|---|---|---|
| 4.822 | 4.656 | 4.592 | 4.739 | 4.489 | 4.509 |

**Conclusion** Here we report no differences in the pH of stomach acid in cadavers from CB horses compared to non-CB horses. Recent work by Scott *et al.* (2017) identified that CB and non-CB stomachs were not anatomically different. Here we demonstrate, from cadavers, stomach pH itself did not differ between CB and non-CB horses. Some care should be taken in interpretation of results, changes within the stomach may have occurred following slaughter, thus cadavers may not be truly representative of the live horse. If there is a relationship between CB and EGUS it is more complex than simply greater acidity in the stomachs of CB horses from an upregulation of gastrin.

**References**

McBride, S.D., Hemmings, A. 2009. Journal of Equine Veterinary Science 29 (1), 10-16.
Nichol, C.J., Davidson, H.P.D., Harris, P.A., Waters, A.J. and Wilson, A.D. 2002. Veterinary Record 151, 658-662.
Scott, L., de Lavis, I. and Daniels, S. P. 2017. Equine Veterinary Education 29 (S8), 11.
Ward S, Sykes B W, Brown H, Bishop A. and Penaluna, L A. 2015. Equine Veterinary Education 27(12), 655-657.

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# The impact of isolation, a mirror and companion on the heart rate and behaviour of horses during loading

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**Application** Horses preferred loading with a companion than loading alone. A mirror was preferential to isolation. The companion and mirror acted as a distraction and reduced the instances of behaviours associated with irritation. This information can be used when dealing with horses that are nervous to load.

**Introduction** Horses are the most commonly travelled type of livestock, however transportation can negatively impact breeding success, performance and stress levels. This study aimed to identify the impact loading alone, with a mirror or with a companion had on the heart rate (HR) and behaviours of the horse. Previous research by Kay and Hall (2009), found that horses travel better with a companion but prefer a mirror to travelling alone, this study did not focus on the loading process, instead on the travelling aspect. HR was significantly lower and behaviours associated with stress fewer.

**Material and methods** Six college horses ($n$=6) of varying breed, gender and age were used. Data were collected over three consecutive days. Each of the horses had travelled previously but had not travelled for a minimum of six weeks prior to data collection. Horses were randomly assigned into three groups to determine the order of the three treatments (isolation, mirror and companion). An Ifor Williams 505 horse trailer was used with the capacity to carry two horses up to 16.2hh. A Polar Equine S610i heart rate monitor, fitted to the equine band, recorded HR continuously, the mean was calculated and recorded. Each horse was habituated to the HR monitor and a rest HR was recorded prior to data collection. The same companion horse was used throughout and was acquainted to each of the subjects. The mirror, one metre square in diameter, was acrylic and safe for use with horses, it was securely attached to the front of the trailer at horse head level. The same handler who was experienced in the transportation of horses, was responsible for the loading and preparing of horses. A haynet was provided for each horse at the front of the trailer. The ethogram contains solely behaviours associated with stress, these have been adapted from McDonnell's Equid Ethogram (2003). Behaviours were recorded using continuous observation. Data was analysed in MiniTab 17 using a general linear model ANOVA to assess HR and a Chi Sqaure test for goodness of fit was used to analyse the behavioural results.

**Results** The behaviours 'eat', 'head turn', 'sniff', 'stamp' and 'step backwards' were found to be significant (table 1). The companion treatment resulted in a reduction in the behaviours 'eat', 'head turn' and 'step backwards'. The behaviour 'stamp' was reduced when the mirror or companion was present there was a significant increase when isolated. 'Sniff' was displayed most frequently in the mirror and companion treatments. HR were found to be insignificant.

**Table 1** Mean frequencies of significant behaviours during loading

| Behaviour | Mean Frequency Isolation | Mean Frequency Mirror | Mean Frequency Companion | p value |
|---|---|---|---|---|
| Eat | 11.5 | 9.5 | 6.83 | $p = 0.029$ |
| Head Turn | 15 | 13.3 | 6.83 | $p = 0.002$ |
| Sniff | 5.3 | 12.17 | 11.67 | $p = <0.001$ |
| Stamp | 3.83 | 1.5 | 1 | $p = 0.002$ |
| Step Backwards | 1.17 | 1.83 | 0.17 | $p = 0.018$ |

**Conclusion** Loading with a companion found horses were more willing to go forward, they also appeared to be distracted by the other horse so behaviours such as 'eat' and 'head turn' were reduced. In a similar way, the mirror acted as a distraction with horses displaying investigative behaviours and fewer behaviours associated with irritation. Horses preferred loading with a companion than isolation, and in line with previous findings a mirror was preferential to isolation.

**Acknowledgements**
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**References**
Kay, R. and Hall, C. 2009. The use of a mirror reduces isolation stress in horses being transported by trailer. Applied Animal Behaviour Science 116(2-4), 237-243.
McDonnell, S.M. 2003. The Equid Ethogram: A practical field guide to horse behavior. 1st edn. New York, NY, United States: Distributed to the trade by National Book Network.

# Characterization of Chilean corralero mare milk in terms of milk composition from elite breeding farms in Central-Southern Chile

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**Application** Mare milk of Chilean corralero breed has the potential to provide an alternative milk for human nutrition characterised by low milk fat content and low energy concentration.

**Introduction** Mare's milk is attracting increasing interest from consumers due to its high content of vitamins and minerals, better digestibility and lower content of fat in comparison with cow's milk. Depending on the region, different breeds of horses are used for this purpose, and milk composition may vary among breeds (Uniacke-Lowe *et al*, 2010). Chilean corralero horse is a relevant breed in Chile given its contribution to the national sport ("rodeo"), farm work with cattle, horse riding and export of stallions (García *et al*, 1997). However, the nutritional value of mare's milk has not been assessed yet. Then, the objective of this study was to characterise mare milk of Chilean corralero breed in terms of milk composition.

**Material and methods** Milk was collected manually in November 2016 (late spring) from 29 mares one hour after foal stopped suckling. Breeding mares came from two elite breeding farms in Central-Southern Chile (Los Rios Region). Mares were healthy, age ranged from 4 to 13 y, were in their 1$^{st}$ to 5$^{th}$ foaling, and body weight ranged 400-450 kg. Mares were grazing a permanent pasture which was sampled once to obtain a composite sample per farm and analysed for dry matter (DM), crude protein (CP), neutral detergent fibre (NDF), acid detergent fibre (ADF) and ash contents. Mares had access to mineral blocks for supplementation with no shed available. Milk samples were analysed for milk fat (MF), milk protein (MP), total solids, ash and gross energy (GE). Lactose content was estimated by difference (lactose = milk solids – MF – MP – total ash). Descriptive statistics were obtained for each group and both were compared in terms of days in milk (DIM), age, number of foaling and milk composition by ANOVA using Genstat 18 (VSNI).

**Results** Both groups were similar in terms of DIM (mean = 48 d), age of mares (mean = 9 y), and number of foaling (mean = 3.75). However, farm V had greater MF, MP, and mineral contents than farm S, leading to a greater total solids content (Table 1) and a greater GE concentration in farm V. These differences could be explained by the variation in pasture quality (Dureau, 1994) since pasture differed noticeably in CP and DM contents (Table 2). Milk composition in this study was between the range reported for mare milk (Uniacke-Lowe *et al*, 2010) with MF content an GE concentration mean values slightly lower than the average values calculated for the species (1.21 g/100 and 480 kcal/kg, respectively).

**Table 1** Mean mare groups characteristics and milk composition of mares from two Chilean corralero breeding farms

| | DIM (d) | Age (y) | No. foaling | MF (g/100ml) | MP (g/100ml) | Lactose (g/100ml) | Total solids (g/100ml) | Total ash (g/100ml) | GE (kcal/kg) |
|---|---|---|---|---|---|---|---|---|---|
| Farm S | 49.6 | 8.5 | 4.3 | 0.61 | 2.05 | 6.56 | 9.64 | 0.42 | 413.2 |
| SEM (n=15) | 5.02 | 0.62 | 0.53 | 0.090 | 0.068 | 0.082 | 0.107 | 0.017 | 6.85 |
| Farm V | 46.4 | 10.4 | 3.2 | 0.94 | 2.32 | 6.49 | 10.22 | 0.47 | 439.3 |
| SEM (n=14) | 5.41 | 1.42 | 0.45 | 0.086 | 0.066 | 0.079 | 0.103 | 0.210 | 6.61 |
| P value | NS | NS | NS | 0.013 | 0.009 | NS | <0.001 | 0.016 | 0.011 |

**Table 2** Nutritional value of one composite pasture snip sample per breeding farm

| | DM (%) | CP (g/100g) | NDF (g/100g) | ADF (g/100g) | Ash (%) |
|---|---|---|---|---|---|
| Farm S | 20.6 | 21.0 | 50.5 | 24.5 | 8.9 |
| Farm V | 24.6 | 15.0 | 52.6 | 27.2 | 8.5 |

**Conclusion** Milk from Chilean corralero mares has a slightly lower MF content and GE concentration than the average value for the species. Mare milk quality shows a potential influence of forage composition, particularly N supply.

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**References**

Uniacke-Lowe, T., Huppertz, T., Fox, P.F. 2010. International Dairy Journal 20(9) 609-629.
Doreau, M. 1994. Le Lait, INRA Editions, 74 (6), 401-418.
García, M., Cabezas, I., Guzmán, R., Valenzuela, S., Merino, V., Pérez, R. 1997. Avances en Ciencias Veterianrias 12(1) 45-52.

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# The effect of four liquid feeding strategies on the growth, carcass quality and feed efficiency of grow-finisher pigs

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**Application** Fermenting the whole diet prior to feeding reduced final live weight, carcass weight and kill out percentage and resulted in a deterioration in feed efficiency when compared with all other feeding regimens.

**Introduction** Fermenting liquid feed prior to feeding can be beneficial to pig gastrointestinal health due to reduced pH, proliferation of lactic acid bacteria and decreased enterobacteria counts (Lawlor *et al.,* 2002; Canibe *et al.,* 2003). However, the effect of diet fermentation on growth and feed efficiency is inconsistent in the literature. Fermenting only the cereal fraction of the diet may be preferable to whole diet fermentation, as microbial decarboxylation of free amino acids is avoided (Canibe *et al.*, 2007). The aim of this study was to compare the effect of four liquid feeding regimens (whole diet fermentation, fermenting the cereal fraction only, fresh liquid feed and single space wet/dry feeding) on growth, feed efficiency and carcass quality of grow-finisher pigs.

**Material and methods** This project received ethical approval from Teagasc Animal Ethics Committee. A total of 216 pigs (29.8kg ± 1.00 SEM) housed in same sex pens of 6 pigs/pen (n=9 pens/treatment) were allocated to 1 of 4 dietary treatments: (1) Single space wet/dry feeders (Wet/Dry) where pigs mixed water and meal at the point of feeding, (2) Fermented cereal diet (Ferm Cer) where the cereal fraction (38% barley, 40% wheat) of the diet was fermented prior to feeding, (3) Fermented whole diet (Ferm Whole) where the whole diet (78% cereal, 22% balancer containing soyabean meal, soya oil, synthetic amino acids, phytase, minerals and vitamins) was fermented prior to feeding, (4) Fresh liquid diet (Fresh) where the diet and water were mixed immediately prior to feeding. All diets were isoenergetic and isonitrogenous. The experiment lasted 70 days during which growth and feed intake were recorded. The Ferm Cer and Ferm Whole treatments were prepared using a starter culture (Sweetsile, Agway, UK) allowing an initial fermentation of 48 hours. Thereafter, fermentation tanks were replenished daily to replace feed consumed by the pigs with either cereal or whole diet at a water:meal ratio of 2.5:1. Body weight and feed intake were recorded throughout the experiment and at slaughter, carcass weight, muscle depth and fat depth were recorded. Data were analysed using the MIXED procedure of SAS 9.4.

**Results** Pigs fed the Ferm Whole diet had lower average daily gain (P<0.001), final live weight (P<0.001), carcass weight (P<0.001) and kill out percentage (P<0.05) as well as poorer feed conversion ratio (FCR; P<0.001) compared to pigs fed all other treatments. Pigs fed Wet/Dry had a better FCR (P<0.001) than those fed the Ferm Cer and Ferm Whole diets but a similar FCR to those fed Fresh.

**Table1** Effect of four liquid feeding strategies on the growth, carcass quality and feed efficiency of grow-finisher pigs

| Item | Treatment | | | | P-Value | | | |
|---|---|---|---|---|---|---|---|---|
| | (1)Wet/Dry | (2)Ferm Cer | (3)Ferm Whole | (4) Fresh | SEM | mix | sex | mix*sex |
| Initial live weight, kg | 30.0 | 29.0 | 29.7 | 30.4 | 1.00 | 0.16 | <0.01 | 0.08 |
| Final live weight, kg | 103.6$^{a}$ | 105.6$^{a}$ | 95.8$^{b}$ | 103.9$^{a}$ | 0.76 | <0.001 | <0.001 | <0.001 |
| Average daily gain, g/d | 1085$^{a}$ | 1081$^{a}$ | 928$^{b}$ | 1077$^{a}$ | 13.54 | <0.001 | <0.001 | 0.24 |
| Average daily feed intake, g/d | 2408$^{a}$ | 2596$^{a,b}$ | 2701$^{b}$ | 2532$^{a,b}$ | 59.76 | <0.01 | 0.22 | 0.82 |
| Feed conversion ratio, g/g | 2.24$^{a}$ | 2.40$^{b}$ | 2.98$^{c}$ | 2.36$^{a,b}$ | 0.04 | <0.001 | <0.001 | <0.01 |
| Carcass weight, kg | 79.2$^{a}$ | 80.8$^{a}$ | 70.9$^{b}$ | 78.7$^{a}$ | 0.98 | <0.001 | 0.29 | 0.58 |
| Kill out, % | 76.7$^{a}$ | 76.6$^{a}$ | 75.2$^{b}$ | 76.2$^{a,b}$ | 0.34 | <0.05 | <0.001 | 0.72 |
| Muscle depth, mm | 49.7 | 48.8 | 46.0 | 48.8 | 1.1 | 0.16 | 0.14 | 0.39 |
| Fat depth, mm | 11.8 | 12.2 | 13.3 | 12.6 | 0.41 | 0.09 | <0.05 | 0.48 |
| Lean meat % | 57.6 | 57.1 | 55.8 | 56.8 | 0.44 | 0.06 | <0.05 | 0.44 |

**Conclusion** Fermenting the whole diet reduced growth, final live weight, carcass weight and kill out percentage, while FCR deteriorated compared with all other treatments. Pigs fed wet/dry diets had better FCR than pigs fed fermented liquid diets but FCR was similar to those fed Fresh.

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**References**

Canibe, N. and Jensen, B. B. 2003. Journal of Animal Science 81, 2019-2031.
Canibe, N., Hojberg, O., Badsberg, J. H. and Jensen, B. B. 2007. Journal of Animal Science 85, 2959-2971.
Lawlor, P. G., Lynch, P. B., Gardiner, G. E., Caffrey, P. J. and O'Doherty, J. V. 2002. Journal of Animal Science 80**,** 1725-1735.

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# The supplementation of cholecalciferol and ergocalciferol sources in finisher pig diets to increases pork vitamin D content and improve pork quality

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**Application** The biofortification of pork is a potential food based strategy for increasing human vitamin D dietary intake, with proceeding benefits of improved pork quality.

**Introduction** Vitamin D inadequacy is now recognised as a public health concern (Cashman *et al*., 2017). Due to this wide-spread inadequacy of vitamin D intake, there is a need for more effective food-based strategic approaches to produce a wider range of sustainable natural vitamin D-enriched foods with potential to increase vitamin D dietary intakes. The fortification of pig diets with vitamin D sources, in order to enhance the pork vitamin D content may have potential to be one natural food-based solution. As well as increasing the vitamin D content of pork, the supplementation of vitamin D to pigs may also have the potential to enhance additional pork quality properties, such as tenderness, colour and antioxidant capacity. Therefore, the objective of the current study was to investigate the effect of inclusion of synthetic and natural vitamin D sources in pig diets on total vitamin D activity of the pork meat as well as pork quality and antioxidant status.

**Material and methods** The experiment was designed as a complete randomised block and approved under University College Dublin Animal Ethics Committee (AREC-13-79-O'Doherty). One hundred and twenty (60 males, 60 females) pigs with initial live weight of 58.0 kg (SD 4.6) were blocked according to live weight and sex, within each block assigned to one of four dietary treatments (n = 30). Dietary treatments consisted of ;( 1) 2000 IU vitamin D□/kg of feed (Vit D□); (2) 50 µg (2000 IU equivalent) of 25-OH-D□/kg of feed (25-OH-D□); (3) 2000 IU vitamin D□/kg of feed (Vit D□); (4) 2000 IU vitamin D□-enriched mushrooms/kg of feed (Mushroom D□). Pigs were weighed at the beginning of the experiment day 0, 28 and day 55 (slaughter). Individual daily feed intakes were recorded using single space computerised feeders. Following overnight chilling of the carcass at 4° C, the *Longissimus thoracis* (LT) was excised for vitamin D analysis carried out by High Performance Liquid Chromatography. The LT steaks were analysed for Warner Bratzler shear force (WBSF) values, cook loss and drip loss. Additionally, LT steaks were stored in modified atmosphere packs (80 % $O_2$: 20 % $CO_2$) for up to 14 days at 4° C for antioxidant and colour analysis. LT steaks were also analysed for lipid peroxidation (LPO), total phenolic compounds (TPC), 2,2-diphenyl1-1-picrylhydrazyl (DPPH) and ferric reducing antioxidant power. Statistical analysis was carried out using PROC MIXED procedure of SAS (SAS, version 9.4).

**Results** Pigs offered the 25-OH-$D_3$ had the highest (P<0.001) LT total vitamin D activity compared to all treatments (Figure 1). Pigs offered the Mushroom $D_2$ diet had higher (P<0.01), final body weight, increased average daily gain and improved feed conversion ratio compared to other treatments (Table 1). There was no effect (P>0.05) of dietary treatment on cook loss, WBSF values and drip loss. There was a significant treatment effect (P<0.05) and time effect (P<0.05) observed for LT lightness (L*), redness (a*) and yellowness (b*) colour. The Mushroom $D_2$ had overall (P<0.05) lower L*, with the highest a* and lowest b* values compared to other treatments. The supplementation of Mushroom $D_2$ and 25-OH-$D_2$ increased (P<0.05) pork antioxidant capacity.

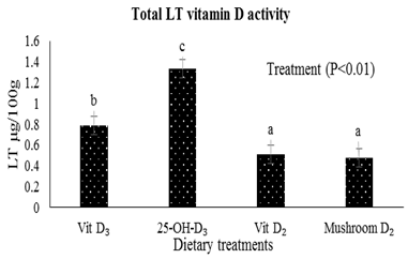


**Figure 1** Effect of dietary treatment on total LT vitamin D activity.

**Table 1** Effect of dietary treatment on animal growth

| Item | Dietary treatments | | | | | P-value |
|---|---|---|---|---|---|---|
| | Vit $D_3$ | 25-OH-$D_3$ | Vit $D_2$ | Mushroom $D_2$ | SEM[2] | Treatment |
| ADG[1] | 0.91[a] | 0.93[a] | 0.95[a] | 1.01[b] | 0.023 | 0.011 |
| ADFI[1] | 2.56 | 2.75 | 2.64 | 2.59 | 0.055 | 0.116 |
| FCR[1] | 2.87[b] | 2.96[b] | 2.83[b] | 2.61[a] | 0.058 | 0.004 |

[1]ADG (kg/d), ADFI (kg/d), FCR (kg/kg). [2]SEM, standard error of the mean

**Conclusion** In conclusion results indicate that the supplementation of 25-OH-$D_3$ exhibited the highest LT muscle vitamin D content, which may be one strategic approach for tackling low human vitamin D intakes. Additionally Mushroom $D_2$ demonstrated potential to improved pig performance, pork colour stability and antioxidant capacity.

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**References**

Cashman, K. D. and Hayes, A. 2017. Meat Science 132, 196-203.

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# Effect of extrusion of field beans on energy and crude protein digestibility (*in-vitro* and *in-vivo*), growth and carcass quality of grow-finisher pigs

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**Application** Field beans are a good energy and protein source and can be fed at up to 37% inclusion in pig diets. Extrusion of field beans increased the digestible energy (DE) but decreased the digestible crude protein (dCP) value of field beans.

**Introduction** European production of pig meat is highly dependent on the importation of soybean meal (SBM). Due to policy support, the area of field beans produced has increased greatly in recent years in Ireland and the U.K. Therefore, there is renewed interest in field beans as an energy and protein source. Extrusion of raw ingredients can increase their nutritional value (Sun *et al*. 2006). It was hypothesised that nutritional value of field beans can be improved by extrusion.

**Material and methods** The same batch of field beans rolled and treated with propionic acid prior to storage was used in 3 experiments to determine its nutritional value for pigs. A portion was extruded (95°C, 5x5x6mm die, EXTRU-tech E525, Sabetha, Kansas). In exp. 1 the *in-vitro* dry matter (DM), organic matter (OM) and CP digestibility of raw field beans (rFB) and extruded field beans (eFB) was determined in duplicate for each sample using an adapted protocol from Boisen and Fernandez (1995). In exp. 2 the digestible energy (DE) and dCP values of rFB and eFB was determined using the difference method (Zhang and Adeola, 2017). A total of 48 male pigs (23.3 kg; ±0.66 SEM) were housed in pairs and allocated to 1 of 3 dietary treatments 1) Control diet based on barley and SBM (13.9 MJ DE/kg, 0.99 g SID Lys/kg), 2) 50% control diet+50% rFB and 3) 50% control diet+50 % eFB. After 7 days of adaptation to the diets, faecal samples were collected over 3 consecutive days, pooled and analysed for DM, gross energy (GE), CP and acid insoluble ash as inert marker. In exp. 3, the effect of replacing SBM by rFB and eFB in grow-finisher diets on growth, carcass quality, apparent ileal digestibility (AiD), and total tract digestibility (ATTD) of DM, OM, GE and CP were investigated. A total of 60 pigs (46.2 kg; ±0.52 SEM) were housed in same sex pairs (n=10 pairs/treatment) and randomly allocated to 1 of 3 dietary treatments (13.1 MJ DE/kg and 0.89 SID Lys/kg): 1) Control diet (CON) barley and soya bean meal diet, 2) 36.7% rFB diet (rFBD) and 3) 36.7% eFB diet (eFBD). The experiment lasted 63 days during which live weight and feed intake was recorded every 2 weeks and average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. Fresh faeces were collected on days 61 and 62 for subsequent ATTD. On day 63 pigs were slaughtered. At slaughter carcass weight, muscle depth and fat depth were recorded and ileal digesta content was collected for AiD determination. Data was analysed using the MIXED procedure of SAS 9.4.

**Results** In exp. 1, *in-vitro* digestibility of eFB compared to rFB was unchanged for DM (73.4 *vs.* 74.4 %; ±0.23 SEM; P=0.12), increased for OM (73.0 *vs.* 74.5 %; ± 0.26 SEM, P=0.02), and increased for CP (90.7 *vs.* 91.0 %; ±0.02 SEM; P=0.04). In exp. 2, the DE value of rFB was 14.38 MJ/kg and the dCP was 22.8 % (on DM basis). The DE value for eFB was 15.75 MJ/kg and the dCP was 21.7 % (on DM basis). In exp. 3, ADFI was higher for rFBD and eFBD compared to CON, while ADG, FCR and carcass quality parameters did not differ among treatments (Table1). Dry matter and OM AiD were higher in pigs fed eFBD than CON but similar to rFBD; DM, OM and CP ATTD was similar for all diets (Table2).

**Table 1.** Growth performance and carcass quality (exp.3)

| | Treatment | | | | |
|---|---|---|---|---|---|
| | CON | rFBD | eFBD | SEM | p-value |
| ADG, g/day | 998 | 1059 | 1027 | 21.5 | 0.12 |
| ADFI, g/day | 2291[a] | 2453[b] | 2403[b] | 43.8 | 0.03 |
| FCR, g/g | 2.30 | 2.32 | 2.35 | 0.027 | 0.42 |
| Carcass weight, kg | 84.4 | 87.9 | 85.7 | 1.13 | 0.12 |
| Kill out, % | 77.2 | 77.7 | 77.1 | 0.37 | 0.48 |
| Fat depth, mm | 11.2 | 12.8 | 12.2 | 0.56 | 0.14 |
| Muscle depth, mm | 60.7 | 59.7 | 57.8 | 2.62 | 0.47 |

**Table 2.** Apparent ileal and total tract digestibilities (exp. 3)

| | Treatment | | | | |
|---|---|---|---|---|---|
| | CON | rFBD | eFBD | SEM | p-value |
| AiD of DM | 65.4[a] | 69.9[a,b] | 72.3[b] | 1.61 | 0.02 |
| AiD of OM | 68.5[a] | 73.2[a,b] | 75.2[a] | 1.46 | 0.01 |
| AiD of CP | 63.7 | 63.3 | 63.3 | 3.2 | 0.99 |
| AiD of Energy | 66.1 | 71.0 | 71.2 | 2.18 | 0.33 |
| ATTD of DM | 88.3 | 85.9 | 88.7 | 0.29 | 0.13 |
| ATTD of OM | 89.9 | 87.9 | 90.1 | 0.27 | 0.15 |
| ATTD of CP | 85.2 | 84.0 | 86.2 | 0.57 | 0.33 |

**Conclusion** Field beans are a good energy and protein source for pigs. Extrusion of field beans did not improve their nutritional value for pigs.

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**References**

Boisen and Fernandez 1995. Animal Feed Science and Technology 51, 29-43.
Sun, T., Lærke, H. N., Jørgensen, H., and Knudsen, K. E. B. 2006. Animal Feed Science and Technology 131, 67-86.
Zhang, F., and O. Adeola. 2017. Animal Nutrition 3, 344-352.

# Growth and haematological assessment of broiler chickens fed bitter leaf (*Vernonia amygdalina)* supplemented diets

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**Application** Attempts at addressing the challenges of antimicrobial resistance in animal production has made researchers focus attention on the exploitation of non-conventional feed resources and herbal plants.

**Introduction** The use of antimicrobial agents as growth promoters are being discouraged due to human and animal health issues, mainly resulting from development of antimicrobial resistance. Attempts at addressing these challenges, as well as motivation by consumer preference towards chemical-free products, have focused researchers attentions on the exploitation of non-conventional feed resources and herbal plants. Some of these useful herbs are indigenous to Africa and have been reported to influence nutrient utilization by chickens (Windisch *et al.,* 2007) and enhance performance of broiler chickens (Carrijo *et al.,* 2005; Odoemelam *et al.,* 2012). This study was undertaken to assess the growth performance, haematological and serum biochemical indices of broiler chickens fed diets supplemented with graded levels of bitter leaf (*Vernonia amygdalina*) meal (BLM).

**Material and methods** One hundred and sixty Marshal Strain broiler chicks were divided into 5 treatment groups of 32 chicks each with 4 replicates. The experimental diets were prepared from commercial broiler starter and finisher feeds such that diet 1 (negative control) contained no leaf meal nor antibiotics, diet 2 contained 0.10% oxytetracycline (positive control), while diets 3, 4 and 5 had BLM inclusion at 1%, 2% and 3% respectively. The feeding trial lasted for 42 days, after a brooding period of two weeks. Starter diet was offered for 14 days during and 14 days after brooding and finisher diet offered for the last 28 days. Data generated during the experiment were subjected to a one-way analysis of variance in a Completely Randomized Design using the general linear model programme of the SPSS computer package and means were separated by Duncan multiple range test.

**Results** Results on growth performance showed that all parameters were significantly affected by dietary treatments. Total feed intake ranged from 7876 g/bird in the 3% BLM diet to 8145 g/bird in the 2% BLM. Final weight gain was least (3577 g/bird) in the negative control diet and highest (3816 g/bird) in the oxytetracycline diet. Birds on 1% and 2% BLM compared favourably with the oxytetracycline birds. Feed conversion and protein efficiency ratios (2.21 and 2.40 respectively) were better in the oxytetracycline diet.

**Table 1** Performance, haematological and serum characteristics of chickens fed oxytetracycline and bitter leaf meal diets

| Performance characteristics | Negative control | 0.10% oxytetra-cycline | 1% bitter leaf | 2% bitter leaf | 3% bitter leaf | SEM |
|---|---|---|---|---|---|---|
| Avg. total feed intake (g) | $7985^{b}$ | $7958^{ab}$ | $8065^{bc}$ | $8145^{c}$ | $7876^{a}$ | 33.63* |
| Avg. final weight (g) | $3577^{c}$ | $3816^{a}$ | $3761^{ab}$ | $3744^{ab}$ | $3688^{b}$ | 29.52* |
| Avg. total weight gain (g) | $3364^{c}$ | $3602^{a}$ | $3547^{ab}$ | $3535^{ab}$ | $3480^{b}$ | 31.08* |
| Feed conversion ratio | $2.37^{c}$ | $2.21^{a}$ | $2.27^{b}$ | $2.30^{b}$ | $2.26^{b}$ | 0.02* |
| Protein efficiency ratio | $2.23^{c}$ | $2.40^{a}$ | $2.33^{b}$ | $2.30^{b}$ | $2.34^{b}$ | 0.02* |
| White blood cell ($x10^{3}/\mu l$) | $87.30^{ab}$ | $82.50^{b}$ | $92.10^{a}$ | $90.00^{a}$ | $87.50^{ab}$ | 1.83* |
| Monocytes (%) | $13.20^{a}$ | $8.50^{b}$ | $9.75^{b}$ | $7.75^{b}$ | $9.20^{b}$ | 0.80* |
| Neutrophils(%) | $19.00^{a}$ | $8.05^{b}$ | $13.2^{ab}$ | $9.90^{b}$ | $9.65^{b}$ | 1.94* |
| Haemoglobin (g/dl) | $11.90^{b}$ | $14.50^{a}$ | $12.40^{ab}$ | $10.80^{b}$ | $12.10^{ab}$ | 0.73* |
| Total protein (g/dl) | $4.55^{a}$ | $3.55^{b}$ | $4.10^{ab}$ | $4.45^{a}$ | $4.10^{ab}$ | 0.21* |

On haematological parameters, monocytes, and neutrophils were significantly highest in the negative control diet, while haemoglobin value of 14.50g/dl, was significantly higher in the oxytetracycline diet. For serum parameters, total protein was least in the oxytetracycline diet with a value of 3.55g/dl.

**Conclusion** The results presented in this study showed that the inclusion of the bitter leaf meal had significant positive effects on broiler performance in comparison to the negative control that contained no additive. It was also clear from this study that the leaf meal showed great promise as a good alternative to the commonly used oxytetracycline antibiotic growth promoter in Nigeria.

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**References**

Mohammed, A. A. and Zakariya, A. S. 2012. Research Journal of Animal Science 6, 38-41.
Windisch, W., Schedle, K., Plitzer, C. and Kroismayr, A. 2007. Journal of Animal Science 86, 140-148.

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# Effects of supplementation with the marine derived polysaccharides, laminarin and chitosan, on the growth performance and faecal consistency of pigs during the post-weaning period

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**Application** Chitosan supplementation improves the effects of laminarin on the growth performance of pigs during the post-weaning period.

**Introduction** The removal of antibiotic growth promoters in pig diets in the EU (EC reg no. 1831/2003) and the imminent ban on zinc oxide use at pharmacological doses are motivating the search for natural alternatives to prevent the stress associated decline in pig performance at weaning. Improvements in daily gains and feed efficiency in the post-weaning period were observed after supplementing the marine polysaccharides laminarin (Heim *et al*., 2014) and a chitosan derivative (Chitooligosaccharide) (Walsh *et al*., 2013). Thus, it was hypothesised that supplementing a combination of laminarin and chitosan would enhance pig growth performance and reduce the incidence of post-weaning diarrhoea. The objective of the study was to investigate the effects of supplementing laminarin and/or chitosan on daily gains, feed intake, feed efficiency and faecal consistency during the immediate post-weaning (PW) period.

**Material and methods** The experiment was designed as a 2×2 factorial arrangement. Fifty weaned pigs were blocked by live weight and litter of origin and randomly assigned to one of four treatments: 1) Basal diet (CON) (n=10); 2) Basal & 200ppm laminarin (LAM) (n=5); 3) Basal & 300ppm chitosan (CH) (n=5); 4) Basal & 200ppm LAM & 300ppm CH (CH & LAM) (n=5). The pen served as the experimental unit with 2 pigs/pen and treatment (1) was assigned 10 pens for a follow-on experiment. The basal diet contained 15.3 MJ/kg of digestible energy, 190 g/kg of crude protein and a total lysine content of 13.5 g/kg. The basal diet comprised of wheat, soya beans (toasted), wheat (cooked), soya meal, corn (dehulled), whey powder, soya protein concentrate, glycerol, calcium carbonate, monocalcium phosphate and was offered for 28 days. Faecal scores were recorded daily on a scale ranging from 1 to 5 as follows: 1 = hard, firm faeces; 2 = slightly soft faeces; 3 = soft, partially formed faeces; 4 = loose, semi-liquid faeces; and 5 = watery, mucous like faeces. Body weight gains and feed intake were recorded weekly. The data was initially checked for normality and analysed by repeated measures using the proc mixed procedure of SAS.

**Results** The effects of treatment on average daily gain (ADG), average daily feed intake (ADFI), feed conversion ratio (FCR) and faecal score (FS) are presented in Table 1. There was an interaction between the supplementation of LAM and CH on ADG ($P<0.01$), ADFI ($P<0.0001$) and FS ($P<0.05$). The CH diet had no effect on ADG or FS and caused a significant decrease in ADFI ($P<0.05$) in comparison to the CON group. However, when LAM and CH were offered in combination, ADG, ADFI and FS were improved in comparison to the LAM group. The FS was negatively correlated with ADG ($R = 0.221$, $P<0.05$) and FCR ($R = 0.40$, $P<0.0001$) during the first 14 days post weaning and negatively correlated with FCR for the duration of the experiment ($R = 0.253$, $P<0.001$).

**Table 1** Effects of laminarin and chitosan on ADG, ADFI, FCR and FS from day 0-28 PW (LSM ± SEM)

| | Treatment | | | | | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|
| | CON | CH | LAM | CH & LAM | SEM | CH | LAM | CH x LAM | Time |
| ADG kg/day | 0.323[a,c] | 0.310[a,b] | 0.271[b] | 0.362[c] | 0.017 | 0.022 | 0.997 | 0.003 | <0.0001 |
| ADFI kg/day | 0.643[a] | 0.595[b] | 0.541[c] | 0.635[a,b] | 0.635 | 0.168 | 0.061 | <0.0001 | <0.0001 |
| FCR (kg/kg) | 2.314 | 2.132 | 2.200 | 2.007 | 0.196 | 0.344 | 0.545 | 0.977 | 0.003 |
| FS | 2.54[a] | 2.72[a,b] | 2.92[b] | 2.66[a] | 0.088 | 0.693 | 0.079 | 0.016 | 0.003 |

[a,b,c] Mean values within a parameter with different superscripts were significantly different.

**Conclusion** The combined supplementation of laminarin and chitosan enhanced pig performance in comparison to supplementation of laminarin alone. However, they did not outperform the control group and thus, further studies with a higher laminarin inclusion level are necessary to determine the potential of the combination as a post-weaning supplement.

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**References**

Walsh, A.M., Sweeney, T., Bahar, B., Flynn, B., O'Doherty, J.V. 2013. Animal 7, 571-579.
Heim, G., Walsh, A., Sweeney, T., Doyle, D., O'Shea, C.J, Ryan, M., O'Doherty, J.V. 2014. British Journal of Nutrition 111, 1577-85.

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# Effects of dietary conjugated linoleic acid supplementation on the growth and immune status of broiler chickens

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**Application** Dietary supplementation with conjugated linoleic acid can modify the metabolism-, carcass, improve the growth-, immune status of birds

**Introduction** The growing consumer demand for functional food products that will promote human health and nutritional values has led to the development of n-3 fatty acids animal products (Zhang *et al*., 2008). Dietary conjugated linoleic acid (CLA) is an additive known to improve the quality (yield, oxidative stability, colour and composition) of poultry meat for safe human consumption (Shin *et al*., 2011). This study evaluates the immune and growth responses of broilers to dietary CLA in a maize-soybean diet.

**Material and methods** The field trial was conducted at the teaching and research farm of the University of Calabar, Nigeria. 200 commercial broilers (Ross 308) chicks were used in a complete randomized design with 5 treatments and 4 replications (pen) of 10 birds each. Broilers were managed under the deep litter system in a 2-phase feeding program during a 42-d production period. Diets were identical in nutrient specifications during the same age period according to NRC (2012) recommendations. Conjugated Linoleic Acid (CLA) was added to the basal diet at 5 levels, 0.00%, 0.01%, 0.02%, 0.03% and 0.04% to constitute treatments 1, 2, 3, 4 and 5, respectively. Birds were offered feed and water *ad libitum*. Birds and feed were weighed on a pen basis at 1, 7, 14, 28, 35 and 42 d of age for the determination of growth rate, feed intake, and feed conversion. Mortality was recorded daily. On d 42, 4 birds per pen were randomly selected, weighed and processed for carcass evaluation. Carcass yield in terms of dressed weight/live weight and immune organs (thymus, spleen and liver) were measured. To assay the efficacy of CLA on the immune status of the chicks, only IBD vaccine was administered on days 14 and 24. Data collected on the growth, haematology, carcass and immune status of the birds were statistically evaluated using the one-way ANOVA procedure of SAS 2004. Means were separated by Tukey's honestly significant differences procedure when the overall $F$-test was $P \leq 0.05$.

**Results** No significant (P>0.05) effects of dietary CLA were observed between treatments for final weight and weight gain. Average daily feed intake and feed conversion ratio were significantly (P<0.05) influenced, with birds on 0.04% CLA recording the best conversion ratio. Dressing percentage became higher (P<0.05) at increasing CLA supplementation. Relative immune organs were higher (P<0.05) in birds fed 0.04% CLA diet compared to other groups. Heterophil:Lymphocyte ratio (H:L), total cholesterol (TC) concentrations and Immunoglobulin-M (Ig-M) (%) were lowered (P<0.05) in the serum of birds fed CLA supplemented diets. While Immunoglobulin-G (Ig-G) (%) increased when chickens were fed diets containing increasing levels CLA. Generally, there were significant improvements in the growth, carcass and immune status of birds following dietary CLA supplementation.

**Table1** Responses of broiler chickens to dietary CLA supplementation

| | Treatments | | | | | | |
|---|---|---|---|---|---|---|---|
| Parameter | 0.00% CLA | 0.01% CLA | 0.02% CLA | 0.03% CLA | 0.04% CLA | SED | P value |
| Final weight, g | 1850 | 1900 | 1767 | 1833 | 1767 | 544.30 | 0.15 |
| Av. Daily WG, g/day | 28.65 | 29.47 | 28.72 | 29.24 | 31.17 | 6.13 | 0.08 |
| Av. Daily FI, g/day | $108.70^{a}$ | $111.20^{a}$ | $82.20^{ab}$ | $100.40^{a}$ | $64.10^{b}$ | 46.18 | 0.03 |
| FCR | $3.86^{b}$ | $3.88^{b}$ | $2.86^{ab}$ | $3.43^{b}$ | $2.06^{a}$ | 1.13 | 0.05 |
| Dressing percentage | $72.00^{ab}$ | $72.20^{b}$ | $74.60^{ab}$ | $75.50^{ab}$ | $78.10^{a}$ | 7.00 | 0.04 |
| Abdominal fat (%) | $0.82^{c}$ | $1.44^{a}$ | $1.37^{a}$ | $1.16^{b}$ | $0.41^{d}$ | 0.22 | 0.05 |
| Thymus (%) | $0.58^{d}$ | $0.59^{c}$ | $0.54^{d}$ | $0.65^{b}$ | $0.66^{a}$ | 0.01 | 0.01 |
| Spleen (%) | $0.13^{d}$ | $0.21^{b}$ | $0.19^{c}$ | $0.24^{a}$ | $0.22^{b}$ | 0.01 | 0.01 |
| Liver (%) | $2.57^{c}$ | $2.38^{d}$ | $2.77^{b}$ | $2.58^{c}$ | $2.84^{a}$ | 0.02 | 0.02 |
| H:L | $0.39^{c}$ | $0.43^{a}$ | $0.42^{a}$ | $0.40^{b}$ | $0.31^{d}$ | 0.02 | 0.01 |
| TC (mg/dl) | $165.70^{a}$ | $153.70^{a}$ | $134.30^{b}$ | $132.00^{b}$ | $124.00^{b}$ | 13.02 | 0.05 |
| I-G (%) | $2.17^{c}$ | $2.23^{c}$ | $2.60^{bc}$ | $2.87^{b}$ | $3.53^{a}$ | 0.16 | 0.05 |
| I-M (%) | $2.51^{ab}$ | $1.89^{b}$ | $2.58^{a}$ | $2.06^{ab}$ | $1.92^{ab}$ | 0.20 | 0.04 |

**Conclusion** This study concludes that for improved growth and health performances, up to 0.04% CLA could be supplemented into broiler diet.

**References**

Zhang, H. J., Guo, Y. M., Tian, Y. D. and Yuan, J. M. 2008. British Poultry Science 49, 213 - 221.
Shin, D., Narciso-Graytan, C., Park, J. H., Smith, S. B., Sanchez-Plata, M. X. and Ruiz-Feria, C. A. 2011. Poultry Science 90, 1340 -1347.

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# *In vitro* evaluation of the effects of a *Laminaria hyperborea* extract on commensal and pathogenic bacterial strains frequently isolated from the gastrointestinal tract of weaned piglets

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**Application** Dietary supplementation with a *Laminaria hyperborea* extract could potentially improve the composition of the gut microbiota in the post-weaning piglet by increasing beneficial bacterial species.

**Introduction** Brown seaweed extracts have many beneficial effects when included in the diet of newly weaned piglets. Dietary supplementation with *Laminaria spp.* extracts were associated with improved intestinal morphology and reduced inflammation of the gastrointestinal tract (Walsh *et al*, 2013). Beneficial shifts in the different bacterial groups of the gut microbiota were also reported (Murphy *et al*., 2013). Commensal *Lactobacillus spp.* and *Bifidobacterium spp.* are thought to have beneficial effects, while members of the *Enterobacteriaceae* family are linked to compromised health. The aim of this study was to determine if a *Laminaria hyperborea* extract could show prebiotic potential and antimicrobial activity *in vitro*.

**Material and methods** *Lactobacillus plantarum*, *L. reuteri, Bifidobacterium thermophilum*, *Escherichia coli* and *Salmonella enterica* ser. Typhimurium were revived from cryoprotective beads and subcultured following standard procedures. All bacterial strains were diluted in 10% medium [de Man, Rogosa and Sharpe (MRS) for lactobacilli and bifidobacteria and Tryptone Soya broth (TSB) for *E. coli* and *S.* Typhimurium] to obtain an inoculum of $10^6$-$10^7$colony-forming unit(CFU)/ml. The *L. hyperborea* extract containing 6.2% laminarin and 14.7% fucoidan was produced using an innovative technology optimised for the extraction of these polysaccharides specifically. A two-fold serial dilution (2 mg/ml-0.25 mg/ml) of *L. hyperborea* extract was made in 10% medium. In 96-well microtiter plates, duplicate wells were inoculated with 100 µl of each concentration of the compound and 100 µl inoculum. Control wells containing 100 µl 10% medium and 100 µl inoculum were included. To confirm sterility, blank wells containing only 10% medium as well as a series of wells containing only the serial dilution of the compound were included. Plates were incubated aerobically for all species except for *B. thermophilum* which was incubated anaerobically at 37 °C for 18 h. Final bacterial concentrations were determined after 10-fold serial dilution, spread plating and incubation at 37 °C for 24 h aerobically or for 48 h anaerobically for *B. thermophilum*. The dilution with 5-50 colonies was selected for the calculation of CFU/ml using the formula CFU/ml=average colony numberx50xdilution. All experiments were carried out with technical replicates on three independent occasions (n=3). Prior to statistical analysis, all data were logarithmically transformed. The PROC GLM was used to analyse the data and PDIFF to separate the means (SAS 9.4). Results are expressed as mean ± standard error.

**Results** An increase of *B. thermophilum* (≤0.7 logCFU/ml, $p<0.05$) was observed at 2 mg/ml and 1 mg/ml concentrations of *L. hyperborea* extract as presented in Table 1. *L. plantarum* (≤0.4 logCFU/ml, $p<0.05$) was increased at all concentrations tested. 1 mg/ml was the optimal dose for both strains. Interestingly, this extract had no effect on the population of *L. reuteri*. No antimicrobial activity against *E. coli* and *S.* Typhimurium was observed at all concentrations of *L. hyperborea* extract.

**Table 3** Bacterial counts following exposure to a serial dilution of *L.hyperborea* extract*

| Bacterial strain | Final bacterial concentration (logCFU/ml) | | | | | SEM |
|---|---|---|---|---|---|---|
| | 2 mg/ml | 1 mg/ml | 0.5 mg/ml | 0.25 mg/ml | 0 mg/ml | |
| *B. thermophilum* | $7.48^{c}$ | $7.56^{d}$ | $7.19^{a}$ | $7.10^{a}$ | $6.85^{a}$ | 0.12 |
| *L. plantarum* | $8.22^{c}$ | $8.35^{d}$ | $8.26^{c}$ | $8.21^{b}$ | $7.97^{a}$ | 0.06 |
| *L. reuteri* | $7.57^{a}$ | $7.47^{a}$ | $7.46^{a}$ | $7.36^{a}$ | $7.19^{a}$ | 0.14 |
| *E. coli* | $9.00^{a}$ | $8.95^{a}$ | $8.89^{a}$ | $8.94^{a}$ | $8.90^{a}$ | 0.05 |
| *S.* Typhimurium | $9.06^{a}$ | $9.08^{a}$ | $8.99^{a}$ | $9.06^{a}$ | $9.04^{a}$ | 0.06 |

* Comparisons were performed between the means of the control (0mg/ml) vs. each treatment.
The mean significance level is defined by different lowercase letters (b<0.05, c<0.01, d<0.001).

**Conclusion** The *L. hyperborea* extract increased the beneficial bacterial strains, *L. plantarum* and *B. thermophilum*, with no effects on the pathogenic strains *in vitro*, indicating a prebiotic potential of this compound that merits further research.

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**References**

Murphy, P., Dal Bello, F., O'Doherty, J. V., Arendt, E. K., Sweeney, T., and Coffey, A. 2013. Animal 7, 1079-1087.
Walsh, A. M., Sweeney, T., O'Shea, C. J., Doyle, D. N. and O'Doherty, J. V. 2013. British Journal of Nutrition 110, 1630-1638.

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# Effects of animal breed and plane of nutrition on nutrient digestibility and energy utilisation of ewes fed fresh grass

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**Application** Good quality grass can sustain a high nutrient utilisation efficiency similar to concentrates.

**Introduction** Fresh grass offers the majority of energy and protein requirements for sheep production in the UK and Ireland (Stergiadis *et al*., 2015). However, there is little information available on comparison of nutrient utilisation efficiencies of fresh grass when offered to different breeds of sheep. The objective of the present study was to evaluate if good quality grass could sustain a high nutrient utilisation efficiency similar to concentrates when offered to 2 breeds of ewes.

**Material and methods** A total of 16 ewe lambs at approximately 13 months of age and 61.5±5.3 kg live weight were used in a 2 × 2 factorial design study, with 2 diets (fresh perennial ryegrass vs. fresh perennial ryegrass plus 0.5 kg/d fresh concentrate) × 2 breeds (Highlander vs. Texel). Each treatment had 4 lambs and treatments were balanced within each breed for animal age and live weight. The feeding period started from early May 2015. The animals were individually housed in pens and fed experimental diets for an adaptation phase of 19 days, and then transferred into respiration calorimeter chambers, remaining there for 5 days, with nutrient digestibility and $CH_4$ emissions measured during the final 4 days. Grass was cut daily in the morning from a single zero-grazing sward which was managed in simulation of grazing condition. Grass was offered *ad libitum* in the morning (10.00am), with the concentrate portion given at the same time for the treatment receiving concentrate (Zhao *et al*., 2015). Water was freely available during the period of study. Data were analysed using GenStat (16th edition) as a 2 × 2 factorial arrangement using the Analysis of Variance.

**Results** The effects of diet and breed on feed intake, nutrient digestibility and energy utilisation are summarised in Table 1. There were no significant interaction effects between diet and breed on any variable evaluated. Grass contained, on average, 197 g/kg DM, and (g/kg DM) 69 ash, 150 crude protein, 459 neutral detergent fibre (NDF), 236 acid detergent fibre (ADF), 215 water soluble carbohydrates (WSC) and 36 lipid, and 13.2 predicted ME (MJ/kg DM). Ewes supplemented 0.5 kg/d fresh concentrates had a significantly lower grass DM intake (P = 0.021) and a trend for a higher total DM intake (P = 0.059). However, the concentrate supplement had no significant effects on any variable of nutrient digestibility or energy utilisation. The Highlander breed had a marginally higher grass DM intake and total DM intake, however, all variables of nutrient digestibility or energy utilisation were similar between the 2 breeds.

**Table 1** Effects of diets and breeds on feed intake, digestibility and energy utilisation of ewes (n= 16)

| | Diet | | | | Breed | | | |
|---|---|---|---|---|---|---|---|---|
| | Grass | Grass+ Conc. | s.e. | P | Highlander | Texel | s.e. | P |
| DM intake (kg/d) | | | | | | | | |
| Grass intake | 1.62 | 1.37 | 0.068 | 0.021 | 1.56 | 1.43 | 0.065 | 0.173 |
| Total intake | 1.62 | 1.81 | 0.068 | 0.059 | 1.78 | 1.65 | 0.065 | 0.173 |
| Nutrient digestibility (g/kg DM) | | | | | | | | |
| DM | 817 | 821 | 9.1 | 0.664 | 822 | 816 | 8.6 | 0.672 |
| OM | 833 | 838 | 9.0 | 0.575 | 838 | 833 | 8.5 | 0.670 |
| Digestible OM in DM | 776 | 779 | 8.2 | 0.750 | 781 | 775 | 7.7 | 0.625 |
| Nitrogen | 703 | 693 | 28.4 | 0.672 | 715 | 681 | 26.9 | 0.404 |
| ADF | 808 | 795 | 11.9 | 0.700 | 806 | 797 | 11.3 | 0.603 |
| NDF | 785 | 770 | 14.0 | 0.700 | 780 | 775 | 13.2 | 0.781 |
| Energy utilisation (KJ/MJ) | | | | | | | | |
| DE/GE | 801 | 804 | 10.7 | 0.691 | 804 | 801 | 10.1 | 0.789 |
| ME/GE | 718 | 720 | 9.6 | 0.476 | 721 | 717 | 9.1 | 0.780 |
| ME/DE | 897 | 895 | 4.3 | 0.429 | 896 | 896 | 4.0 | 0.913 |

**Conclusion** The present study demonstrated that neither concentrate supplementation nor ewe breed had a significant effect on nutrient digestibility or energy utilisation efficiency, although concentrate supplementation significantly reduced grass DM intake. These results indicate that sheep can utilise good quality grass as effectively as that including concentrates in the diet and thus improving grazing grass quality is the key to increase the nutrient utilisation efficiency of sheep production.

**References**

Stergiadis, S., M. Allen, X.J. Chen and T. Yan. 2015. Journal of Dairy Science 98, 3257-3273.
Zhao, Y. G., A. Aubry, N. E. O'Connell, R. Annett, and T. Yan. 2015. Journal of Animal Science 93, 5764–5773.

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# Enhancing the eating quality of concentrate fed lambs

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**Application** Lamb produced from concentrates can have a similar chemical composition and shelf life characteristics to that produced from grass.

**Introduction** In the UK, consumers prefer grass to concentrate finished lamb. However, a proportion is finished of concentrates. Grass finishing is associated with higher welfare, a shorter supply chain and higher eating quality (Fisher *et al.,* 2000). Dietary factors such as polyunsaturated fatty acids (PUFAs) and vitamin E are known to influence the shelf life and eating quality of lamb, with grass finished lamb being associated with a higher PUFA and vitamin E content, (Wood, 2005). Within meat, vitamin E acts as an anti-oxidant, reducing PUFA oxidation, colour deterioration and drip loss (Macit *et al*., 2003). The objective was to investigate dietary strategies to produce concentrate finished lamb with a similar chemical composition and shelf life characteristics to grass finished lamb.

**Materials and methods** Forty Suffolk x Texel ewe lambs with a mean live weight (LW) of 29.0 kg were allocated by LW to either grazed grass (G) or one of three concentrate diets formulated to reflect either a barley based concentrate (B), or alternatives based on dried grass (DG) or sugar beet pulp (SB). All concentrates were formulated to provide similar levels of metabolisable energy, ether extract (45 g/kg DM) and had a similar effective rumen degradable protein/fermentable metabolisable energy ratio. Concentrate B contained 60 mg/kg vitamin E and 18 g/kg Megalac$^{(TM)}$, whereas, concentrates DG and SB contained 250 mg/kg vitamin E and 15 or 27 g/kg linseed oil, respectively. Lambs offered diet G were grazed on a mixed sward consisting of predominately perennial ryegrass, whereas lambs offered diets B, DG and SB were housed individually on sawdust and offered concentrates as course mixes *ad-libitum*. Animal performance was monitored weekly, and lambs selected for slaughter once they reached 40 kg, prior to processing as described Brown and Williams (1979). The *longissimus dorsi* muscle was dissected out from the right side and analysed for fatty acids by gas chromatography. The left leg was removed and cut into three 1.5 cm thick leg steaks containing both *semimembranosus* and pelvic limb muscles. These steaks were vacuum packed and conditioned at 2-4 °C for six days prior to simulated retail display (2-4 °C, 700 lux (16 h on 8 h off) using modified atmosphere packaging (0.75 $O_2$ and 0.25 $CO_2$) for a further 7 days, and measurement of colour stability and lipid oxidation (malonaldehyde, TBARS). The right leg was removed, and *semimembranosus* muscle analysed for vitamin E content by HPLC. The data were analysed by ANOVA as a randomised block design using Genstat 17.

**Results** Lambs offered diets B, DG and SB had a higher (P<0.001) daily gain, slaughter weight and cold carcase weight than those offered diet G. Following 7 days simulated retail display, lambs offered diets G and B tended (P=0.081) to have a higher a* (Redness) than those offered diets DG or SB. Lambs offered diets DG or SB had a higher (P<0.001) muscle C18:3n-3 and vitamin E content than those offered B. There were no differences between treatments in lipid oxidation.

**Table 1** Effect of dietary treatments on performance, muscle C18:3 *n-3*, vitamin E content, colour stability and lipid oxidation (following 7 days of simulated retail display).

| | G | B | DG | SB | SED | P |
|---|---|---|---|---|---|---|
| Slaughter weight (kg) | 38.1$^{b}$ | 42.4$^{a}$ | 41.2$^{a}$ | 41.9$^{a}$ | 0.57 | <0.001 |
| Daily gain (kg/day) | 0.06$^{b}$ | 0.38$^{a}$ | 0.33$^{a}$ | 0.37$^{a}$ | 0.03 | <0.001 |
| Cold carcase weight (kg) | 16.42$^{b}$ | 20.35$^{a}$ | 19.26$^{a}$ | 20.39$^{a}$ | 0.52 | <0.001 |
| C18:3n-3 (g/kg muscle) | 0.19$^{b}$ | 0.12$^{b}$ | 0.53$^{a}$ | 0.52$^{a}$ | 0.02 | <0.001 |
| Vitamin E (µg/g muscle) | 2.61$^{a}$ | 1.88$^{b}$ | 2.38$^{ab}$ | 2.36$^{ab}$ | 0.26 | 0.046 |
| L* (Lightness) | 44.8$^{ab}$ | 44.7$^{ab}$ | 43.3$^{b}$ | 45.3$^{a}$ | 0.65 | 0.027 |
| a* (Redness) | 15.8 | 15.7 | 14.6 | 14.4 | 0.67 | 0.081 |
| b* (Yellowness) | 10.5$^{b}$ | 9.46$^{a}$ | 9.54$^{a}$ | 9.47$^{a}$ | 0.24 | 0.001 |
| TBARS (mg/kg muscle) | 4.30 | 4.25 | 4.07 | 3.47 | 0.79 | 0.700 |

**Conclusion** Lamb performance on diet G was lower than expected. Lambs finished of concentrates containing linseed oil and higher vitamin E, had similar muscle C18:3 *n-3*, vitamin E and shelf life to those finished off grazed grass.

**References**

Brown, A.J. and Williams, D.R. 1979. Agricultural Research Council. Bristol.
Fisher, A.V., Enser, M., Richardson, R.I., Wood, J.D., Nute, G.R., Kurt, E., Sinclair, L.A., and Wilkinson, R.G. 2000. Meat Science 55, 141-147.
Macit, M., Aksakal, V., Emsen, E., Esenbuğa, N., and Aksu, M. 2003. Meat science 64(1), 1-6.
Wood, J.D. 2005. Encyclopaedia of Animal Science. Marcel Dekker p 558-570.

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# Identification, comparison and validation of robust rumen microbial biomarkers for methane emissions using diverse *Bos Taurus* breeds and basal diets

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**Application** Identification of robust biomarkers for methane ($CH_4$) emissions associated with rumen microbial functional genes will help to predict $CH_4$ emissions across diverse breeds and diets and to implement a breeding strategy.

**Introduction** Recent studies have highlighted the exciting opportunity that volatile fatty acids (VFA) concentrations (Negussie *et al.,* 2017) and rumen microbial biomarkers of methane emissions could enable breeding of cattle which emit less $CH_4$ and ultimately may lower agricultural greenhouse gas emissions (Roehe *et al.,* 2016). The aim of this study was the comparison of these biomarkers to identify those highly correlated with $CH_4$ emissions across diverse breeds and diets.

**Material and methods** The data in this study were obtained from samples collected in 3 independent experiments balanced for breed type (Aberdeen Angus, Charolais, Limousin and Luing) and diet, and selected for whole metagenome sequencing. In total, 50 animals were evaluated offered forage or concentrate diets (Rooke *et al.,* 2014; Duthie *et al.,* 2016, 2017). Post-mortem rumen digesta samples were used and total DNA was extracted prior to metagenomics analysis. We used the same protocols applied in Roehe *et al.* (2016) that identified functional microbial genes based on the KEGG genes database. For 16S rRNA gene analysis, the genomic reads were aligned to the Kraken database (Wood and Salzberg, 2014). Methane emissions were measured individually during 48 h in respiration chambers (Rooke *et al.* 2014). A combination of network analysis using Miru (Kajeka, UK) and Partial least squares analysis (PLS, Version 9.1 for Windows, SAS Institute Inc., Cary, NC, USA) was used to identify the most correlated microbial populations (genus level) or microbial genes associated with $CH_4$ emissions (Variable Importance in Projection, VIP).

**Results** In this study, the PLS results comparing several potential biomarkers for $CH_4$ emissions identified 37 factors (including 22 individual genes) with a VIP>0.80 and explained 42% of the variation in residual $CH_4$. Main factors are summarized in Table 1. Other parameters like the Acetate-to-Propionate ratio were not identified as potential biomarker.

**Table 1** Partial least squares analysis identifying the main biomarkers for $CH_4$ emissions within the microbiome.

| Factor[1] | VIP | Coefficient[2] | Function/microbial gene |
|---|---|---|---|
| *Methanotorris* | 1.69 | 0.14 | Hydrogenotrophic methanogen |
| *Methanobrevibacter* | 1.37 | 0.12 | Hydrogenotrophic methanogen |
| *Methanohalophilus* | 1.09 | -0.09 | Methylotrophic methanogen |
| *Faecalitalea* | 0.99 | -0.09 | Amino acid metabolism |
| A:B | 0.88 | -0.01 | Archaea:Bacteria ratio |
| K00672 | 1.32 | -0.05 | formylmethanofuran-tetrahydromethanopterin N-formyltransferase |
| K00581 | 1.08 | -0.02 | tetrahydromethanopterin S-methyltransferase subunit E |
| K00150 | 1.06 | -0.02 | glyceraldehyde-3-phosphate dehydrogenase (NAD(P)) |
| K01959 | 1.02 | -0.02 | pyruvate carboxylase subunit A |
| K00123 | 0.92 | 0.01 | formate dehydrogenase, alpha subunit |
| K00400 | 0.89 | 0.01 | methyl coenzyme M reductase system, component A2 |

[1] Microbial genus, A:B ratio, or KEGG gene. [2] Coefficient of correlation.

**Conclusion** This study confirms the interest of targeting functional microbial genes as robust biomarkers. Most of the identified microbial genes were directly involved in the hydrogenotrophic methane synthesis pathway while methylotrophic methanogens showed less importance in explaining $CH_4$ emissions. In addition, most of the genes identified as biomarkers grouped in the same cluster within a functional genes network and this result was reproduced over three independent trials.

**Acknowledgements** This project was funded by BBSRC and, the Scottish Government with aligned funding of AHDB, Defra and the UK Agricultural Greenhouse Gas Inventory Research Platform.

**References**

Duthie, C-A., Haskell, M., Hyslop, J. J., Waterhouse, A., Wallace, R. J., Roehe, R. and Rooke, J. A. 2017. Animal 13, 1-8.
Duthie, C-A, Rooke, JA, Troy, S, Hyslop, JJ, Ross, DW, Waterhouse, A and Roehe, R. 2016. Animal 10**,** 786-795.
Negussie, E., De Haas, Y., Dehareng, F., Dewhurst, R. J., Dijkstra, J., Gengler, N., Morgavi, D. P., Soyeurt, H., Van Gastelen, S., Yan, T., and Biscarini, F. 2017. Journal of Dairy Science 100**,** 2433-2453.
Roehe, R., Dewhurst, R. J., Duthie, C-A., Rooke, J. A., McKain, N., Ross, D. W. Hyslop, J. J., Waterhouse, A., Freeman, T. C., Watson, M. and Wallace, R. J. 2016. PLoS Genetics 12: e1005846.
Rooke, J. A., Wallace, R. J., Duthie, C-A., McKain, N., de Souza, S. M., Hyslop, J. J., Ross, D. W., Waterhouse, A. and Roehe, R. 2014. British Journal of Nutrition 112, 398–407.
Wood, D. E. and Salzberg, S. L. 2014. Genome Biology Evolution 15**,** R46.

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# Defining the inflammatory gene signature that precedes the development of uterine disease in *postpartum* dairy cows

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**Application** This study contributes to our understanding of the pathogenesis of uterine disease and may aid the identification of prognostic biomarkers to improve disease outcomes.

**Introduction** Following calving, inflammation of the uterus is common during the first week postpartum. However, whilst the majority of cows resolve this inflammation a significant proportion fail to do so. Our previous work identified transcriptomic differences between cattle that resolve inflammation and those that develop uterine disease as early as 7 days postpartum (DPP). Here, we hypothesise that excessive expression of endometrial inflammatory markers contributes to development of uterine disease.

**Material and methods** Endometrial epithelial cells were collected using cytobrushes at 7 and 21 days postpartum (DPP) from 147 mixed parity Holstein Friesian cattle on a commercial dairy farm. Cows were classified as healthy or with uterine disease (purulent vaginal discharge; PVD and cytological endometritis; CYTO) on the basis of vaginal mucus score (Williams *et al*. 2005) and >18% polymorphonuclear cell infiltrate into the endometrium (Kasimanickam *et al*. 2004) at 21 DPP. Day 21 postpartum was chosen for disease diagnosis in line with previous studies that have found a negative correlation between uterine disease and subsequent reproductive performance (Dubuc *et al*. 2010).

**Results** Of the cattle sampled, 78% had high uterine inflammation (>18% PMN) at 7 DPP. By 21 DPP, 28% had developed CYTO and 29% had both CYTO and PVD. RNA-seq (n=30 cows) identified clear differences in transcriptomic profiles of these cells at 7 DPP between cows that subsequently resolve inflammation and those which develop uterine disease. Differential expression of 376 genes was observed between healthy and CYTO cows, and 1654 between healthy and CYTO + PVD cows (P<0.05). Pathway over-representation analysis of genes differentially expressed between healthy and diseased cows identified significant changes in immune-related pathways. The cytokine-cytokine receptor interaction pathway, the NOD-like receptor signalling pathway and the Toll-like receptor signalling pathway were up-regulated in cattle with both CYTO and CYTO+PVD, suggesting that there is a core inflammatory signature present early postpartum that is associated with the onset of uterine disease, including up-regulation of *IL1A, IL1B, NLRP3, IL17, IL8, TNF, TLR2* and *TLR4* (Figure 1).

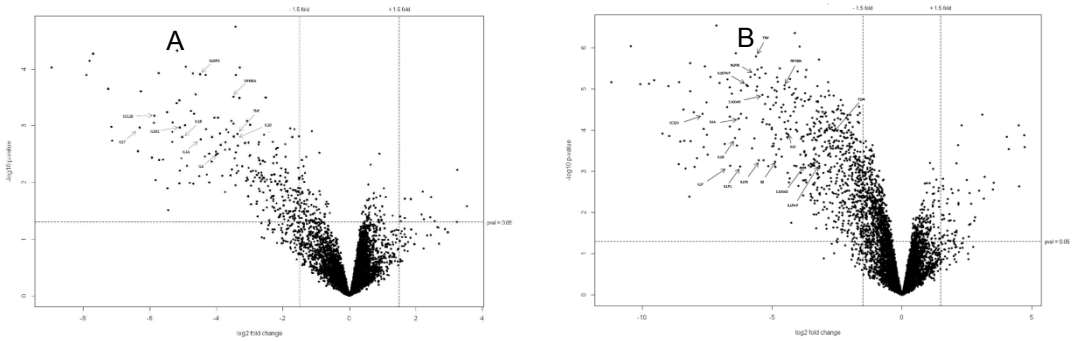


**Figure 1** The majority of differentially expressed genes were upregulated in cattle that developed uterine disease. Gene expression data are presented as volcano plots for each comparison (A: healthy vs. CYTO; B: healthy vs. CYTO+PVD), using log values of the fold change and P value. Each point represents a single gene, with those that survived the cut off values of P<0.05 and FC>1.5 deemed significantly differentially expressed.

**Conclusion** Identification of an inflammatory signature associated with the onset of uterine disease has contributed to our understanding of the pathogenesis of uterine disease and may aid the identification of prognostic biomarkers to improve disease outcomes.

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**References**

Dubuc J, Duffield T F, Leslie K E, Walton J S and LeBlanc S J. 2010. Journal of Dairy Science 93 (11), 5225-5233.
Kasimanickam R, Duffield T F, Foster R A, Gartley C J, Leslie K E *et al*. 2004. Theriogenology 62 (1-2), 9-23.
Williams E J, Fischer D P, Pfeiffer D U, England G C, Noakes *et al*. 2005. Theriogenology 63 (1), 102-17.

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# Impaired locomotion in purebred Jersey and Holstein-Friesian cows, associations with activity measures and hoof surface temperature

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**Application** Activity and hoof surface temperature were associated with locomotion score in Holstein-Friesian cows. No such effects were observed in a sample of purebred Jersey cows.

**Introduction** Studies of automated locomotion impairment (lameness) detection have generally successfully been able to identify non-lame (0) and moderately and severely lame cows (2-3) (Alsaaod *et al*., 2015). Though possible with specialised equipment such a video analysis, the detection of slightly lame cows (1) has proven more challenging. Studies using activity sensors have generally used a coarser measure of lame / non-lame, often excluding slightly lame cows. Slightly lame cow detection currently requires a highly skilled observer to detect. Automated detection of slightly lame animals would thus allow earlier intervention in many cases.
This study assesses associations between locomotion score (0-2 only - excluding severely lame cows) and two different precision technologies. Hoof temperature from thermal imaging of hooves and activity measures from an accelerometer sensor are considered. In addition, how applicable approaches developed in one breed are on another breed is explored. This is assessed by contrasting associations found in groups of Holstein-Friesian and purebred Jersey cows.

**Material and methods** Cow locomotion score was ascertained by one trained human observer using a 0-3 scale (Archer *et al*., 2010). The leg worn RumiWatch pedometer / accelerometer (Converter V.0.7.4.5, Itin+Hoch GmbH, Liestal, Switzerland) was the activity sensor used. The pedometer trial design consisted of selecting approximately equal numbers of cows scoring 0, 1 and 2 to wear the pedometers. The activity the day after locomotion scoring of 19 Jerseys and 15 Holstein-Friesian cows were assessed (technical faults caused the disparity in sample size). Thermal image data were collected using the FLIR T430sc camera (FLIR Systems Inc., Stockholm, Sweden) using methods as described in (Byrne *et al*., 2017). For the thermal imaging, a convenience sample was selected and associations were assessed within the samples of 19 Jerseys cows (as above) and 76 Holstein-Friesian cows.

**Results** The accelerometer derived Activity Index (AI) was strongly correlated with impaired locomotion among a group of 15 Holstein-Friesian cows (Spearman's rho -0.8, p=0.001). AI is defined as 'The averaged variance of 3-dimensional acceleration in 10-s segments' (Alsaaod *et al*., 2015). However, for AI, no association with locomotion score could be discerned for a group of 19 purebred Jersey cows. Using thermal imaging of 76 Holstein-Friesian cows, the average hoof temperature of all 4 hooves was positively correlated with the locomotion score ($P<0.05$). However, no consistent association with locomotion score was observed with hoof temperature within the 19 Jersey cows.

**Conclusion** Thermal image data of 19 purebred Jersey cow' hooves showed no consistent association with locomotion score. In contrast, a clear association was found among 76 Holstein-Friesian cows for all four hooves. As only hind feet were identified as problem feet in the 76 Holstein-Friesian group by the scorer, this indicates a thermal effect in all non-lame hooves. AI as measured by a leg worn accelerometer was found to be strongly associated with lameness in Holstein-Friesian cows but not Jersey purebred cows. Both the thermal and activity findings indicate distinct breed specific effects. These could be attributable to breed differences in physiology, lameness related behaviour or different lameness pathology prevalence rates in the studied groups.
These findings may have implications for the application of automated locomotion measurement, and potentially, for Precision Livestock Technology in general. The challenges of using sensors technology to detect lameness are well documented (Van Nuffel *et al*., 2015). The findings of this study indicate this may be further complicated by breed effects. When farmers are considering investing in a Precision Livestock Technology and they have multiple breeds of cattle or minority breeds, they may thus benefit from considering which breeds were used to develop and calibrate the technology. In future, breed effects should be considered in the validation of Precision Livestock Technologies, in particular for those assessing locomotion.

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**References**

Alsaaod, M. Niederhauser, J.J. Beer, G. Zehner, N. Schuepbach-Regula, G. *et al* 2015. Journal of Dairy Science 98(9), 6236–6242.
Archer, S., Bell, N. and Huxley, J. 2010. In Practice. 32(10), 492–504.
Byrne, D. Berry, D. Esmonde, H and McHugh, N. 2017. Journal of Animal Science. 95(2), 970–979.
Van Nuffel, A. Zwertvaegher, I. Pluym, L. Van Weyenberg, S. Thorup, V. *et al*. 2015. Animals 5(3), 861–885.

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# A model to predict age specific calving rate from age at first calving and calving interval in cattle

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**Application** Assessment of methods to translate age at first calving and calving intervals for a breed from experimental studies into annual calving rates for assessment of breeding strategies.

**Introduction** Prediction of age specific calving rates, the probability of an animal having a calf within a certain age bracket, is important for modelling the biological and economic performance of herds comprised of different breeds and crosses. However, these data are often not available whereas age at first calving (AFC) and calving interval (CI) are much more commonly reported, particularly in Sub-Saharan Africa. The objective was to use AFC and CI means and standard deviations as inputs to predict age specific calving rates. Two variations of a deterministic model and one simulation are described and tested using parameters from Ethiopia and Ireland. Predicted calving rates are compared across models and systems.

**Material and methods** In model A, AFC mean and standard deviation were used to define a normal distribution for age at first calving. The probability of the first calving occurring within each of 20 sequential annual age classes was calculated. For the first age class, mean CI was added to the midpoint age in the class, producing a mean age at second calving if the first calving occurred within the first age class. CI standard deviation was used to produce a normal distribution around this new mean and the probability of second calving occurring within each age class and first calving occurring within the first age class was calculated. This was repeated for each age class of first calving. The second calving probabilities were summed across first calving age classes (weighted by the derived proportions in each first calving age), giving the total probability of second calving occurring within each age class. These steps for second calving were repeated for 3$^{rd}$-20$^{th}$ calvings. Probability was summed across parities, giving age class calving rates. A problem with model A is that the mid age in a class is rarely equal to the mean age of calving in that class. To reduce error, model B reduced the length of each age class to 0.1 years. The results were then summed within the original 20 age classes. A simulation was used to test these two models. A data set of 100,000 individuals with values of AFC and CI was simulated so that mean and standard deviation of AFC and CI were equal to the input parameters. Calving dates were then calculated for each individual. The number of calvings occurring within each of the 20 age classes was divided by the total number of individuals to calculate age class calving rates. This was repeated 10 times and an average across replicates was taken. Parameters from two varying systems were used to test the models and simulation: farms in Oromiya Region in Ethiopia, where mean AFC=3.89(σ=0.61) years and mean CI=1.54(σ=0.43) years (Ayalew *et al.* 2004) and Ireland, where mean AFC=2.20(σ=0.37) years and mean CI=1.02(σ=0.09) years (Evans *et al.* 2006).

**Results** Both models give very similar results in both case studies (Figure 1). However, the results of the simulation with the Ethiopian parameters show on average higher calving rates than either deterministic model. In Ethiopia, the large means of AFC and CI as well as the high standard deviations lead to low yearly calving rates. In Ireland, the CI was close to a year and had much smaller variation and this led to a much higher yearly calving rate, close to 100%.

**Conclusion** The similarities between models A and B suggest the error introduced by using the mid-point rather than the mean age of calving in a class is not important in the tested scenarios. Calving rate in Ethiopia predicted by the simulation was greater than that predicted by models A and B due to an individual in the simulation having a consistent calving interval over life, whereas the models assume no correlation between calving intervals within an individual. Both scenarios are interesting as in reality calving intervals across life will show some, but not complete, correlation.

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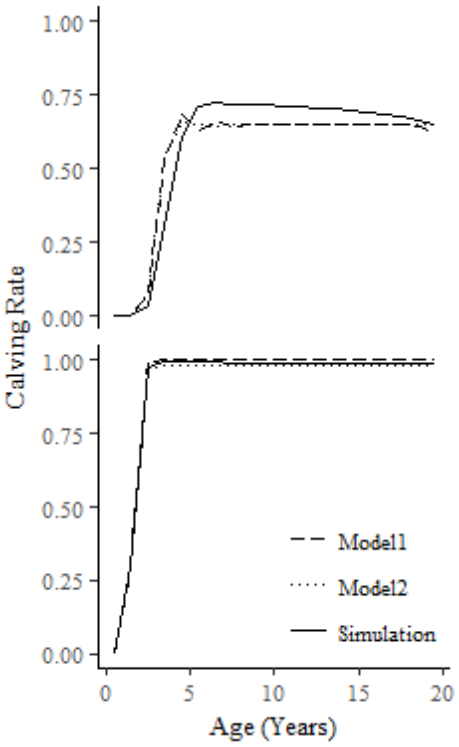


**Figure 1** Predicted calving rates by age in Ethiopia and Ireland

**References**

Ayalew, W., van Dorland, A. and Rowlands, J. (eds) 2004. Design, execution and analysis of the livestock breed survey in Oromiya Regional State, Ethiopia. Oromiya Agricultural Development Bureau, Addis Ababa, Ethiopia.
Evans, R.D., Wallace, M., Garrick, D.J., Dillon, P., Berry, D.P. and Olori, V. 2006. Livestock Science 100, 216-230.

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# Comparison of utilisation performance of perennial ryegrass (*Lolium perenne L.*) cultivars under grazing conditions

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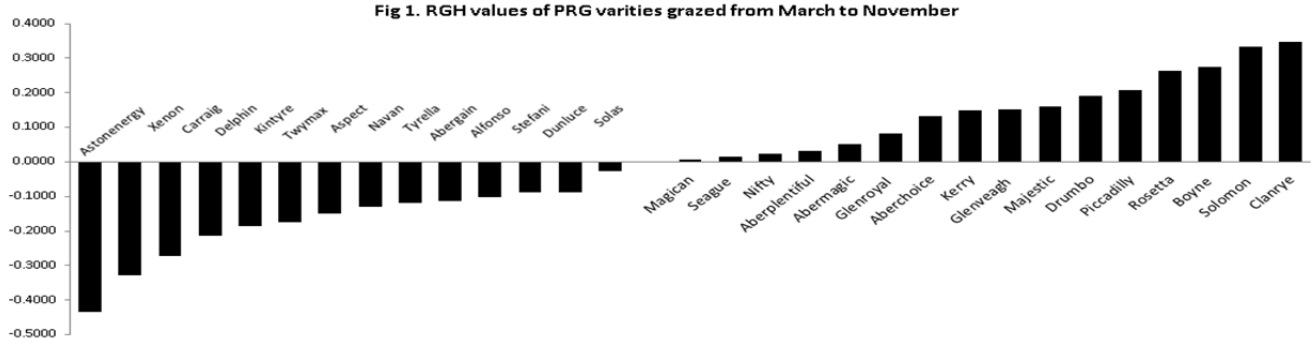
**Application** Cultivars of perennial ryegrass differ in their level of grazing utilisation with post-grazing heights of cultivars ranging from 3.8cm to 4.8cm.

**Introduction** Increased utilisation of pasture increases profitability of dairy cattle systems by reducing the requirement for concentrate supplementation. Differences in perennial ryegrass (PRG; *Lolium perenne* L.) traits such as DM yield, persistence and quality have been quantified. Such analysis has allowed superior cultivars of perennial ryegrass (PRG) to be selected. The suitability of cultivars to intensive grazing based production systems is another trait of interest to farmers as it affects overall pasture utilisation and therefore profit. The objective of this study was to examine utilisation performance of varieties by comparing post-grazing heights of individual varieties after grazing by dairy cows.

**Material and methods** In October 2016, a paddock (0.7 ha) subdivided into 90 plots (8x4.5m) was sown with cultivars of PRG from the thirty leading varieties from the Irish Department of Agriculture, Food and the Marine recommended list, under a randomised block design. Fifteen diploid varieties of intermediate and late heading types were used: Aberchoice, Abermagic, Boyne, Clanrye, Drumbo, Glenroyal, Glenveagh, Kerry, Majestic, Nifty, Piccadilly, Rosetta, Solomon, Stefani and Tyrella. Fifteen tetraploid cultivars: Abergain, Aberplentiful, Alfonso, Aspect, Astonenergy, Carraig, Delphin, Dunluce, Kintyre, Magician, Navan, Seagoe, Solas, Twymax and Xenon were also sown. The plots were rotationally grazed by dairy cows from March to October with eleven grazings achieved. The numbers of cow allocated varied throughout the season but on average, 60 cows grazed the paddock. Cows grazed the paddock in 3 sections (30 plots/section). Cows were moved on to the next section when the majority of plots were grazed to a height of 4cm or less. Residency time was never longer than 6 hours in each section. Prior to grazing a 1.2x6m section of each plot was harvested with an Etesia mower to a height of 3.5cm. Mown herbage was weighed and 0.1 kg of grass was dried at 90˚C to determine DM content. A Jenquip rising plate meter was used to measure grass height prior to grazing and to measure post-grazing height of the plots. The data were analysed using the GLM procedure of SAS, with block, grazing event and variety used as variables in the model.

**Results** Variety affected post grazing height (P<0.001). There was a 1cm difference between the lowest and highest mean height recorded; Astonenergy (3.7cm) had the lowest post-grazing height and Clanrye (4.8cm) had the highest. Pre-grazing height and mass are positively correlated with post-grazing height. To adequately account for varieties achieving greater pre-grazing heights, the post-grazing height of each variety was predicted with a linear model, taking account of the pre- and post-grazing heights of all the varieties. Residual Grazed Height (RGH) is calculated as the difference between achieved post-grazing height and predicted post-grazing height. Varieties achieving lower RGH (i.e. those have greater negative values) were assumed to have greater utilisation than expected during grazing. From these analyses Astonenergy had the greatest grazing utilisation performance (RGH=-0.43) and Clanrye had the worst (RGH=+0.35).

Fig 1. RGH values of PRG varities grazed from March to November




**Conclusion** Differences in utilisation efficiency exist between varieties. This is a key trait of importance for farmers and affects the ease of grazing management of pastures. Both farmers and plant breeders are conscious of the benefits of identifying varieties with superior grazing characteristics. This study shows that there is significant variation amongst varieties of PRG for the post-grazing height trait thus facilitating genetic selection of varieties with superior grazing capacity. It is expected that such varieties will have better utilisation performance whilst maintaining high DM production.

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# Characterisation of best linear unbiased estimates generated from national genetic evaluations of reproductive performance, survival, and milk production in dairy cows

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**Application** Best Linear Unbiased Estimates (BLUEs) provide an indication of dairy herd phenotypic performance after accounting for differences in animal-level environmental and genetic characteristics.

**Introduction** Animal performance is a function of both the genotype of the animal and the environment it is (and was) exposed to. Failure to improve management in line with the requirements of the genetically elite germplasm hampers the actual realisation of the benefit in genetic gain. The aim of the present study was to describe herd-level factors contributing to herd-year BLUEs and understand the influence certain herd-level factors have on dairy cow fertility, milk, and survival.

**Material and methods** Contemporary group solutions from the national Irish dairy cow fertility genetic evaluations on 3,445,557 cows were used. Traits included age at first calving, calving to first service interval from parity 1 to 3 as separate traits, number of services from parity 1 to 3 as separate traits, calving interval from parity 1 to 5 as separate traits, survival from parity 1 to 5 as separate traits, and 305-day milk yield from parity 1 to 5 as separate traits. Contemporary group BLUEs were collated into herd-year BLUE effects. Only data from spring calving herds that had ≥30 calving events for each year in the 10-year period from 2007 to 2016 inclusive were retained (5,177 herds). Herd-year characteristics of interest included: 1) geographical location, 2) whether the herd was milk recording, 3) herd size, 4) herd expansion rate, 5) herd-level use of artificial insemination, 6) proportion of cows in the herd that were born in that herd, 7) the proportion of cows in the herd registered with a breed society, and 8) the proportion of cows in a herd that calved in the first 42 days of the calving season. A single herd-year BLUE for each trait averaged across parities was calculated. The associations between herd-level characteristics and the six BLUE traits were quantified using linear mixed multiple regression models where the dependent variable was the herd-year BLUE.

**Results** Correlations between the same trait in different parities were all weak to moderate (0.15 to 0.48) with the exception of milk yield, which was strongly correlated between parities (0.77 to 0.87). Within parity correlations between the herd-year BLUEs for each trait were weak to moderate and ranged from -0.15 to 0.39. Mean herd-year BLUE for age at first calving reduced by over 10 days from the year 2007 to 2014; mean BLUE for calving to first service interval increased by 2.8 days. Mean herd-year BLUE for milk yield increased by over 100 kg from the year 2007 to 2015. The repeatability across years of mean herd-year BLUEs for all traits ranged from 0.14 (survival) to 0.72 (milk yield). All herd-level characteristics were associated with the herd-year BLUEs. Mean BLUE for calving interval lengthened as herd size became larger, expansion rate increased, a larger proportion of dairy cows were purchased, and an increased number of animals were registered with a breed society (Figure 1); mean BLUE for calving interval also lengthened as the proportion of cows calved within the first 42 days of the calving season reduced as well as the herd-level use of artificial insemination reduced (Figure 1).

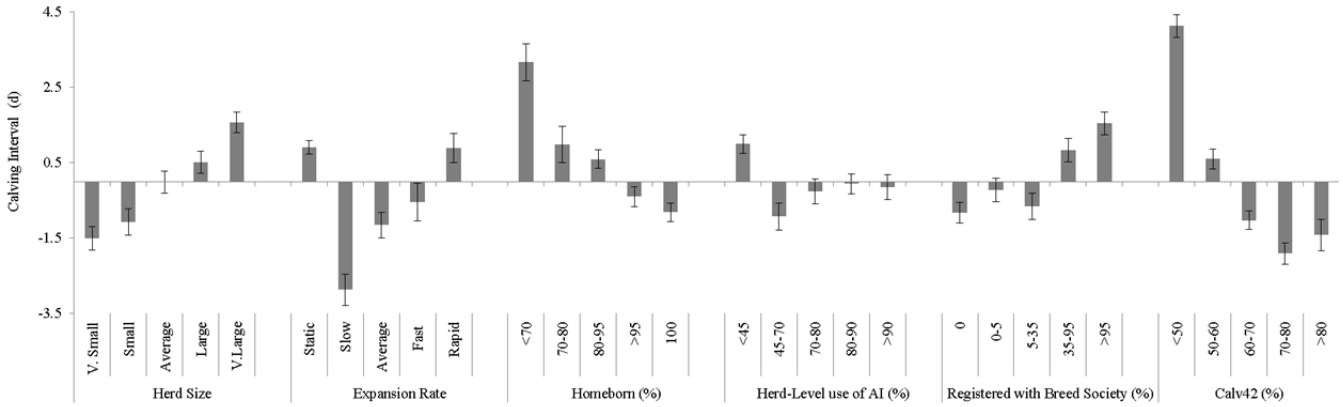


**Figure 1** Mean best linear unbiased estimate for calving interval for each category of six herd-level characteristics.

**Conclusion** All herd-level characteristics considered in the present study were associated with variability in herd-year BLUEs for fertility, milk yield and survival; such information can be incorporated into decision support tools.

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# The effect of a 14 day immune supplement on the performance of newborn dairy calves

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**Application** Prevention of scouring in early life is important in reducing later ill health and performance effects. The inclusion of a health supplement in the milk fed to calves at birth can improve dry matter intake to support growth and development.

**Introduction** Enteric infections in the early days of calf life are responsible for approximately 40% of calf mortality rates and contribute to the ongoing burden of disease affecting calf development. Inclusion of measures to prevent ill health can mitigate these risks supported by effective nutritional provision and environmental management. The first priority is to ensure successful immunity is established through high quality colostrum feeding which provides protective antibodies to the calf from the mother. Additional protection may be provided through feed supplements. Trouw Nutrition have developed a unique supplemental product which supports gut immunity and health as it contains antibodies against the major pathogens *Cryptosporidium parvum, Rotavirus, Coronavirus, E. coli* (K88, K99, 987P, and F41P), *Clostridium perfringens* (type A, C and D) and *Campylobacter fetus-jejuni.* This study seeks to investigate the effects of including this supplement in the calf's first feeds by assessment of calf performance and health through to weaning.

**Material and methods** 64 Holstein Friesian dairy calves were assigned to experimental groups a) treatment or b) placebo at birth. Calves were weighed at birth and allocations were balanced for live weight and sex. Within 6 hours of birth, calves received 3 L of their mother's colostrum via an oesophageal feed tube. The colostrum was pre-mixed with 30 g of supplemental powder consisting of a) gut health additive or b) non-treated powder. Subsequent feeds included 10 g of supplemental powder mixed into colostrum for the first three days, followed by 10 g powder added to milk replacer up to 14 days of age. Calves received 2 L colostrum twice each day before moving onto a higher provision of milk replacer at 10-15% BW allowance. This was prepared fresh at 150 g/L dilution over two equal feeds per day. Water was provided *ad libitum* alongside a custom blend of starter concentrate and a bucket of chopped straw. The experimental treatment was terminated on day 15 and calves were monitored up to 70 days of age recording levels of intake, growth and health. Milk replacer intake was measured at each feed up to 63 days and individual intake of starter and water was recorded up to 69 days. Calves were weighed on a weekly basis from 7-70 days. Faecal scoring (FS) was conducted daily and in the case of calf scouring (FS>2) electrolytes were provided with administration of antibiotics as per veterinary instruction. All episodes of calf ill health were monitored and recorded on a daily basis.

**Results**

**Table 1** Feed intake measured through the study

| | | A | B | SE | Sig.(*p*) |
|---|---|---|---|---|---|
| **Average Daily Intake** | | | | | |
| Milk replacer (gDM/day) | d3-63 | 788.7 | 790.3 | 11.6 | NS |
| Concentrate (gDM/day) | d3-70 | 570.5 | 515.7 | 27.0 | 0.005 |
| DMI (gDM/day) | d3-63 | 1168 | 1120 | 16.5 | 0.001 |
| Water (ml/day) | d3-70 | 1760 | 1450 | 60.5 | 0.001 |
| **Cumulative Intake (d3-70)** | | | | | |
| Milk replacer (kgDM) | | 48.5 | 47.9 | 0.17 | NS |
| Concentrate (kgDM) | | 34.5 | 30.9 | 0.64 | 0.025 |
| DMI (kgDM) | | 83.0 | 78.9 | 0.65 | 0.034 |
| Total Water (L) | | 80.3 | 65.7 | 2.52 | 0.002 |

Calves receiving the health supplement (Group A) demonstrated increased intake of concentrate and subsequently had significantly greater average dry matter intake than those calves on the placebo (Group B). This was coincided with a significant increase in water uptake by group A calves, although no significant difference in milk replacer intake was recorded. A significant ($p<0.05$) difference in live weight was noticed during weaning with average weight 73.6 kg and 69.5 kg measured on day 56 for group A or B calves respectively. However, this difference was not considered to be significant by day 70 with average daily gain of 0.67 kg/day in group A comparable to 0.63 kg/day by group B. There was no significant difference in the occurrence of scour or pneumonia episodes between the two experimental groups, yet a slight increased number of animals on the placebo powder were noted to incur scouring effects (44%) compared to those receiving treatment (37%).

**Conclusion**
The inclusion of health supplements in the milk feeding of dairy calves from birth encourages increased concentrate and water uptake to promote efficient growth.

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# *Dichelobacter nodosus* metapopulations and the epidemiology of footrot in an endemically infected flock

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**Application** Widely endemic in the UK, footrot can lead to severe lameness, loss of condition and reduced productivity thus having a great economic impact and costs around £24 million per annum. With this in mind, understanding the pathogenesis of footrot and the involvement of *Dichelobacter nodosus* in its progression is a key topic in the sheep industry and will help improve knowledge towards suitable vaccine development and refine the strategies for combating this disease within flocks.

**Introduction** Footrot, primarily caused by *Dichelobacter nodosus*, is responsible for approximately 70% of foot lesions in sheep in the UK. There are two clinical presentations of footrot: interdigital dermatitis (ID), an inflammation of the skin between the digits, and severe footrot (SFR) where hoof horn separates from the underlying sensitive tissue. Several virulence factors have been identified in *D. nodosus* however, the presence of virulence factors does not correlate with the presence of severe disease. *D. nodosus* exists in a community of multiple strains (metapopulation) and one hypothesis is that disease severity is dependent on several strains co-existing to cause disease, rather than one 'virulent strain'.

**Material and methods** A flock of 99 ewes from one farm were assigned either parenteral and topical antibiotics (AIS) or foot trim and topical antibiotics (FTS) if a footrot treatment was required (locomotion score >2, scoring from 0 – 4). The interdigital skin of feet of 50 of these ewes was swabbed and footrot disease status recorded on a minimum of 16 occasions over 10 months (separate score for ID and SFR both from 0 – 4). This project is using foot swabs from two feet from 25 of the 50 ewes. DNA was extracted, and the number of *D. nodosus* cells (load) quantified by quantitative PCR, from 845 foot swabs. All samples were also inoculated onto culture plates as part of a previous study from which 93 isolates were cultured from 47 (6%) samples. Multiple regression and 2-way ANOVA were used to test for statistical significance of results below.

**Results** *D. nodosus* was detected in 68% of samples. Key new results are that *D. nodosus* load significantly increased with ID score ($p < 0.01$) but there was no relationship between *D. nodosus* load and SFR score (**Table 1**). *D. nodosus* load decreased significantly one week after treatment with either AIS or FTS ($p < 0.05$). There was no significant change in *D. nodosus* load for samples from when only parenteral antibiotics was given for another health reason ($p > 0.05$). This indicates that AIS and FTS were effective in reducing the external load of *D. nodosus*, possibly due to the action of the topical antibiotics. The load of *D. nodosus* was significantly higher in samples which did have isolate(s) cultured ($p < 0.01$), indicating that isolates do not come from a random sample of skin swabs.

**Table 1** Change in the number of *Dichelobacter nodosus* cells (load) plus standard error with ID and SFR score.

| ID Score | No. | No. *D. nodosus* +ve samples (%) | Mean $\log_{10}$(load)+1 of *D. nodosus* +ve samples, SFR +ve & -ve ± SE | Mean $\log_{10}$(load)+1 of *D. nodosus* +ve samples, SFR +ve only ± SE |
|---|---|---|---|---|
| 0 | 521 | 315 (60.5) | 3.89 ± 0.06 | 4.14 ± 0.15 |
| 1 | 168 | 118 (70.2) | 4.17 ± 0.09 | 4.05 ± 0.26 |
| 2 | 61 | 56 (91.8) | 4.59 ± 0.11 | 4.63 ± 0.21 |
| 3 | 38 | 35 (92.1) | 4.70 ± 0.17 | 4.67 ± 0.26 |
| 4 | 57 | 54 (94.7) | 4.97 ± 0.12 | 4.69 ± 0.25 |

**Conclusion** The results suggest *D. nodosus* is driving the progression of ID. Topical antibiotics may be key to reducing the load, and therefore spread, of footrot-causing bacteria. The use of quantitative PCR to quantify *D. nodosus* load was more representative than that of culture. Next steps are to analyse the *D. nodosus* serogroups and strain community profile.

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**References**

Smith, E. M., Green, O. D. J., Calvo-Bado, L. A., Witcomb, L. A., Grogono-Thomas, R., Russell, C. L., Brown, J. C., Medley, G. F., Kilbride, A. L., Wellington, E. M. H. and Green, L. E. 2014. The Veterinary Journal 201, 295-301.

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# Long-term effect of removing prophylactic antibiotics from weaner diets on performance and health indicators of finisher pigs

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**Application** Removal of antibiotics (AB) from weaner feed resulted in a 95% reduction in AB usage. Despite an initial reduction in performance, pigs that did not receive in-feed AB had similar performance at the finisher stage to pigs receiving in-feed AB.

**Introduction** Mis/over-use of AB contributes to antibiotic resistance posing a risk to both human and animal health. Potential restrictions on prophylactic AB use in pig feed is placing greater focus on alternative strategies such as targeted parenteral administration of AB and vaccinations combined with management and housing improvements. The aim of this study was to quantify the effects of removing prophylactic AB from the diets of weaner pigs on their health and performance at the finisher stage on a commercial farm with a high level of AB usage.

**Material and methods** The study had ethical approval from the Teagasc Animal Ethics Committee (TAEC 40/2013). It was carried out on a 300 sow commercial farrow-to-finish farm positive for PRRSv, APP, *M. hyopneumoniae* and influenza. In-feed prophylactic AB (Sulfadiazine-Trimethoprim, Pfizer Ltd., 14.4mg/kg BW/d; for 5 d/wk) was used during the nine week weaning stage to address clinical problems. Six batches of 140 pigs each were weaned, tagged and allocated over a six-week period into two groups where in-feed AB were removed from the diet of one group (NOAB) and were maintained in the other group (AB) [weaning weight (NOAB 9.2±0.61;AB 9.2±0.62 Kg)]. Pigs were managed as per usual farm practice and they were weighed on transfer between each production stage. Feed intake was recorded daily as well as all mortalities and all parenteral administrations of AB (which were administered in both treatment groups as required). Average daily gain (ADG), average daily feed intake (ADFI) and feed conversion ratio (FCR) were calculated. Pigs were individually scored for tail lesions prior to slaughter. Production data were analysed in SAS v9.3 by general linear models (PROC GLM) while mortality rate and number of injections were analysed using a Chi-square test (PROC FREQ). Tail lesions were analysed by linear mixed models (PROC MIXED). Pearson correlations (PROC CORR) were calculated between initial BW, mortality rate and number of injections during the whole weaner stage as well as between final BW and 'pig days' (no. days to reach slaughter).

**Results** AB pigs showed higher ADFI and ADG than NOAB pigs in the first weaner stage indicating that even when used at therapeutic levels, antibiotics have growth promoting effects (Table 1). However, FCR did not differ between the two groups at any stage. Removal of in-feed AB increased the number of parenteral treatments (NOAB 25% vs. AB 13.8% of the animals treated; P<0.001) during the entire weaner stage while there was no difference in finisher pigs (NOAB 34% vs. AB 32%; P=0.406). While the mortality rate was not different at weaning (2.1%vs.1.9%; P=0.806) it tended to be higher in NOAB pigs during the finisher stage (3.1%vs.1.3%; P=0.099). BW at weaning was correlated with the percentage of parenteral treatments (r=60.7; P=0.036) and tended to be correlated with mortality (r=56.3; P=0.056) while at the finisher stage initial BW was correlated with the no. of days that pigs took to reach slaughter (r=62.7; P<0.001).

**Table 4** Production data (ADG, ADFI, FCR, BW) for pigs provided with in-feed antibiotics (AB) and for pigs with no in-feed antibiotics (NOAB) during the first and the second weaner stages and the finisher stage

| | First weaner stage | | | Second weaner stage | | | Finisher stage | | |
|---|---|---|---|---|---|---|---|---|---|
| | NOAB | AB | *P* | NOAB | AB | *P* | NOAB | AB | *P* |
| ADGg | 402.2±18.2 | 435.6±13.0 | 0.018 | 711.0±32.3 | 743.7±42.6 | 0.774 | 865.4±29.3 | 882.2±29.3 | 0.893 |
| ADFIg | 584.6±39.8 | 646.5±28.8 | 0.048 | 1380.9±29.3 | 1440.2±60.1 | 0.589 | 1811.1±31.1 | 1818.8±36.9 | 0.984 |
| FCR | 1.48±0.03 | 1.52±0.03 | 0.483 | 1.95±0.05 | 1.95±0.04 | 0.944 | 2.10±0.04 | 2.07±0.04 | 0.853 |
| BWKg | 21.9±0.85 | 23.0±0.70 | 0.032 | 41.4±1.36 | 43.3±1.4 | 0.218 | 99.4±1.5 | 101.4±1.9 | 0.483 |

Abbreviations: ADG-average daily gain; ADFI-average daily feed intake; BW-body weight; FCR-feed conversion ratio. Data are presented as mean±SEM (standard error of the mean).

**Conclusion** Despite the initial negative effects of the NOAB treatment on ADG, ADFI and parenteral treatments, differences were no longer significant at finishing and pigs were as efficient as AB pigs at all stages of production. However, the tendency towards higher mortality in NOAB than AB pigs may indicate better protection against disease over time. The relationships found in the study reflect the strong link between BW at weaning and susceptibility to disease.

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# Johne's Disease - A novel social network analysis perspective

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**Application** The identification of high risk herds using social network analysis (SNA) can reduce the effort required to control Johne's disease (*JD)* and provide an evidence-based approach to the development of risk-based surveillance activities and disease prevention programs.

**Introduction** Traditionally in Ireland, the approach to *JD* was based on passive surveillance of clinical cases with non-responsive chronic diarrhoea, largely by submission of a faecal sample to the Veterinary Laboratories Service (VLS-DAFM) and subsequent control by voluntary removal of the affected animal. In most cases, the infection was largely well established in the herd by the time *Mycobacterium avium paratuberculosis* (*MAP*) was detected. In recent years, a different and more proactive approach aimed at estimating disease risk and earlier detection of infection has been adopted by many farmers (repeated serology testing). The movement and trade of live animals from farm to farm, plays a primary role in the introduction and spread of infectious agents, especially *MAP*. Social Network Analysis (SNA) provides a methodology for the analysis and illustration of the relationship between the movements of animals and the transmission of a contagious pathogen associated with those movements. Hence the objectives of this study were to: 1) describe the demographics of confirmed *JD* positive (*JD* + ) animals in Ireland, 2) to construct and characterise the connections made between herds by the movements of *JD* + animals through different premises (*JD* network), 3) to increase our understanding of the latent spatio-temporal herd-to-herd transmission of the disease and 4) to provide a disease surveillance framework for the identification of those herds (nodes) more likely to facilitate the spread of infection.

**Material and methods** Laboratory data covering 11 years and consisting of faecal mycobacterial culture results of animals with persistent non-responding diarrhoea were organised in an adjacency matrix formed by a collection of nodes (herds) and an array of directed arcs (movements) linking the nodes *(JD* network). Once the database was tidied, selected datasets were extracted and graphed, and then the relationships between herds were explored and analysed by their connections (movements). Three programs for large network analysis and visualization were used: *UCINET, PAJEK* and *R*. A network is a collection of units of interest or nodes (herds, animals,etc.) that may or may not be connected (arcs/edges) to each other through a relationship of some sort (movement, lineage, etc.). A movement event was defined as the change from an identified premise of origin (Herd No.) to other identified premise of destination (Herd No., factory, knackery, etc.). Since each animal movement implied a certain direction, the graph produced was a directed network. The dataset contains 1220 confirmed (faecal culture) JD+ animals and 1089 herds (nodes) and a total 1413 movements, including movements to factories, knackeries and 728 movements excluding them. Since infection with MAP normally happen early in life, a herd in which an *JD* + animal is born (herd of origin) was classified as 'source herd'. Quantification of how many 'source herds' are connected directly or indirectly between, within each component, represents a measure of disease transmission. An assumption was made that premises sharing JD+ animals are potentially capable of spreading JD. Descriptive statistics of the network and nodes were calculated (infection chain, centrality indicators, geodesic distances, density, etc.). Inferential statistics were attended by comparing the JD network with random network model (Erdos-Renyi random network) with similar network characteristics.

**Results** The network was fragmented into 415 components (sub-networks where nodes are connected within, but do not have ties with, other sub-networks) ranging from 1 to 50 nodes. Out of the 258 components with two or more nodes, 59 had two or more connected 'source herds' nodes, representing 23% of these subset and 14% of the total number of components. In addition, herds with high ingoing contact chain and in-degree showed strong association (asymmetric Rajski 0.63 and 0.60 respectively) with those source herds. Preliminary results show a significant association between source herds and disease transmission by the movement of JD+ animals. I was also found that 179 JD+ animals (110 of beef breed and 69 dairy; 163 females and 16 males) were connected by lineage.

**Conclusion** SNA analysis provided a practical approach to assess disease transmission and identify those premises more likely to facilitate the spread of JD, thus providing a framework for the development of a risk-based Johne's surveillance program.

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**References**

De Nooy, W., Mrvar, A., Batagelj, V. 2005. Exploratory Social Network Analysis with Pajek, Cambridge University Press, New York.

# Untangling the pangenomes of the *Butyrivibrio* group

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**Application** Exploring the phylogeny of the *Butyrivibrio* group will provide a basis for further understanding the complex interactions of the rumen microbiota. This is imperative to fully understand the consortium of microbial enzymes that are responsible for the catalysis of multifaceted reactions, such as biohydrogenation.

**Introduction** The *Butyrivibrio* group is composed of the most active bacterial species involved in the biohydrogenation of C18 unsaturated fatty acids (UFA); this includes the genera *Butyrivibrio* and *Pseudobutyrivibrio*. The former genus was proposed based on the discovery of a group of conformationally similar rumen bacteria which were capable of producing large amounts of butyric acid via glucose fermentation. At the time, it was noted that the inherent variability resulting from classification based on metabolic profile could lead to difficulties in defining other species-specific patterns. This approach to classification has become antiquated with the development of sequencing technologies and downstream bioinformatics analysis. This study aims to investigate the taxonomic relatedness and functional capacity of the ruminal *Butyrivibrio* group using the 73 genomes.

**Material and methods** Seventy one geneomes were already available in JGI (Hungate 1000) or Genbank and the genomes of two additional bacterial strains (*B. proteoclasticus* JK669 and *P. xylanivorans* MZ8), obtained through the Rowett Research Institute, were completed by ourselves. DNA extraction was performed using a FastDNA™ SPIN Kit for Soil (MP Biomedicals) and quantified using an Epoch BioTek (US) microplate spectrometer with Gen5-Take3 software on a Take3 plate. After submission to MicrobesNG, the samples were subject to sequencing by the Illumina HiSeq 2500 platform, using 2x250bp paired-end reads and with x30 coverage. All 73 genomes were annotated using Prokka V1.12 via the Galaxy platform with a similarity E-value cut-off of 1e-06. The 16S rDNA were then aligned using the Aligner Pipeline of the Ribosomal Database Project (RDP), Release 11, Update: September 30, 2016. The alignment was used to create a phylogenetic tree using FastTree V2.1.10 with default parameters. An additional tree was constructed using 40 gene markers as per Wu and Eisen (2008). The resulting tree files were uploaded to the Interactive Tree Of Life (iTOL), changelog version 3.5.2. *Clostridium beijerinckii* NCIMB 8052 and *Lactobacillus acidophilus* NCFM were used as outliers, and both trees were rooted by the latter.

**Figure 1** Phylogenetic comparison of the Butyrivibrio group based on 16S rDNA comparions

**Results** Phylogenetic analysis of 16S rDNA (Figure 1) and 40 marker sequences revealed three distinct clades within the *Butyrivibrio* group. As expected, *Butyrivibrio* and *Pseudobutyrivibrio* appear to form independent clades, with the exception of *B. fibrisolvens* strains, which form the third. The exception to this is the placement of *B. proteoclasticus* JK669 and *Butyrivibrio* sp. NC2002, which both fell within the *Pseudobutyrivibrio* clade on the 16S tree. The latter fell within its expected clade (grouped with *Butyrivibrio*) in the 40 marker tree.

**Conclusion** 16S rDNA and 40 marker taxonomic analysis highlights the extensive variation within the *Butyrivibrio* group, in particular between *B. fibrisolvens* and the rest of the group. These results emphasise the need for further research into the *Butyrivibrio* group. Future research will focus on pangenomic analysis, conducting whole genome analyses in order to determine a set of core and accessory genes (and therefore core and accessory functions) across the same 73 genomes.

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**References**

Wu, M. and Eisen, J. 2008. A simple, fast, and accurate method of phylogenomic inference. Genome Biology 9(10), p.R151.

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# Evaluating the field-level performance of grazing livestock systems based on high-resolution primary data

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**Application** The proposed framework quantifies the economic performance of individual fields that collectively constitute rotational grazing, providing useful information for commercial farmers to improve their animal management strategies.

**Introduction** With increasing concern for global warming and food security, the importance of improving livestock production efficiency has never been greater (Eisler *et al*., 2014). In order to compare system-wide performances of multiple farming strategies, various forms of metrics, such as nutrient use efficiency, carbon footprint per unit of final product and annualised net farm income, have been developed to date (Klapwijk *et al*., 2014; McAuliffe *et al*., 2017 and 2018). These metrics, however, are typically designed to produce a single score that represents the entire area of landholding, and therefore unable to account for spatial heterogeneity across different fields on the farm. Using high-resolution primary data from the North Wyke Farm Platform (NWFP), an intensively instrumented farm-scale livestock research facility in Devon, UK, this study proposes a novel framework to evaluate the field-by-field performance of grazing systems and biophysical mechanisms causing its variability.

**Material and methods** The NWFP (50°46'10" N, 3°54'05" W) comprises three hydrologically independent small-scale farms locally known as "farmlets". Each farmlet has ~22 ha of grassland divided into seven fields, with self-contained infrastructure including a silage clamp, a manure midden and a cattle housing facility. Each farmlet carries (a) 30 Charolais x Hereford-Friesian calves; and (b) 50 Suffolk x Mule ewes and their lambs sired by Charollais rams, both of which are randomly allocated across the three farmlets at the beginning of each season and then rotated approximately every 2-3 weeks within the assigned farmlet. Over three grazing seasons (2011, 2012 and 2013), field-level performances of animals were evaluated by the means of the animal liveweight gain attributable to each field. The gain attained while grazing on a particular field was derived based on fortnightly weighing of all animals, with values on non-weighing days imputed under the assumption of a constant daily gain between two weighing events. The gain attained while being housed was assigned to the field that produced the silage fed to animals. As the exact origin of silage fed on a given day could not be identified, the entire liveweight gain achieved by each individual animal while indoors was apportioned across fields according to their contributions to silage production. Finally, the total liveweight gain attributable to each field was computed as the sum of gains from grazing and from silage, across all animals allocated to the farmlet, and converted to the per hectare value to eliminate the effect of field size. Using the dataset thus prepared ($n$ = 21 with three repeated years), multivariate regression analysis was conducted to identify primary determinants of field-level animal productivity, with a particular focus on soil parameters that are easily measurable in the commercial environment.

**Results** Across the farmlets and years, the average field-level performance was quantified to be 463.9 (± 175.1) kg ha$^{-1}$ year$^{-1}$. Neither the farmlet nor year had a statistically significant ($p < 0.10$) effect on this value. The results of regression analysis suggested that an increase in the soil organic carbon (SOC) stock, amongst other covariates, led to a greater liveweight gain from that field ($p = 0.02$), while intra-field heterogeneity in SOC was shown to be detrimental to animal production ($p < 0.01$). Furthermore, a larger SOC stock was also associated with a lower level of water discharge from the corresponding field and, consequently, smaller losses of soil inorganic nitrogen.

**Conclusion** As the vast majority of commercial enterprises around the world adopt rotational grazing systems, data from field-scale randomised controlled trials are unlikely to reveal the complete mechanistic processes linking soil, pasture, animals and, ultimately, the whole-farm income. The proposed framework based on field-scale data from a farm-scale trial offers a potential solution to this issue, while at the same time contributing to the derivation of economically and environmentally efficient animal rotation strategies. Forthcoming work undertaken under this approach is expected to assist the development of scientifically robust and easy-to-measure metrics for agricultural sustainability, which can support sustainable decision making by commercial livestock producers.

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**References**

Eisler, M.C., Lee, M.R.F., Tarlton, J.F., Martin, G.B., Beddington, J. *et al*. 2014. Nature 507, 32-34.
Klapwijk, C.J., van Wijk, M.T., Rosenstock, T.S., van Asten, P.J.A., Thornton, P.K., Giller, K.E. 2014. Current Opinion in Environmental Sustainability 6, 110-115.
McAuliffe, G.A., Takahashi, T., Harris, P., Orr, R.J., Lee, M.R.F. 2018. Journal of Cleaner Production 171, 1672-1680.
McAuliffe, G.A., Takahashi, T., Mogensen, L., Hermansen, J.E., Sage, C.L., Chapman, D.V., Lee, M.R.F. 2017. Journal of Cleaner Production 165, 1163-1173.

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# Efficacy of ivermectin and fenbendazole on Irish cattle farms

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**Application** A study on the efficiency of ivermectin and fenbendazole was carried out on 16 Irish cattle farms and found that on 12 of the 16 farms the reduction in the faecal egg counts of the calves was less than 95% for both drugs. On 4 of the 16 farms there was a failure of ivermectin but the population of gut worms was susceptible to fenbendazole.

**Introduction** In grass-based production systems such as Ireland, control of gastrointestinal nematode infection is dependent on the availability of efficacious anthelmintic products. A direct and unavoidable consequence of continuous use of anthelmintics is the development of drug resistant nematodes. There are currently three classes of broad spectrum anthelmintics available for the control of gastrointestinal nematodes in cattle, benzimidazoles (BZ), levamisoles (LV) and macrocyclic lactones (ML). Anthelmintic resistance (AR) is a major issue for ruminant livestock production worldwide. Since the 1960's cases of AR have been reported on sheep farms all over the world. In more recent years, AR has become an issue on cattle farms in New Zealand, Argentina and the UK with resistance to macrocyclic lactones (ML) and benzimidazoles (BZ) being reported (Kaplan and Vidyashankar, 2012). While anthelmintic resistance has been confirmed on two beef research farms in Ireland (O'Shaughnessey *et al*., 2014), the extent of anthelmintic resistance on a wider scale is unknown. The aim of the current study was to investigate the prevalence of resistance to benzimidazole and macrocyclic lactone products on Irish cattle farms.

**Material and methods** Two anthelmintic products from different chemical families were used in this study, ivermectin (ML) and fenbendazole (BZ). Sixteen farms, geographically spread over the Republic of Ireland, took part in the study in 2017. Farms required a minimum of 40, first grazing season calves in order to take part in the study. Herd level faecal egg count (FEC) was monitored fortnightly from the 1st of May 2017. Farmers facilitated this by collecting fifteen fresh faecal samples from their group of calves and sending them to the lab in Teagasc. A composite faecal sample was generated by combining 5 g of faeces from each sample. This composite sample was then used to carry out the FEC. FEC was carried out using the mini FLOTAC with a sensitivity of 5 eggs per gram (epg) (Coles *et al*., 2014). Once herd level FEC reached at least 100 epg, Teagasc staff visited the farm. Twenty calves from the grazing group were selected at random and faecal samples taken directly from their rectum. These calves were then weighed, marked and treated with ivermectin subcutaneously at a rate of 1 ml per 50 kg bodyweight (Ivomec, Merial Animal Health). A second group of 20 calves were again selected at random, faecal samples taken *per rectum* and the calves weighed, marked and treated with oral fenbendazole at a rate of 7.5 ml per 100 kg bodyweight (Panacur, Intervet Ireland Limited). The calves continued to co-graze for the duration of the study. Fourteen days post anthelmintic treatment the farm was re-visited and faecal samples collected *per rectum* from all calves. In order to determine anthelmintic treatment efficacy a Faecal Egg Count Reduction Test (FECRT) (Coles *et al*., 2006) was carried out.

**Results** On all 16 farms there was evidence of resistance to ivermectin as treatment failed to reduce the FEC by >95% on any farm. For 15 farms reductions in FEC varied between 3% and 89% however, on 1 farm faecal egg count increased between the first and second sampling after treatment with ivermectin. On 12 farms there was also evidence for fenbendazole resistance with FEC reductions varying between 15 % and 93 %. On 4 farms fenbendazole was effective, resulting in reductions in egg counts >95%.

**Conclusion** The FECRT showed that ivermectin and fenbendazole failed to reduce the FEC of the calves by > 95% on 16 and 12 farms respectively, suggesting reduced efficacy of both anthelmintic products on these farms. On only 4 farms were nematode populations shown to be susceptible to fenbendazole. These results suggest that there is widespread failure of both macrocyclic lactone and benzimidazole products in the effective control of nematode populations on Irish cattle farms.

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**References**

Coles, G.C., Bosco, A., Cringoli, G., Maurelli, M.P., Musella, V., Rinaldi, L. 2014. Acta parasitology 59(4), 625-628
Kaplan, R.M., Vidyashankar, A.N. 2012. Veterinary Parasitology 186, 70-78
Coles, G.C., Jackson, F., Pomroy, W.E., Pichard, R.K., Von Samson-Himmelstjenna, G., Silverstre, A., Taylor, M.A., Vercruysse, J. 2006. Veterinary Parasitology 136, 167-185
O'Shaughnessey, J., Earley, B., Mee, J.F., Doherty, M.L., Crosson, P., Prendiville, R., Macreilli, M., deWaal, T. 2014. Veterinary Record 175, Issue 5

# Ewe breed differences in cervicovaginal mucus properties at a natural and synchronised oestrus

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**Application** This study provides novel insights into factors influencing cervical sperm transport which would facilitate the practical use of cervical artificial insemination for sheep using frozen-thawed semen.

**Introduction** The only effective method for artificial insemination in sheep using frozen-thawed semen consists of a laparoscopic procedure, but this requires veterinary expertise and is welfare unfriendly. The alternative transvaginal method involves depositing semen at the cervical opening, as it is not possible to transverse the sheep cervix due to its complex anatomy. However, internationally this method yields low pregnancy rates. The only exception to this is in Norway, in which vaginal deposition of frozen-thawed semen to a natural oestrus yields good pregnancy rates (>60%). Research in Ireland has demonstrated this is due to the ewe breed (Donovan *et al*. 2004), since sperm can transverse the cervix in greater numbers in some breeds (Belclare) than in other breeds (Suffolk). Therefore, the aim of this study was to characterise the differences in the cervical secretions in Belclare and Suffolk ewes during the follicular and luteal phases of both a synchronised and natural cycle.

**Material and methods** Cervicovaginal mucus was collected from Suffolk and Belclare ewes (n=30 per breed) during the breeding season at the follicular (Day 0) and luteal (Day 10) phases of a synchronised (14 day progestagen sponge + 400 IU equine chorionic gonadotropin at sponge removal) and a natural cycle (ewes were checked for oestrus using a vasectomised ram). Mucus from each animal, at each time point, was assessed for weight (g), colour and viscosity after aspiration. The colour was measured via a scoring system from 1 to 7 (1: clear, 2: clear-cloudy, 3: cloudy, 4: cloudy-milky, 5: milky, 6: milky-creamy, 7: creamy). For viscosity, a small drop of mucus was placed into the Leja chamber (IMV Technologies, L'Aigle, France) and the time (sec) to fill this chamber was recorded (A maximum of 420 sec was allowed for mucus to fill the chamber). Following completion, the ewes were resynchronised and the experiment was repeated. Analysis of mucus weight, colour and viscosity data was carried out using repeated measures ANOVA in SPSS (version 24.0, Armonk, NY).

**Results** Luteal mucus was less abundant, cloudier in colour and more viscous than follicular phase mucus ($P<0.05$). There was a significant effect of ewe breed on mucus viscosity but not on mucus colour or weight. There was a ewe breed by stage of cycle interaction for mucus viscosity which was represented by Belclare ewes having more viscous mucus than Suffolk ewes at the follicular phase of both the synchronised and natural oestrus ($P<0.05$; Figure 1). There was no effect of synchronisation on mucus weight, viscosity or colour.

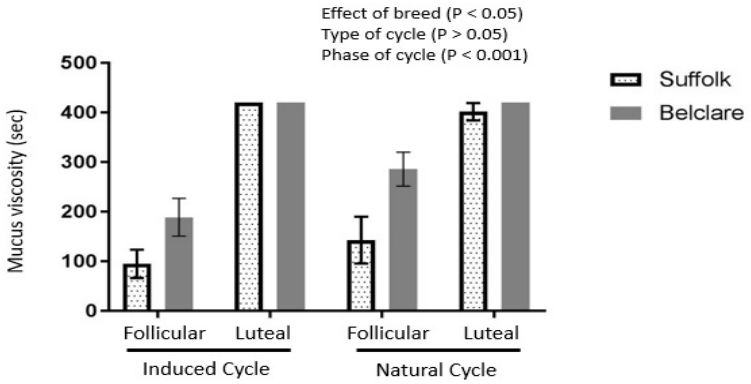


**Figure 1** Mean mucus viscosity from Suffolk and Belclare ewes at the follicular and luteal phases of a synchronised and natural cycle. Means are reported as ± standard error of the mean (SEM) of two replicates.

**Conclusion** This study demonstrates that the amount of mucus secreted in Suffolk and Belclare ewes is similar but they differ in their rheological properties. A more in depth analysis of the cervical tissue and its mucus composition is required to understand why sperm transport differs between these two ewe breeds.

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**References**

Donovan, A., Hanrahan, J.P., Kummen, E., Duffy, P. and Boland, M.P. 2004. Animal Reproduction Science 84, 359-368.

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# Identification of transcriptional variants in skeletal muscle tissue of cattle undergoing compensatory growth

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**Application** This study provides an insight into SNPs involved in compensatory growth in cattle. This data following validation could contribute to identifying DNA based biomarkers for the selection of cattle with a greater ability to undergo compensatory growth.

**Introduction** Compensatory growth or catch-up growth is defined as a physiological process by which an animal has the ability to accelerate its growth after a period of under nutrition (Hornick *et al*., 2000). It can be effectively exploited in beef production systems by restricting feed intake during periods such as Winter when feed costs are high and then allow animals to undergo compensatory growth when pasture becomes available in Spring. Currently a number of genes have been identified as contributing to the expression of compensatory growth in cattle (Connor et.al. 2010, Keogh, 2015) and these genes may harbour SNPs which may serve as molecular biomarkers for the selection of cattle with a better ability to undergo compensation. The objective of this study was to assess these genes for transcriptional variants in skeletal muscle tissue. Genes investigated included those involved in growth regulation, lipid and steroid metabolism, ATP binding, transcriptional regulation, phosphoprotein and oxidation reduction processes.

**Material and methods** This study utilised tissue collected as part of the study of Keogh (2015). Thirty Holstein-Friesian bulls were assigned to one of two groups: (i) restricted feed allowance for 125 days (RES; n=15) followed by *ad libitum* access to feed for 55 days or (ii) *ad libitum* access to feed throughout the trial (ADLIB; n=15). The first 125 days was denoted as Period 1 and the subsequent 55 days, Period 2. During Period 1 RES were managed to achieve a target mean daily growth rate of 0.6 kg/day and 1.9 kg/d for ADLIB. *M. longissimus dorsi* biopsy samples were collected from all animals on day 15 of re-alimentation in Period 2. Total RNA was isolated and RNA sequencing performed on an Illumina Hiseq 2000. Sequence reads were checked for quality using FastQC, with low quality reads removed using Trim Galore. Reads were then aligned to the bovine genome (UMD3.1) using STAR. Variant calling was performed as per GATK guidelines (McKenna *et al*., 2010) on 53 genes known to be important to the expression of compensatory growth. Duplicate reads were removed from those that had mapped and remaining reads were subjected to local realignment and base-score recalibration. Variant calling was performed using HaplotypeCaller. Finally, the Ensembl Variant Effect Predictor programme was used for annotation of identified variants.

**Results** The bulls displayed compensatory growth with the RES group growing at 1.8 times of the ADLIB group, with an ADG of 2.5 kg/day apparent during re-alimentation in Period 2. Of the 53 genes selected 411 transcript variants were identified in cattle undergoing compensatory growth versus restricted cattle. 65% of the variants identified were synonymous variants with 33% missense variants and 1% were frameshift variants. Of the variants identified, those within 3 particular genes were of high impact, namely; *ASNS, PRDX6 and ELN. ASNS* is involved in protein and ATP binding and was found to harbour a frameshift variant. Similarly a frameshift variant was also discovered in *PRDX6* which is involved in redox regulation of the cell. This gene may also play a role in the regulation of phospholipid turnover as well as in protection against oxidative injury and was also found to be differentially expressed in a previous hepatic based study (Connor *et al*., 2010). Finally a splice acceptor variant was identified in the *ELN* gene which is involved in the regulation of proliferation and organization of vascular smooth muscle and was previously identified as a variant in the 1000 bull project (identified SNP rs42069643).

**Conclusion** This study provides an insight into the molecular mechanisms regulating the compensatory growth phenomenon in cattle. Transcriptional variants identified in the current study may, following validation, hold potential for use as DNA based biomarkers for the selection of cattle with a superior ability to undergo compensatory growth following a period of dietary restriction.

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**References**

Connor, E.E., Kahl, S., Elsasser, T.H., Parker, J.S., Van Tassell, C.P., Baldwin, R.L., and Barao, S.M. 2010. Functional & Integrative Genomics 10(1), 39-51.
Hornick, J.L., Van Eenaeme, C., Gerard, O., Dufrasne, I. and Istasse, L. 2000. Domestic Animal Endocrinology 19, 121-132.
Keogh, K., 2015, PhD thesis, UCD, Dublin.
McKenna, A., Hanna, M., Banks, E., Sivachenko, A., Cibulskis, K., Kernytsky, A., Garimella, K. , Altshuler, D., Gabriel S. and Daly, M. 2010. Genome Research 209, 1297-1303.

# Effect of dietary oil type and vitamin $D_3$ level during gestation on sow and litter performance

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**Application** Replacing soya oil with salmon oil in the gestation diet did not improve sow or litter performance. An 800IU/kg dietary inclusion of vitamin $D_3$ improved piglet growth to weaning compared to 2000IU/kg inclusion.

**Introduction** Increasing litter size has been accompanied by an increase in the proportion of small, weak, unviable piglets that often do not survive the first few days of life. It is therefore important to identify strategies to improve piglet viability at birth and survivability to weaning as a solution to improve overall sow output. Previous research suggests that fish oils, rich in omega-3 fatty acids, can improve piglet vitality and vigour at birth, increase piglet growth to weaning and reduce pre-weaning mortality. The current recommended vitamin $D_3$ inclusion rate for gestating and lactating sows is 800IU/kg (NRC, 2012) but levels up to 2000IU/kg are commonly included in UK and Irish gestation diets. Recently, increasing vitamin $D_3$ levels (200-2000IU/kg) in sow gestation diets was found to reduce stillbirths and increase piglet birth and weaning weight (Lauridsen *et al* 2010, Weber *et al* 2014). The objective of this study was to investigate the use of salmon oil and vitamin $D_3$ inclusion levels in sow gestation diets to improve piglet growth and survival to weaning.

**Material and methods** The study was a 2x2 factorial design, involving 120 multiparous sows (30/treatment) and was conducted at AFBI, Hillsborough, Co. Down, between April 2016 and May 2017. Dietary treatments were fed from day 30 of gestation until parturition and contained either 2.5% soya or salmon oil and two inclusion levels, 'high' or 'low', of vitamin $D_3$ (2000 and 800IU/kg, respectively). Sow weight, back-fat depth ($P_2$) and body condition score was recorded at days 28 and day 107 of gestation and at weaning. Gestation and lactation feed intake was also recorded. Litters from sows whose farrowings were attended (n=80) were used for vitality measures. For each piglet (n=1143); time of birth, birth interval, vitality score, sex and birth weight was recorded. Each piglet was marked with their birth order and observed for time to first suckle. Piglets were weighed again at day 1 to allow for colostrum intake to be estimated according to the method of Theil *et al* (2014). Crown to rump length and abdominal circumference were also measured to allow for Ponderal index and body mass index (BMI) to be calculated as per the method of Baxter *et al* (2008). Piglets from all litters (n=1841) were weighed at day 1, day 14 and day 28 (weaning). Data were analysed using Genstat (18$^{th}$ edition). Sow and litter variables were analysed with linear mixed model methodology using REML estimation. Variables that were discrete and had a poisson distribution e.g. total born, born alive, were analysed with generalised linear mixed model methodology. Vitality score was analysed using a multilevel mixed-effects ordered logistic regression. In all analyses, sow and rep were included as random effects; parity and treatment were included as fixed effects with additional fixed effects added in a stepwise manner and those that were significant were retained in the model.

**Results** Average daily gain (ADG) of piglets from day 14 to weaning was significantly greater for those born to sows fed soya oil 800IU/kg vitamin $D_3$ compared to soya oil 2000IU/kg (298.20 vs. 269.50g/day, respectively, $P<0.05$). Sow live-weight, back-fat depth, body condition score, gestation length, feed intake or lactation length did not differ between treatments ($P>0.05$). Sows fed soya oil during gestation ate on average 12.1kg more during lactation than salmon oil fed sows ($P=0.004$), as such soya oil sows tended to be heavier at weaning ($P=0.061$). There was no effect of treatment on total born, born alive, pre-weaning mortality, ADG of the litter or number weaned. Gestation treatment did not affect piglet vitality score. For all litters, piglets from sows fed soya oil had significantly greater ponderal index and BMI ($P<0.001$) compared to piglets from sows fed salmon oil. Vitality scored piglets from sows fed 2000IU/kg during gestation were significantly heavier at birth (1.41 vs. 1.33kg, respectively $P=0.019$) with a tendency to have a greater colostrum intake ($P=0.082$) than piglets from sows fed 800IU/kg. Day 1 weight of piglets was significantly higher when born to sows offered 2000IU/kg ($P<0.05$). Piglets from sows fed 2000IU/kg and had a greater crown-to-rump length ($P<0.001$) than piglets from 800IU/kg fed sow. However piglets from sows fed 800IU/kg were heavier at weaning ($P=0.023$), had a higher ADG from day 14 to weaning ($P=0.007$) and had a greater Ponderal index ($P=0.039$) when compared to piglets from 2000IU/kg fed sows.

**Conclusion** The use of salmon oil in sow gestation diets did not improve piglet vitality at birth or survival and growth to weaning. Although 2000IU/kg vitamin $D_3$ increased piglet birth and day 1 weight, an inclusion level of 800IU/kg vitamin $D_3$ increased piglet growth during the late suckling period and weight at weaning, therefore this study supports the current recommendation of 800IU/kg vitamin $D_3$ for gestating sows.

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**References**

Baxter, E., Jarvis, S., D'Eath, R.B., Ross, D.W., Robson, S.K. *et al*. 2008. Theriogenology 69, 773-783
Laurdisen, C., Halekoh, U., Larsen, T., Jensen, S.K. 2010. Journal of Animal Science 88, 202-213.
NRC. 2012. Nutrient requirements of Swine: Eleventh Revised Edition. The National Academies Press.
Theil, P.K., Flummer, C., Hurley, W., Kirstensen, N.B., Labouriau, R., Sorensen, M.T. 2014. Journal of Animal Science 92, 5507-5519.
Weber, G.M., Witschi, A.K.M., Wenk, C., Martens, H. 2014. Journal of Animal Science 92, 899-909.

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# Using novel animal measurements to predict dry matter intake in lactating dairy cows (*Bos taurus)*

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**Application** Including linear body measurements, grazing behaviour and infrared thermography (IRT) with known energy sinks can increase the ability to predict dry matter intake (DMI) by 5%.

**Introduction** Identifying cows that efficiently convert grazed grass into milk has become increasingly important due to high production costs and increasing global food demand. Selective breeding for efficiency would require DMI estimates for large numbers of animals. Estimating DMI through the n-alkane technique is a time consuming and expensive process, and is impractical to conduct at commercial farm level. The objective of this study was to use novel animal measurements with the aim of enhancing the predictability of DMI over and above that explained by known energy sinks.

**Materials and methods** During 2015, DMI was estimated for 120 Holstein/Friesian cows on three occasions (May, June and September), on the Teagasc Dairygold Research farm, Kilworth, Co. Cork. These cows were also part of a stocking rate (SR) study. DMI was estimated using the n-alkane technique as described by Mayes *et al*. (1986) and modified by Dillon and Stakelum (1989). Milk yields were recorded daily and milk composition weekly. Solids corrected milk (SCM) yield was calculated as described by Tyrell and Reid (1965). Body weight (BW) and BCS were recorded the week before and after DMI. Back length, hip width, rump width, withers height and muzzle circumference were recorded once for each animal between August and September. Heart girth, chest girth and body depth were measured twice, once after the first grazing of a 36 hour allocation (full) and after the last grazing of a 36 hour allocation (empty) between the intake estimation periods. Each cow was linear scored once during lactation, in May, by the Irish Holstein Friesian Association. Three blood pressure measurements were taken within one milking between the second and third DMI measurement. Heart rate was measured for every animal over one 24 hour period using polar heart monitors. Images of the eye, ribs, front and back hoofs were captured from the left side of each animal using an IRT camera after the second intake run. Animals were fitted with Institute of Grassland and Environmental Research behaviour recorders for a 24 hour period between each intake run. In total 48 independent variables had univariate regression models generated using SAS PROC REG to determine their association with DMI. Variables that had a P-value of less than or equal to 0.25 were retained for backward linear regression, where variables with a P-value <0.05 were retained in the model. All models were adjusted for parity, stocking rate treatment, body weight and SCM yield. Model residuals were standardised and normality checks were performed.

**Results** Known energy sinks (SCM, BW) and adjustment variables (parity, SR treatment) predicted DMI with an R-squared of 69.6%. Of the 48 variables of interest, 14 variables progressed to the backward linear regression phase. Grazing bout duration, heart girth (full) and eye temperature ultimately remained after the final phase. Including grazing bout duration, heart girth and average eye temperature increased the R-squared for DMI from 69.6% to 74.3%. Grazing bout duration and heart girth were positively associated with DMI while average eye temperature was negatively associated.

**Table 1** Variables of interest retained by the backward linear modelling phase

| Specific Measurement | Measurement Type | P-value | R-squared |
|---|---|---|---|
| Grazing bout Duration | Grazing Behaviour | 0.0357 | 0.012 |
| Heart Girth (Full) | Linear Measurements | 0.0167 | 0.015 |
| Average Eye Temperature | Thermography | 0.0052 | 0.020 |

**Conclusion** The predictability of DMI increased to 74.3% by including grazing bout duration, heart girth (full) and average eye temperature in addition to SCM, body weight and parity while accounting for SR treatment effects. As technology improves and the use of automation increases on commercial farms, this may become a viable method of estimating DMI. These findings will be validated using an independent dataset.

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**References**

Dillon, P. and Stakelum, G. 1989. Irish Journal of Agricultural Research 8, 104.
Mayes, R. W., Lamb, C. S. and Colgrove, P. M. 1986. The Journal of Agricultural Science 107(1), 161-170.
Tyrrell, H. F. and Reid, J. T. 1965. Journal of Dairy Science 48(9), 1215-1223.

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# The effect of milk cooling rate on milk microbiological quality and composition

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**Application** Pre-cooling milk prior to it entering the bulk tank is effective in rapidly reducing milk temperature and could be useful in reducing bacterial growth during storage, thereby preserving milk quality.

**Introduction** Milk leaves the cow's udder at approximately 35 ˚C, which is a favourable temperature for bacterial growth (Walstra *et al*., 2006). Therefore, milk cooling and refrigerated storage are necessary after milking, in order to reduce bacterial growth rates. An increase in bacterial counts in milk could result in hydrolysis of protein, fat and lactose, thus affecting milk processing characteristics and nutritional value. The pre-cooling of milk using plate heat exchangers (prior to entering the bulk tank) rapidly reduces milk temperature and also could rapidly reduce bacterial growth rates. The aim of this study was to investigate the effect of pre-cooling milk at different rates on the microbiological quality and composition of milk.

**Material and methods** Spring-calving dairy cows (n=210) were milked in a 30-unit side-by-side milking parlour, twice daily over two three-week periods. The volume of milk collected during each milking was distributed equally into three identical bulk tanks (morning: 800 L/ tank; afternoon: 500 L/ tank), using shut-off valves to control the flow rate. Prior to the milk entering the bulk tanks, three pre-cooling treatments were applied: no plate cooler (NP), single-stage (SP) and double-stage (DP) plate cooler; which pre-cooled milk to 32.0 ± 1.4 ˚C, 17.0 ± 2.8 ˚C and 6.0 ± 1.1 ˚C, respectively. In the SP treatment, milk exchanged heat with ground water (at approximately 15 ˚C). In the DP treatment, milk was pre-cooled in two stages: ground and ice-water (at approximately 0 ˚C) were used in the first and second stage, respectively. Milk was added to the bulk tanks twice daily for 72 h and stored at 3 ˚C. Milk line samples were collected to access the microbiological quality of milk entering the tanks. After the initial morning milking, duplicate milk samples were collected from each bulk tank once the milk temperature reached 3 ˚C, corresponding to 0 h samples (one milking). The subsequent samples (24, 48 and 72 h) were collected prior to the addition of milk from the morning milkings of the following days, when the bulk tanks contained milk from 2, 4 and 6 milkings, respectively. The samples were analysed for a range of bacteria, as well as composition and somatic cell count (SCC). The data were analysed using the MIXED procedure in SAS 9.3, with period (1 and 2), week (1, 2 and 3), pre-cooling system (NP, SP and DP) and storage time (0, 24, 48, and 72 h) as fixed effects.

**Results** The average total bacterial count (TBC) for the milk line samples was 3.35 ± 0.29 $\log_{10}$ cfu/ mL, indicating that milk of good microbiological quality was produced. The TBC means at 0 h for NP, SP and DP were 3.55, 3.57 and 3.50 ± 0.09 $\log_{10}$ cfu/ mL, respectively; while the psychrotrophic bacterial count (PBC) means were 3.11, 3.04 and 3.07 ± 0.11 $\log_{10}$ cfu/ mL, respectively. The bulk tanks that received the NP, SP and DP treatments cooled milk to 3 ˚C in 2 h, 1 h and 20 min, respectively. The differences in cooling times were expected to affect the initial bacterial counts within each tank; however, there was no significant difference between treatments (P>0.05). The different pre-cooling treatments also did not affect any of the bacterial counts over the storage period up to 72 h (P>0.05). This could be due to the reduction of the blend temperature within each bulk tank as the volume of milk at 3 ˚C increased after each milking; consequently, the milk was cooled faster. However, after 72 h, numerical differences were observed between the bacterial counts in the milk volumes subjected and not subjected to pre-cooling. The TBC means at 72 h for NP, SP and DP were 3.90, 3.77 and 3.71 ± 0.09 $\log_{10}$ cfu/ mL, respectively; the mean PBC were 3.38, 3.28 and 3.25 ± 0.11 $\log_{10}$ cfu/ mL, respectively. The low bacterial growth rates also indicate that the storage temperature of 3 ˚C was effective in preventing an increase in bacterial numbers over the storage period. The pre-cooling treatments and storage time had no impact on milk composition (P>0.05). The average fat, protein, lactose and total solids were: 4.42 ± 0.08 %, 3.58 ± 0.07 %, 4.78 ± 0.07 % and 13.38 ± 0.13 %, respectively.

**Conclusion** The bacterial count and composition of milk are minimally impacted when the milk was stored at 3 ˚C for 72 h, whether the milk is pre-cooled or not; however, milk entering the tank should have good initial microbiological quality. Considering the numerical differences in bacterial counts between treatments at 72 h, the use of the SP or DP pre-cooling systems is recommended to maintain low levels of bacteria and possibly reduce cooling energy costs.

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**References**

Walstra, P., Wouters, J. T. M. and Geurts, T. J. 2006. 2nd ed. Taylor and Francis Group, LLC.

# Iodine levels in bulk tank milk produced by dairy cows during the mid- and late-lactation periods

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**Application** The consumption of excess iodine by humans is related to thyroid dysfunctions. Bovine milk is one of the main iodine sources for humans and therefore it is relevant to monitor on-farm factors that could contribute to high iodine levels in milk.

**Introduction** Two major factors influencing milk iodine levels are (i) ingestion of iodine through animal feeds and (ii) application of teat disinfectants containing iodine to cows. Incorrect feeding management, such as utilisation of rations with higher levels of iodine than required or unnecessary supplementation, can result in high concentrations of iodine in milk. Borucki Castro *et al.* (2012) reported a linear relationship between dietary iodine content and milk iodine concentration. With regard to the use of iodine teat disinfectants, the incomplete removal of those products from the teat surface prior to cluster attachment can increase the risk of direct transfer of iodine to milk. Iodine may also be absorbed through the teat skin, with consequent effects on milk iodine levels. The British Agricultural Research Council, the German Society of Nutrition & Physiology and the US National Research Council recommend that the daily iodine consumption should be 10 to 12 mg/cow/day. The aim of this study was to investigate the concentration of milk iodine levels on Irish dairy farms during mid- and late-lactation periods and identify the possible sources of this iodine in the milk.

**Material and methods** This study was undertaken on 67 spring-calving dairy farms (herd size: 21 to 469 cows), located in the Kilkenny and Waterford region of Ireland, during the mid- (June) and late- (October) lactation periods. Milk samples were collected from the top inlet of each of 67 bulk tanks using sterilised sample dippers, after appropriate agitation. Iodine was quantified through inductively coupled plasma mass spectroscopy, using an Agilent ICPMS 7700x system. Questionnaires were completed on-farm, in which information on iodine content in animal feed and use of teat disinfectants containing iodine was captured.

**Results** Average milk iodine levels on the 67 dairy farms were similar during the mid- and late-lactation periods: at 116 ppb (range: 10–561 ppb) and 109 ppb (range: 3–1121 ppb), respectively. During the mid- and late-lactation periods, 13 and 12 farms, respectively, produced milk with iodine levels higher than 150 ppb, which is the target level for the production of milk powder. Among those farms, 5 farms had high levels of iodine in both lactation periods. Milk iodine levels ranged between 178 and 561 ppb and between 154 and 1121 ppb on the farms with iodine levels higher than 150 ppb in the mid-lactation and late-lactation periods, respectively. The daily iodine intake from the dairy rations on a per cow basis on the farms that were higher than the 150 ppb limit varied from 31.8 to 100 mg of iodine/cow/day (mid-lactation) and 75.0 to 87.5 mg iodine/cow/day (late-lactation). It was not possible to observe a linear relationship between dietary iodine content and milk iodine concentration in the present study, given that the questionnaires did not cover other potential factors that could contribute to iodine levels in milk (e.g., boluses, addition of iodine to drinking water, mineral licks) and the contribution of grass. Regarding teat disinfection, in the mid- and late-lactation period, 4 of the 13 farms and 2 of the 12 farms, respectively, were using iodine teat disinfectants. Those products were also used on other farms among the 67 (mid-lactation: 3 farms; late-lactation: 3 farms); however, the iodine levels in their milk samples was lower than 150 ppb.

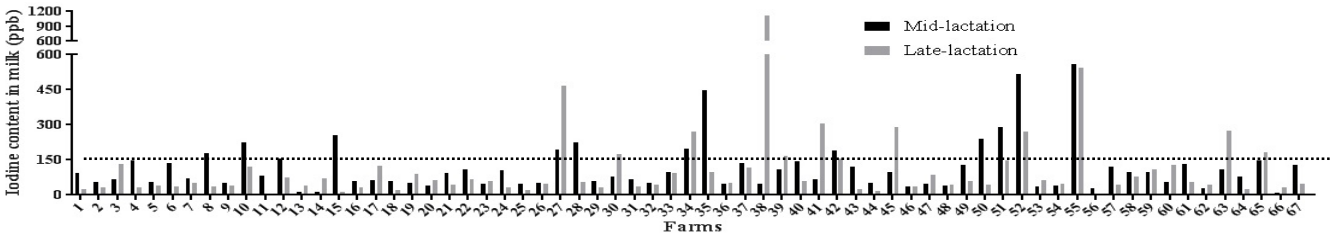


**Figure 1** Iodine levels in milk samples collected during the mid- and late-lactation period on 67 dairy farms

**Conclusion** The concentration of iodine in Irish dairy rations is higher than necessary and combined with the misuse of iodine-based teat disinfectants will result in higher levels of iodine than required in milk destined for infant formula manufacture.

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**References**

Borucki Castro, S. I., Berthiaume, R., Robichaud, A. and Lacasse, P. 2012. Journal of Dairy Science 95, 213–220.

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# Scrutinizing of *Spirulina Platensis* on growth performance and carcass characteristics of Japanese quail

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**Application** Nowadays, organic feeding of birds is very important to ensure poultry meat consumers' health. *Spirulina Platensis* is a blue-green algae that has high nutritive value due to its valuable nutrient content.

**Introduction** *Spirulina* is a microscopic single cell algae which grows in fresh water and has a simple structure but a complex composition. It is a concentrated source of food containing nutraceutical, antioxidants, and probiotic properties. Moreover, it is an important source of the blue photosynthetic pigmented protein C-phycocyanin, which has strong antioxidant and anti-inflammatory properties. Interestingly, *Spirulina* is known for its wide ranging biological activities, like prevention of anemia because of high iron and vitamin contents (Hemalatha *et al*., 2012), inhibition of herpes simplex infection (Ferreira Hermosillo *et al*., 2011). The aim of the present study was to study the effect of *Spirulina Platensis* on growth performance and carcass characteristics of Japanese quail.

**Material and methods** A total of 150 one-day-old Japanese quail *(Coturnix coturnix Japonica)* were used in a completely randomized design with 5 treatments and 3 replicates. The birds were randomly allocated to 15 pens with wood shavings litter (ten birds per pen). Average minimum-maximum temperatures and average relative humidity recorded inside the barn during the experiment were 24.25±1.15ºC - 35.20±1.50ºC, and 65±5%, respectively. The main ingredients of the diets included corn, soybean meal and corn feed meal. The diets were formulated to meet the nutrient requirements of the quail as recommended by Silva and Costa (2009). The diets were all made to be iso- caloric and iso- nitrogenous. Experimental diets including control diet (with no additive) and 4 levels of algae (2.5, 5, 10 or 20 g / kg diet) were fed to birds from 1 to 35 d of age. *Spirulina platensis* algae samples were cultivated on July of 2017 in ACECR, Sari, Mazandaran, Iran. The birds feed intake, body weight gain and feed conversion ratio (FCR) was measured weekly. The percentages of Carcass yield, breast, drumstick and femur of the quail were studied at 35 d. European production efficiency factor (EPEF) was calculated at the end of the experiment. Data from this experiment were analyzed by analysis of variance using GLM procedures (SAS institute, 2008).

**Results** Feeding quail with 5 g *Spirulina*/ kg diet resulted in a higher feed intake, body weight gain, European production efficiency factor and breast percentage compared to control group (Table 1). Quail that received 20 g Spirulina/ kg diet consumed more feed, however, they had a lower carcass yield compared to the control group ($P<0.05$). There was no significant difference among the percentages of drumstick and femur of quail ($P>0.05$).

**Table 1** Effects of *Spirulina Platensis* on quails' growth performance and carcass characteristics ($P<0.05$)

| Treatments | Feed intake (g) | Weight gain (g) | FCR | *EPEF | Carcass Yield (%) | Breast (%) | Drumstick +Femur (%) |
|---|---|---|---|---|---|---|---|
| Control | 642.81$^{c}$ | 211.45$^{c}$ | 3.039$^{ab}$ | 198.73$^{b}$ | 67.89$^{a}$ | 25.50$^{b}$ | 19.30 |
| 2.5 g Spirulina / kg | 655.46 $^{bc}$ | 216.76$^{bc}$ | 3.024$^{ab}$ | 204.84$^{b}$ | 69.06$^{a}$ | 29.23$^{a}$ | 17.33 |
| 5 g Spirulina / kg | 668.10 $^{ab}$ | 228.03$^{a}$ | 2.930$^{b}$ | 222.20$^{a}$ | 68.33$^{a}$ | 29.37$^{a}$ | 17.62 |
| 10 g Spirulina / kg | 675.11 $^{ab}$ | 217.65$^{bc}$ | 3.102$^{a}$ | 200.68$^{b}$ | 65.00$^{ab}$ | 26.07$^{b}$ | 18.08 |
| 20 g Spirulina / kg | 694.48 $^{a}$ | 219.26$^{b}$ | 3.168$^{a}$ | 201.47$^{b}$ | 63.42$^{b}$ | 25.01$^{b}$ | 17.60 |
| SEM | 7.57 | 2.197 | 0.076 | 4.88 | 1.107 | 0.697 | 0.508 |
| P value | 0.0104 | 0.0047 | 0.048 | 0.032 | 0.0263 | 0.0023 | 0.1236 |

Means within the same row with uncommon superscript differ significantly ($P < 0.05$). *European production efficiency factor

**Conclusion** Results of the present study indicated that using 5 g *Spirulina Platensis* algae / kg diet during 1-35 d of age improved body weight gain, EPEF and breast percent of Japanese quail. *Spirulina Platensis* can be consider as an organic dietary supplement in quails' nutrition, however, more research is needed on its value in poultry nutrition.

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**References**

Ferrira-Hermosillo, A., Torres-Duràn, P.V., Shamosh-Halabe, S., Juàrez-Oropeza, M.A. 2011. Revista Internacional de Cienciay.
Hemalatha K., Pugazhendy, K., Jayachandran, K., Jayanthi, C., Meenambal, M. 2012. International Journal of Chemical and Analytical Science 3, 1509-1512.
SAS, 2008. Statistical Analysis System. SAS Users Guide Statistics. Version 9.2. Ed. SAS institute Inc., Cary, NC, USA.
Silva, J. H. V., and F. G. P. Costa, 2009. 2. ed. Jaboticabal: FUNEP. 110.

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# Comparing the efficacy of teat disinfectant products against bovine mastitis-causing contagious and environmental bacteria

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**Application** Environmental bacteria, in comparison to contagious bacteria, had a higher resistance to the range of teat disinfectant products. Disinfectant products containing a lactic acid mix were most effective against both groups of bacteria.

**Introduction** The most prevalent mastitis-causing bacteria fall into one of two categories; contagious and environmental bacteria (Cheng *et al.,* 2010). Therefore, determining the most effective teat disinfectant against both types of bacteria can help to reduce the incidence of new intramammary infections. The aim of this study was to compare the efficacy of a range of teat disinfectant products against strains of environmental and contagious bacteria.

**Material and methods** Microbiological and molecular methods were used to identify bacteria strains from bulk tank milk samples that could be associated with intramammary infections. In this study, a disc diffusion assay test (CLSI, 2015) was used to determine the antimicrobial susceptibility of bacteria against 10 commercially available teat disinfectant products differing in active ingredients. The control disinfectant (dip) contained 0.5% iodine, while dips 1, 4 and 6 contained lactic acid and chlorhexidine, dips 3 and 10 contained lactic acid and salicylic acid, dip 2 contained chlorhexidine, dip 5 contained ammonium lauryl sulphate, dip 7 contained lactic acid, dip 8 contained diamine and dip 9 contained chlorine dioxide. To prepare the inoculum, cultures were grown overnight on Mueller Hinton (MH) agar. These colonies were suspended in maximum recovery diluent and the suspension adjusted to a 0.5 McFarland standard. Then, 100 µl of this suspension was spread onto a MH agar plate using an L-shaped spreader and left to dry for 15 minutes. Next, 6 mm filter paper discs were soaked in the test disinfectant for 30 seconds. Discs were placed onto agar plates using a sterile forceps, ensuring the entire disc touched the agar. Plates were incubated, inverted, at 37 ˚C for 24 hrs. The zones of inhibition were measured in millimetres (mm) using an electronic caliper. Zones of inhibition are the absence or presence of bacteria around each disc. It is an indirect measurement of the ability of the disinfectant to inhibit the bacterial strain.

**Results** After identification, bacterial strains were placed into one of two groups; environmental or contagious. The environmental group contained *E. faecalis*, *H. alvei*, *L. lactis*, *A. viridans*, *S. Liquefaciens* and *S. marcescens*, while the contagious group contained *S. devriesei, S. hominis*, *S. epidermis*, *S. chromogenes*, *S. haemolyticus* and *S. xylosus.* In comparison to the control, all teat disinfectant products had a similar efficacy or higher, with the exception of disinfectant 5 against contagious bacteria (Table 1). Overall, teat disinfectants were more effective against contagious than environmental bacteria, with teat disinfectant products containing lactic acid and salicylic acid (10) and lactic acid and chlorhexidine (1, 4, and 6) being the most effective against both groups of bacteria. Dip 10 was the most effective product against environmental bacteria with an average zone of 21 mm. Dips 4, 6 and 1 were also very effective with zones of 20 mm 18 mm, and 20 mm, respectively. A similar result was obtained for contagious bacteria with dip 10 the most effective, with an average zone of 25 mm, with dips 4, 1 and 6 obtaining zones of 24 mm, 23 mm, and 22 mm, respectively. For both groups, dip 5 was the least effective product, yielding zones of 12 mm and 16 mm, respectively.

**Table 5** Efficacy of teat disinfectant products against environmental and contagious bacteria (mm)

| Dip | C | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Enviro. | 15.36 $^{a}$ | 20.38 $^{b}$ | 16.87 $^{a}$ | 17.38 $^{a}$ | 20.39 $^{b}$ | 11.51 $^{b}$ | 18.32 $^{b}$ | 15.79 $^{a}$ | 15.18 $^{a}$ | 14.38 $^{a}$ | 21.46 $^{b}$ |
| Con. | 17.75 $^{a}$ | 22.55 $^{b}$ | 21.45 $^{b}$ | 19.53 $^{b}$ | 23.77 $^{b}$ | 15.78 $^{a}$ | 22.24 $^{b}$ | 19.76 $^{b}$ | 20.48 $^{b}$ | 21.67 $^{b}$ | 25.1 $^{b}$ |

C=Control, Enviro=Environmental, Con=Contagious, Dip=Disinfectant
$^{ab}$ denotes significant difference (P<0.05) within bacterial groups, for each teat disinfectant compared to the control
Control=Iodine, 1, 4 and 6=lactic acid and chlorhexidine, 3 and 10=lactic acid and salicylic acid, 2=chlorhexidine, 5=ammonium lauryl sulphate, 7=lactic acid, 8=diamine and 9=chlorine dioxide

**Conclusion** Results show that the disc diffusion assay was a suitable method for comparing the efficacy of products. Environmental bacteria had a higher resistance to all teat disinfectant products tested. Products most effective against both contagious and environmental bacteria contained lactic acid. This study demonstrates that there is a wide range of disinfectant ingredients available which are comparable or more effective than iodine based products for teat disinfection.

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**References**

Cheng, D., Zhu, S., Yin, Z., Ding, W., Mu, Z *et al*., 2010. African Journal of Microbiology Research 4, 1110-16.
CLSI 2015. Performance Standards for Antimicrobial Susceptibility testing.

# Proximate composition of *Stylosanthes hamata* as influenced by phosphorus sources and method of propagation

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**Application** An understanding of better source of manure and propagation methods will help producers anticipate and plan for changes in forage quality which will favour efficient and improve livestock production.

**Introduction** The mixed crop–livestock farming system practiced in the tropics and subtropics will be most affected by increase in demand for livestock products as more than 85% of the world's cattle, sheep and goats are held in these mixed systems in the tropics (Gardiner and Devendra, 1995). Addition of forage legumes in livestock feeding will aid more roughage intake and digestion for livestock (Adu *et al*., 1992). The major nutrients required for increased sward and survival of legumes in pasture is phosphorus. This research therefore aims at evaluating the effects of phosphorus sources and sowing methods on proximate composition *Stylosanthes hamata*.

**Material and methods** The experimental site was cleared, ploughed and harrowed before establishment. Poultry manure and Organo-mineral fertilizer (Aleshinloye) were used as the desired phosphorus sources while the method of propagation was the broadcasting and drilling method. The rate of application for both manures was 80kg P/ha. The quantity of each manure applied, was calculated based on the pre-determined chemical composition (i.e phosphorus) content of the manures. *Stylosanthes hamata* (seeds) were scarified and sowed based on the treatment allotment at a seed rate of 5 kg/ha. The experiment was therefore a 3 x 2 factorial arrangement consisting of three (3) phosphorus sources: (No manure i.e control, Poultry manure and Organo-mineral fertilizer (Aleshinloye)) and two propagating methods: (broadcast and drill) and was laid out in a Randomized Complete Block Design (RCBD) with three (3) replicates. Samples (*S. hamata*) were harvested at 10 weeks after sowing, weighed and oven dried at $65^0$C to a constant weight for proximate analysis according to the technique of A.O.A.C. (2000). Data were analysed using the general linear model and treatment means separated using Duncan's Multiple Range Test of the SAS software package (SAS 1999).

**Results** *S. hamata* fertilized with poultry manure recorded the highest ($P<0.05$) dry matter content while the highest ($P<0.05$) Crude protein content was recorded for Aleshinloye fertilized *S. hamata*. The broadcast method of propagation gave the highest ($P<0.05$) ash content. The interaction between the factors (i.e phosphorous sources and sowing method) were not significant ($P>0.05$) except on the ash content.

**Table 1** Effects of phosphorous sources and propagation method on proximate composition of *S. hamata*

| Factors | DM | CP | EE | ASH |
|---|---|---|---|---|
| | | % | | |
| **Phosphorus sources** | | | | |
| Control | 95.30[ab] | 11.84[b] | 11.83 | 11.38 |
| Aleshinloye | 93.75[ab] | 20.24[a] | 12.66 | 11.63 |
| Poultry | 95.50[a] | 18.71[a] | 12.02 | 10.25 |
| **SEM** | 1.07 | 1.28 | 1.34 | 0.39 |
| **Propagation method** | | | | |
| Broadcast | 94.30 | 16.99 | 11.49 | 11.80[a] |
| Drilling | 93.90 | 17.30 | 12.25 | 10.25[b] |
| **SEM** | 0.76 | 0.98 | 0.84 | 0.60 |
| **Phosphorus x Propagation method** | 0.060 | 0.900 | 0.500 | 0.040 |

[ab]: means on the same column with different superscript are significantly different.

**Conclusion** Organo-mineral fertilizer (i.e Aleshinloye) produced plants (*S. hamata*) with highest crude protein contents which suggest it as a fertilizer of high release of minerals (i.e Phosphorous) while the broadcasting way of propagation adopted in this study produced plants which had the higher content of Ash.

**References**

Adu, I. F, Fajemisin, B. A and Adamu, A. M. 1992**.** The utilization of sorghum stover fed to sheep as influenced by urea and graded levels of lablab supplementation. In: Small Ruminant Research and Development in Africa. Proceedings of the 1st Biennial Conference of the African Small Ruminant Research Network, ILRAD, Nairobi, Kenya, 10-14 December, 1990. ILCA Nairobi, Kenya. Rey B, Lebbie S H B and Reynold R (editors) 367–374.
Gardiner P. and Devendra C. (eds). 1995. Global Agenda for Livestock Research. Proceedings of a consultation, ILRI, Nairobi, Kenya. 118.

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# Regrowth potential of a grass/legume mixture in response to short-term grazing trial

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**Application** The productivity of sown pasture is hinged on its ability to regrow sufficient herbage for sustainable livestock grazing.

**Introduction** Defoliation of herbage through grazing or cutting is critical to yield responses of pasture plants. The ability of a forage plant to produce high regrowth yield can be linked to its capability to produce more leaf and tiller density, which enhances soil coverage and better weed control (Olanite *et al*., 2009), following grazing or cutting regimes. Understanding the potential of pasture regrowth is important to maximize production from sown pasture fields. The aim of the present study was to assess the regrowth potential (DM YIELD) of *Panicum/Stylo* pasture in response to short-term grazing by White Fulani (WF) calves.

**Material and methods** A demarcated section of 5 ha sown pasture field having *Panicum maximum* (Ntchisi) and *Stylosanthes guianensis* as the major botanical components was used in this study. Two forage densities (dense and sparse) were grouped into two treatments in a randomized complete block design, with each pasture type covering 15 x 5 m, and constituting a block with three replicates. Each treatment had a caged area of 1 $m^2$ for sample collection. Six WF calves with average body weight of 82kg were allowed to graze in each pasture type from 7:30 h to 9:30 h (three days per grazing period) at six-week interval. Twelve weeks after the grazing trials, herbage dry matter yield of regrowth as influenced by grazing was evaluated for both caged and uncaged areas of each plot. Herbage in two 0.5$m^2$ quadrat were randomly harvested to 10cm stubble height in the uncaged area of each plot, while those in the caged areas were harvested to the same height. Materials harvested from each quadrat in each plot was weighed (Total fresh weight, TFW), and the fresh weight of sub samples were taken (FSsW). The sub-samples were oven dried at 65$^oC$ until constant weight (dry subsample weight, DSsW). Percentage dry matter (%DM) content was calculated, and the DM yield for each treatment was estimated by multiplying the fresh weight of harvested forage from each treatment by its %DM i.e FSsW x %DM = DM yield. The sub sample DM yield was used for extrapolation in t $Ha^{-1}$. Data were analysed using t-test in R statistics (R Core Team, 2015).

**Results** There was a significant ($P<0.05$) difference in the dry matter yield of regrowth as influenced by grazing in the plots with dense biomass. The caged area recorded higher yield of regrowth (2.28 t $Ha^{-1}$) than the grazed area (1.69 t $Ha^{-1}$), presumably due to higher production of tillers. The total dry matter yield obtained in the grazed area of dense plot was lower, but very similar to that obtained in sparse plot. There was no significant difference in the dry matter yield recorded for both caged and grazed areas of sparse plots.

**Table 1** t-test Result comparing Total Dry Matter Yield (t $Ha^{-1}$) from caged and grazed areas of both dense and sparse plot

| | Dense | | | Sparse | | |
|---|---|---|---|---|---|---|
| | Mean | SEM | P value | Mean | SEM | P value |
| Caged | 2.28 | 0.09 | **0.001** | 2.05 | 0.10 | 0.17 |
| Grazed | 1.69 | 0.11 | | 1.70 | 0.21 | |

SEM: Standard error of mean.

**Conclusion** short-term grazing resulted in decreased dry matter yield of regrowth in the grazed areas of both dense and sparse pasture biomass compared to the caged areas. In this situation, management strategies should include possible ways of augmenting the intake requirement of the animals, for example by providing conserved forage in the form hay or silage.

**References**

Olanite, J. A., Tarawali, S. A. and Aken'Ova, M. E. 2009. Journal of Agricultural Science and Environment 9(1), 28-38.
R Core Team, 2015. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria ISBN 3–900051-07–0, URL. http://www.R-project.org/.

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# Growth and faecal egg count response of naturally grazing West African dwarf sheep to supplementation of neem (*Azadirachta indica*) leaf meal concentrate

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**Application** The supplementation of neem leaf meal concentrate diet improved the growth performance with a reduction in faecal egg count of sheep grazing natural pastures.

**Introduction** Neem *(Azadirachta indica*) is a readily available unconventional feedstuff that can be incorporated into the feed of the animals to promote their growth. The leaves of neem plant contain appreciable amount of protein, carotene and adequate amount of trace minerals with phytochemical compounds (Ascher *et al.,* 1995, Okoli *et al*., 2001) which may be helpful in promoting growth and management of intestinal worms in livestock. The aim of the present study was to evaluate the effect of supplementation of varying inclusion levels of neem leaf meal in a concentrate diet on the growth and faecal egg count of West African Dwarf sheep grazing natural pastures.

**Material and methods** Twenty (20) West African Dwarf sheep aged 5-6 months and average weight of 11.15±0.49kg was grouped into 4 dietary treatments in a completely randomized design. The animals were tagged for identification and allowed to graze natural pastures for six (6) hours daily (0800hrs – 1400hrs). Fresh neem leaves were chopped for effective drying, milled to leaf meal and incorporated into a 16% crude protein concentrate diet at 0g, 5g, 10g, 15g neem leaf/ 100g concentrate /animal /day, respectively. Bodyweights of the animals were taken on weekly basis for the 90-day experimental period. Faecal samples of about 2 to 4 grams were obtained directly from the rectum of each sheep at the beginning of the experiment and on weekly basis. Data collected were subjected to one-way analysis of variance in a completely randomized design and analysed using SAS (1999) and means were separated (Duncan, 1955).

**Results** Growth of sheep improved ($P<0.05$) with the inclusion of neem leaf concentrate compared to the control, albeit the growth of sheep decreased with an increase in NLM inclusion in the diets (Table 1). Average weight gain (g/day) ranged from 29.00 to 45.44 in sheep supplemented with 0 and 5g NLM inclusion, respectively. A range of 33.38 to 88.00% reduction in faecal egg count was observed across treatments with sheep supplemented with NLM having the higher % reduction compared to the unsupplemented control treatment, possibly as a result of phytochemical compounds in neem leaves.

**Table 1** Weight gain and faecal egg count of West African Dwarf sheep supplemented with neem leaf meal concentrate

| Parameters | 0gNLM | 5gNLM | 10gNLM | 15gNLM | ±SEM |
|---|---|---|---|---|---|
| Initial weight (kg) | 11.48 | 11.06 | 10.81 | 11.25 | 0.49 |
| Final weight (kg) | $14.09^{b}$ | $15.15^{a}$ | $14.24^{b}$ | $14.44^{b}$ | 0.65 |
| Weight gain (kg) | $2.61^{c}$ | $4.09^{a}$ | $3.43^{b}$ | $3.19^{b}$ | 0.34 |
| Average daily weight gain (g/day) | $29.00^{c}$ | $45.44^{a}$ | $38.11^{b}$ | $35.44^{b}$ | 2.28 |
| Initial faecal egg count (egg/g) | $800^{a}$ | $833^{a}$ | $533^{b}$ | $563^{b}$ | 13.33 |
| Final faecal egg count (egg/g) | $533^{a}$ | $100^{b}$ | $133^{b}$ | $143^{b}$ | 9.17 |
| % Reduction in faecal egg count | $33.38^{b}$ | $88.00^{a}$ | $75.05^{a}$ | $78.80^{a}$ | 3.21 |

$^{a,b,c}$ Means with same superscripts within the rows are not significantly different ($P>0.05$)
NLM- Neem leaf meal

**Conclusion** The improved performance of sheep supplemented with neem leaf meal concentrate in this study indicates its potential to help increase growth and decrease faecal egg counts in sheep grazing natural pastures. The dietary inclusion of neem leaf meal at 5% in a concentrate diet could therefore play a valuable role in supplying supplemental nutrients for the optimum performance of sheep grazing natural pastures.

**References**

Ascher, H., Isman, K. R. S. 1995. The Neem tree: *Azadirachta indica* and other Meliaceous Plants. Weinheim, Germany.
Duncan, 1955. Multiple Range and Multiple F test, Biometrics 11, 1- 42.
Okoli, I.C., Ebere, O. O., Emenalom, M. C., Uchegbu and Esonu B. O. 2001. Indigenous Livestock Production Paradigms Revisited. III: An assessment of the proximate values of most preferred indigenous browses of South Eastern Nigeria. Tropical Animal Production Investigation 4, 99-107.
SAS 1999. Statistical Analysis System user guide: Statistics version 8 editions, statistical analysis institute INC, Cary. NC.

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# Chemical composition and ruminal degradation characteristics of West African fodder trees

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**Application** Fodder trees represent a significant potential source of protein for ruminants in the tropics. However, usage is limited by incomplete knowledge of the factors affecting their nutritional value.

**Introduction** Ruminant production in many parts of sub-Saharan Africa is hampered by a shortage of feed and low-quality forages. The use of fodder trees represents a source of highly nutritious feed to supplement bulk feeds during the dry seasons. They can serve as a rich source of crude protein and minerals especially during the dry season when livestock production is severely depressed. However, fodder trees and shrubs are known to contain anti-nutritional factors such as tannins, saponins and non-protein amino acids. This may limit their palatability or inclusion rate in the ration. Whilst a considerable amount of work has been done to determine the chemical composition and the dry matter degradability of fodder trees, the sheer variety of the species being used across the continent means that there is more work to be done. There is also significant variation in the chemical composition and ruminal degradability in the same tree species growing in different environments (Tefera *et al* 2008). Samples of foliage from West African trees were investigated for their potential as feed supplements for ruminants. Samples of Acacia nilotica (AN), Azadirachta indica (AI), Guiera senegalensis (GS), Lannea acida (LA), Parkia biglobosa (PB), Piliostigma reticulatum (PR) and Ziziphus mauritiana (ZM) were collected from three distinct ecological zones in Nigeria (The Sahel Savannah (Zone 1), the Sudan Savannah (Zone 2) and the Northern Guinea Savannah (Zone 3).

**Material and methods** Samples were analysed for dry matter (DM), crude protein, ash, neutral detergent fibre, acid detergent fibre, lignin and tannins. Sample were incubated with rumen fluid and *in vitro* gas production measured at 0, 1, 2, 3, 4, 6, 8, 12, 24, 48, 72 and 96h. The equation $y= a + b (1 – e^{(-ct)})$ was used to describe the kinetics of gas production (Ørskov & McDonald, 1979). Methane concentration was determined at 24 and 48h. After 96h, the incubation was stopped and volatile fatty acid (VFA) concentrations were determined. Freeze-dried residues from the incubation were used to estimate *in vitro* DM and organic matter digestibility. The effect of forage variety and interaction with site of collection was determined by analysis of variance in Genstat 18th Edition (VSN International, Hemel Hempstead, UK).

**Results** Both species and ecological zone influence the chemical composition, the extent and rate of *in vitro* gas production (Table 1) and the pattern and extent of methane and VFA production ($p < 0.001$, SED for extent and rate of gas production 9.10 and 0.455 for the effect of plant and 15.76 and 0.788 for the interaction of plant and zone respectively).

**Table 6** The rate (c, %h) and extent (a+b, ml) of gas production of West African fodder trees grown in different environmental zones incubated in rumen fluid *in vitro*

| Plant | Crude protein (%) | | | Acid detergent lignin (%) | | | Extent (a+b, ml) | | | Rate of degradation (c, %/h) | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Zone | | | | | | | | | | | |
| | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 | 1 | 2 | 3 |
| AN | 11.06$^{a1}$ | 15.09$^{a2}$ | 15.13$^{a2}$ | 6.43$^{a}$ | 6.97$^{a}$ | 6.07$^{a}$ | 65.5$^{a}$ | 74.9$^{a}$ | 91.3$^{a}$ | 3.31$^{a}$ | 2.44$^{a}$ | 1.94$^{a}$ |
| AI | 15.33$^{b1}$ | 14.08$^{b2}$ | 22.23$^{b3}$ | 14.55$^{b}$ | 12.43$^{b}$ | 14.24$^{a}$ | 106.5$^{b}$ | 58.4$^{bd*}$ | 95.3a | 3.25$^{a}$ | 4.29$^{bc}$ | 3.43$^{b}$ |
| GS | 11.13$^{a}$ | 11.97$^{c}$ | 11.61$^{c}$ | 23.21$^{c}$ | 18.57$^{b}$ | 23.07$^{b}$ | 37.9$^{c}$ | 26$^{9c}$ | 25.1$^{b}$ | 5.73$^{b}$ | 5.10$^{c}$ | 3.96$^{b}$ |
| LA | 11.62$^{a1}$ | 13.86$^{b2}$ | 8.94d$^{3}$ | 10.98$^{ab}$ | 13.70$^{b}$ | 9.140$^{a}$ | 37.4$^{c}$ | 48.4$^{d}$ | 38.7$^{bd}$ | 4.23$^{c}$ | 2.71$^{a}$ | 3.26$^{b}$ |
| PB | 14.58$^{b1}$ | 13.98b1 | 13.09$^{e2}$ | 10.01$^{ab}$ | 14.50$^{b}$ | 2.63$^{a}$ | 48.0$^{c}$ | 88.1$^{a*}$ | 31.5$^{b}$ | 3.38$^{ac}$ | 2.69$^{a}$ | 3.03$^{bc}$ |
| PR | 11.28$^{a1}$ | 14.30$^{bd2}$ | 12.77$^{e3}$ | 2.92$^{a}$ | 17.50$^{b}$ | 15.66$^{a}$ | 54.4$^{a}$ | 58.5$^{bd1}$ | 63.7$^{c13}$ | 4.52$^{bc}$ | 3.70$^{bd}$ | 4.21$^{bd}$ |
| ZM | 17.70$^{c1}$ | 13.74$^{be2}$ | 17.72$^{f1}$ | 11.15$^{abd}$ | 10.75$^{a}$ | 11.09$^{a}$ | 67.4$^{a}$ | 63.1$^{b1}$ | 55.6$^{cd3}$ | 2.85$^{b}$ | 2.91$^{ad}$ | 3.86$^{b}$ |

**Conclusion** All tree species had CP content above 10% suggesting they could be used as supplements to bulk fodder. However, not all the species are degraded uniformly in the rumen. Chemical composition therefore is not reliable as the sole predictor of suitability.

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**References**

Ørskov, E. and McDonald, I. 1979.The Journal of Agricultural Science 92(2), 499-503.
Tefera, S., Mlambo, V., Dlamini, B., Dlamini, A., Koralagama, K. and Mould, F. 2008. Animal Feed Science and Technology 142, 99-110.

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# Impact of a reseeded sward replacing a permanent pasture on milk yield, milk components and sward productivity in the first year of establishment

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**Application** When a new sward is established to replace a degraded permanent pasture the milk production and land productivity can be improved in the first year even before the sward has fully established. It is expected that the benefits increase further in subsequent years.

**Introduction** Reseeding permanent pasture when it becomes unproductive or when its quality declines, mainly due to a decline in yield and botanical deterioration (De Vliegher and Carlier, 2007), is a vital farm management decision; especially concerning performance during the first year of establishment. Reseeding allows favourable species and varieties to be sown with improved traits such as increased yield, enhanced nutritional value or disease resistance, but establishment in the first year is critical to good returns. The objective of this study was to assess the impact on milk production and components of dairy cows grazing a reseeded perennial ryegrass sward replacing a permanent pasture in the first year of establishment.

**Material and methods** Fourteen hectares were reseeded with a *Lolium perenne* cultivar (AberMagic) in September 2015. Besides, 14 ha of a permanent pasture (PP) were allocated to the experiment. Allocation to treatment was based on commercial decisions based on pasture availability. Fifty-five autumn calving dairy cows were allocated to graze the PP and 70 cows to the reseeded sward (RS). Cows were turned-out for on/off rotational grazing on April 18th (2016), and from May 4th cows grazed 24 h a day. Cows received 2 kg/d concentrate in the milking parlour. Milk production, milk fat (MF), milk protein (MP), lactose (ML) contents and somatic cell count (SCC) were assessed on two occasions in May and June. Snip samples of both swards were collected periodically from April to June 2016 and were analysed for WSC, crude protein (CP), fibre fractions and metabolizable energy contents. Herbage yield was not directly measured. Baseline data of milk yield and components prior to the beginning of the trial were used as covariates and the effects of the body condition score and lactation number were included as factors in the linear mixed models ("cow" as the random effect) using Genstat 18 (VSNI).

**Results** Nutritional value differed between pasture across samplings, with PP showing higher CP and WSC than RS at the beginning of the grazing season. The WSC content of the RS only surpassed that of PP in the sampling of May 3rd (Table 1). The decrease in WSC of the RS in the last sampling could be explained by the difference in time of regrowth (longer in the PP). The RS showed a higher milk production and lactose content in May and a lower SCC (Table 2). No differences were observed in milk traits in June. An additional output of the system was 33 t DM ensiled from the RS, whilst no silage was made from PP from the same land area, representing a higher carrying capacity of the sward.

**Table 1** Crude protein (CP) and water soluble carbohydrates (WSC) content of a RS and a PP on April, May and June 2016.

| | CP content (g/100 g DM) | | | | | | WSC content (g/100 g DM) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 08/04 | 19/04 | 03/05 | 17/05 | 08/06 | SEM | 08/04 | 19/04 | 03/05 | 17/05 | 08/06 | SEM |
| HSG | 22.4 | 18.8 | 15.1 | 18.6 | 27.4 | 0.517 | 16.8 | 22.7 | 27.1 | 16.7 | 10.8 | 0.663 |
| PP | 23.9 | 21.0 | 15.6 | 20.4 | 17.6 | 0.517 | 18.3 | 25.3 | 25.8 | 16.3 | 19.1 | 0.663 |

**Table 2** Mean milk yield and milk components recorded on May 4th and June 10th 2016 from cows grazing a RS and a PP

| | Yield (L/cow) | | Milk fat (%) | | Milk Protein (%) | | Lactose (%) | | SCC (x1000) | | LogSCC | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | May | June | May | June | May | June | May | June | May | June | May | June |
| RS | 24.6 | 20.9 | 4.2 | 3.74 | 3.47 | 3.6 | 4.41 | 4.42 | 59.4 | 64.9 | 1.774 | 1.812 |
| PP | 22.9 | 21.7 | 4.25 | 3.71 | 3.5 | 3.62 | 4.34 | 4.48 | 113.3 | 66.4 | 2.054 | 1.822 |
| SED | 0.58 | 0.91 | 0.100 | 0.136 | 0.069 | 0.072 | 0.025 | 0.024 | | | 0.059 | 0.061 |
| P value | 0.005 | NS | NS | NS | NS | NS | 0.007 | 0.06 | | | <0.001 | NS |

**Conclusion** Replacing a permanent pasture with a new reseeded sward with a high quality perennial ryegrass cultivar had little impact on milk quality or yield at a cow level but increased land productivity and subsequently potential yield at a herd level in the first year of establishment.

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**References**

De Vliegher, A. and Carlier, L. 2007. Grassland Science in Europe vol. 12.

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# Effect of *Macroptilium atropurpureum* (siratro) inclusion in *Cenchrus ciliaris* (buffel grass) pasture on herbage chemical composition and *in vitro* digestion and fermentation characteristics

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**Application** *Cenchrus ciliaris* cv Molopo is a perennial grass with low nutritive attributes when preserved as dry hay. Intercropping with Siratro improved its nutritive value and *in vitro* fermentation.

**Introduction** *Cenchrus ciliaris* is native to Botswana but the nutritive value of Molopo variety is not adequate for maintenance and production by ruminants. Its crude protein ranges between 30 and 100 g/kg and digestibility from 300 to 600 g/kg. Therefore, intercropping it with a legume would improve its nutritional value. Previous research in Botswana (APRU 1979) evaluated biomass production of Cenchrus/Siratro sward and not its nutritive value. Therefore, the objective of this study was to determine the nutritive value and fermentative parameters of Cenchrus/Siratro sward.

**Material and methods** Samples of Cenchrus (C), Cenchrus/Siratro sward (CS) and Siratro (S) were randomly obtained during 2015 and 2016 from field plot BUAN farm where S proportion in CS swards varied between 17 and 20%. Three sampling were obtained during the rainy and dry seasons in each year and pooled according to C, CS and S. In 2016, during the dry season, cenchrus/siratro sward was not available. Sub-samples were obtained and standard AOAC procedures were used to determine chemical composition in duplicates. *In vitro* gas production was measured in two runs using calibrated syringes following modified methods of Menke and Steingass (1988). Samples (0.5g), in duplicates, were filled into Ankom fibre bags and incubated in syringes containing rumen/buffer mixture. Ankom buffer solutions A & B were used and syringes incubated in a water bath at 39$^{o}$C for 3, 6, 12, 24, 48, 72 and 96hrs during which gas volume was recorded. *In vitro* dry matter digestibility (IVDMD) was determined after incubation once NDF was completed on the residues. Gas data was fitted into Ørskov and McDonald' (1979) equation without the zero hr but with a lag time. Effective degradability (ED=bc/(c+k); Ørskov, 1992) was estimated at assumed outflow rate of 0.03/hr ($ED_3$) or 0.05/hr ($ED_5$). Differences due to forage type was determined using GLM of SAS (2002-08). Season, year and their interactions were also specified in the model (not reported). The results are reported as LSD means ± SEM.

**Results** CS (Cenchrus/Siratro) was intermediate to C (Cenchrus) and S (Siratro) for CT (condensed tannins), ADL (Acid detergent lignin), NDF (Neutral detergent fibre), CP (crude protein), IVDMD (*in vitro* dry matter digestibility) and Lag time, (Table 1).

**Table 1** Nutritive value and *in vitro* digestibility (g/kg DM) and *in vitro* fermentative characteristics of cenchrus/siratro sward.

| | Ash | CT* | ADF | ADL | NDF | CP |
|---|---|---|---|---|---|---|
| Nutritive value | | | | | | |
| C | 88.8±13.51 | 1.8±0.48$^{b}$ | 488.8±6.20$^{b}$ | 86.8±7.32$^{b}$ | 821.9±9.96$^{c}$ | 71.8±0.28$^{c}$ |
| CS | 91.0±21.36 | 2.2±0.76$^{b}$ | 480.8±9.80$^{b}$ | 95.7±11.57$^{ab}$ | 776.5±15.75$^{b}$ | 87.1±0.45$^{b}$ |
| S | 86.9±13.51 | 4.5±0.48$^{a}$ | 399.2±6.20$^{a}$ | 117.6±7.32$^{a}$ | 526.8±9.96$^{a}$ | 171.6±0.28$^{a}$ |
| P value | 0.9865 | 0.0059 | <0.0001 | 0.0337 | <0.0001 | <0.0001 |
| Fermentation | | | | | | |
| | b (ml/0.5g) | c (ml/hr) | Lag (hrs) | ED3 (ml/0.5g) | ED5 (ml/0.5g) | IVDMD (g/kg) |
| C | 53.1±0.60$^{b}$ | 0.044±0.0006$^{b}$ | 2.1±0.12$^{c}$ | 31.5±0.49$^{b}$ | 24.8±0.42$^{b}$ | 537.5±12.69$^{c}$ |
| CS | 53.9±0.95$^{b}$ | 0.044±0.0009$^{b}$ | 1.7±0.19$^{b}$ | 32.2±0.77$^{b}$ | 25.5±0.66$^{b}$ | 601.6±15.54$^{b}$ |
| S | 58.5±0.60$^{a}$ | 0.048±0.0006$^{a}$ | 1.2±0.12$^{a}$ | 36.2±0.49$^{a}$ | 28.8±0.42$^{a}$ | 702.9±12.67$^{a}$ |
| P value | <0.0001 | <0.0001 | <0.0001 | <0.0001 | <0.0001 | <0.0001 |

*CT; condensed tannins, ADF; Acid detergent fibre, ADL; Acid detergent bound lignin, NDF; Neutral detergent fibre; CP = crude protein, IVDMD; *in vitro* dry matter digestibility. Different superscripts within a row denotes significant difference at P<0.05.

**Conclusion** Siratro inclusion in Cenchrus pasture offers an opportunity to improve the nutritive value of cenchrus.

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**References**

Animal Production Research Unit 1979. Botswana Government Printers. Gaborone.
Menke, K. H. and Steingass, H. 1988. Animal Resources Development 28,7-55.
Ørskov, E.R. 1992. Protein Nutrition in Ruminants. 2nd Edition Academic Press, London.
Ørskov, E.R. and McDonald, I. 1979. Journal of Agricultural Science 92, 499-503.

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# Mixing or delaying weaning did not improve piglets' post-weaning growth performance

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**Application** Mixing litters before weaning or delaying weaning three days had no effect on pigs' growth and feed conversion ratio up to 10 weeks of age.

**Introduction** Weaned piglets undergo a chain of stressful events (e.g., transition from liquid to solid food, separation from the dam, mixing with unfamiliar piglets, etc.) usually resulting in anorexia and growth impairment (Kim *et al*., 2012). Reducing the number of challenges at weaning, such as avoiding mixing, has been considered to help piglets cope with weaning (Ekkel *et al*., 1995). The objective of the present work was to study 1) the effect of mixing litters before weaning and 2) the effect of delaying weaning on piglets' growth performance after weaning.

**Material and methods** Two experiments were performed at AFBI experimental farm. In Exp. 1, 54 sows (parities 1 to 6) and their litters were allocated in pairs to one of the following treatments: T1) litters kept individual during lactation and weaned as normal; T2) two litters mixed from day 7 post-farrowing and weaned as normal; T3) two litters kept individual, mixed at weaning but stayed in the farrowing pen for 3 days before being moved to the weaning facility; T4) two litters mixed from day 7 post-farrowing and at weaning they stayed in the farrowing pen for 3 days before being moved to the weaning facility. In Exp. 2, at weaning 24 litters were allocated to one of the following treatments: TA) litters weaned as normal; TB) litters kept in the farrowing pen without the sow for one day; TC) litters kept in the farrowing pen without the sow for three days. In both experiments, ten piglets per litter were individually tagged and monitored (balanced for sex and weight). At weaning (28 days) litters were moved to weaning facilities according to each treatment and allocated in groups of 20. Piglets were weighed at birth, day 7 (Exp. 1), weaning, and at 7 and 10 weeks of age. Average daily feed intake (ADFI), average daily gain (ADG), and feed conversion ratio (FCR) after weaning was recorded. Data was analysed using GenStat 16$^{th}$ edition. All models were analysed using the litter as the experimental unit.

**Results** Results from Exp. 1 are presented in Table 1. The average body weight (BW) for all the pigs was 8.5±1.70, 15.0±2.83, and 26.4±6.01kg at weaning, 7 and 10 weeks of age, respectively. No treatment effect was observed on piglets' ADG, ADFI or FCR after weaning ($P > 0.05$, respectively). Similarly, in Exp. 2 the average BW for all the pigs was 9.4±0.21, 17.3±0.60, and 32.6±1.03kg at weaning, 7 and 10 weeks of age, respectively. No treatment effect was observed on piglets' ADG, ADFI or FCR after weaning ($P > 0.05$, respectively).

**Table 1** Effect of treatment (Exp. 1) on piglets' growth performance after weaning

| | T1 | T2 | T3 | T4 | **SED** | **P** |
|---|---|---|---|---|---|---|
| nº pigs/nº sows | 124/14 | 109/13 | 137/14 | 139/14 | | |
| Weaning to 7 weeks of age | | | | | | |
| ADG, g | 446 | 420 | 434 | 425 | 22.2 | 0.661 |
| ADFI, g | 500 | 486 | 489 | 474 | 23.0 | 0.746 |
| FCR | 1.13 | 1.16 | 1.13 | 1.12 | 0.049 | 0.859 |
| 7 to 10 weeks of age | | | | | | |
| ADG, g | 704 | 665 | 745 | 702 | 49.3 | 0.742 |
| ADFI, g | 1252 | 1192 | 1242 | 1185 | 50.0 | 0.445 |
| FCR | 1.80 | 1.82 | 1.67 | 1.69 | 0.075 | 0.161 |
| Weaning to 10 weeks of age | | | | | | |
| ADG, g | 575 | 543 | 589 | 563 | 31.5 | 0.512 |
| ADFI, g | 876 | 838 | 859 | 833 | 31.8 | 0.514 |
| FCR | 1.14 | 1.15 | 1.10 | 1.11 | 0.032 | 0.336 |

**Conclusion** None of the management strategies oriented to reduce weaning stress in our studies resulted in improved growth performance after weaning. Delaying weaning, mixing before weaning or mixing and delaying weaning did not improve growth and feed intake. However, pigs' welfare and behaviour or performance to slaughter was not evaluated.

**References**

Kim, J.C., Hansen, C.F., Mullan, B.P., and Pluske, J.R. 2012. Animal Feed Science and Technology 173, 3–16.
Ekkel, E.D., van Doorn, C.E., Hessing, M.J., and Tielen, M.J. 1995. Journal of Animal Science 73, 1544–1551.

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# Assessment of carcass traits and internal organs of broilers fed 'wood ash digested' maize cobs based diets as part replacement of fibre fortified with grandizyme

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**Application** The most important factors militating against broiler production in Nigeria is feed, hence, it becomes imperative that alternative feed stuffs are sought to replace the conventional ones whose availability is becoming scarce due to high demand by other livestock species.

**Introduction** Broiler production in Nigeria is facing high cost of feed ingredients, estimated to be over 70% of the intensive production hence problematic due to high competition for its use by other livestock species. Maize cobs are seen littering the surroundings, markets and streets, constituting a nuisance. The present study seeks to evaluate the use of maize cobs as a partial replacement of fibre source ingredients. However, limitations in its use in poultry diets need to be overcome in order to enhance digestibility. Aruby and Pierson (2003) stated that enzymes have been used to supplement the bird's developing endogenous enzymes in breaking down complex food substances, hence the use of wood ash digestion and grandizyme fortifier. The objective of this experiment is to evaluate the performance of broilers fed with dietary inclusion of maize cob digested with wood ash that is fortified with enzyme.

**Material and methods** Dried maize cobs were collected from the feed mill of the Oyo State College of Agriculture and Technology. Wood ash was collected from a bakery in Igbo-Ora, it was filtered and was soaked in water at the rate of 500g per liter. The residue was removed and the milled maize cobs were soaked in the lyre solution for a period of 72 hours, drained and sundried to a constant weight. Biovet – Yc enzyme that was used in the experiment was purchased from reputable drug market. It is derived from Trichodermal viride, which was developed to complement the digestive enzyme of poultry and recommended to be included at the rate of 500g/tonne of feed. The 40 marshall broilerwere fed and watered *ad-libitum* on commercial broiler starter diet for 4 weeks. At the end of fourth week, birds were randomly allotted into five dietary treatments of 40 chicks per treatment in a completely randomized design (CRD) A-E. The control A had no Wood Ash Digested Maize Cob (WADMC) nor enzyme, while diets B, C, D and E had 5, 10, 15 and 20% inclusion levels of WADMC respectively, with 5g of the enzyme per 100kg of feed. Each diet was fed to each treatment of 40 birds in four replicates starting from four weeks after starter diets and data collected were subjected to Duncan Multiple Range Test and means were separated.

**Results** The results obtained for weight gain at the end of the eighth week shows that TE20%) was significantly ($P<0.05$) different in values for both starter and finisher stages for the five treatments. The gradient increased in size and values, however, TA(0%) had the least weight gain ($P>0.05$). The study showed distinctive effects on the primal cuts of the broiler carcasses. Values obtained for gizzard showed significant differences between TC(10%), TD(15%), and TE(20%) while TA(0%) and TB(5%) showed no significant difference in their gizzard (7.09, 8.04, 6.83 and 7.93, 7.13), respectively. It appears however, that maize cobs fortified with grandizyme treatment level helps in preventing veterinary intervention. Keelhows significant ($P<0.05$) differences between the TC(105) and TE(20%) as reflected in their means values because the more the inclusive level in diets, the higher the response of the body growth rate and the back meat. The thigh and drumstick cuts treatments for A, C and E did not differ significantly from each other in their values. However, birds in TB(5%) and TD(15%) which had high inclusion levels of treated maize cobs fortified with grandizyme had low convertion feed to meathan the other treatments. Finally, the keel cuts of treatments A and E were significantly different ($P<0.05$) from that of treatments B and D positively. However, birds in treatment E recorded incidence of prolapse which could be due to the high level of fiber in the diet Adegbola and Okonkwo, (2002).

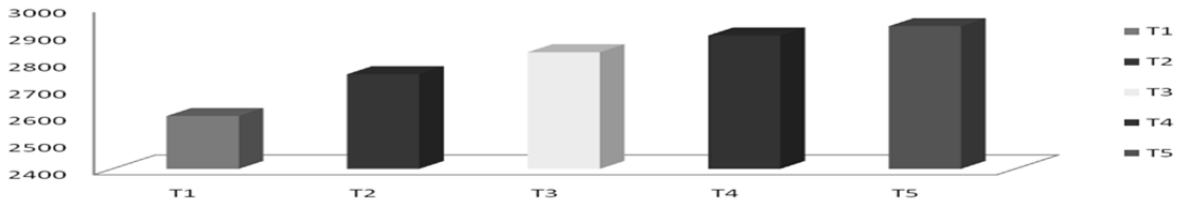


**Fig 1: Weight gain (g) of birds fed varying levels of wood ash treated maize cobs based diet fortified with enzyme**

**Conclusion** It could be concluded that the performance and carcass characteristics of WADMC based diets as partial replacement of fiber at 10 and 15% levels of inclusion fortified with 500g/T of enzyme was better than those on 5 and 20%.

**References**

Adegbola, T.A. and Okonkwo, J.C. 2002. Nigeria. Journal of Animal Production 29 (1), 21-26.
Aruby, M. and Person, E.E.M. 2003. Poultry Science Journal 91, 322-326.

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# Pasture finishing of early-maturing suckler-bred cattle at 19 months of age: bulls versus steers

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**Application** Aberdeen Angus-sired suckler steers can be adequately finished at pasture at 19 months of age, whereas bulls cannot.

**Introduction** As grazed grass is considerably cheaper than conserved forages and concentrate feeds in temperate climates (Finneran *et al*., 2012), finishing cattle from pasture is less costly. Compared to steers, bulls have biological advantages in terms of growth rate and feed efficiency (O'Riordan *et al*., 2011). Suckler bull beef production usually involves a post-weaning indoor feeding period based on conserved forage supplemented with concentrates or concentrates offered *ad libitum* (O'Riordan *et al*., 2011). Within these production systems achieving a commercially acceptable carcass fat score (6, scale 1-15) is a primary market requirement. There is relatively little published information comparing the performance of pasture-finished bulls and steers. Therefore, the objective of this study was to evaluate the growth, carcass and selected meat quality traits of spring-born Aberdeen Angus-sired suckler bulls and steers finished at pasture at 19 months of age.

**Material and methods** Thirty weaned, spring-born Aberdeen Angus-sired bulls ca. 8 months old were blocked on weight and from within block randomly assigned to a bull or steer finishing system (n=15). Castration was carried out by a veterinarian. During the indoor winter period, animals were offered grass silage to appetite, supplemented with 1.6 kg dry matter (DM) daily of concentrates. Animals were turned out to pasture on 7 April and rotationally grazed *Lolium perenne*-dominant swards (ca. 6.5 bulls/ha) for 193 days. Total grazing area was split into equal-sized farmlets comprised of paddocks. Both treatments were allocated herbage of similar pre-grazing height (8.9 cm) and mass (1474 kg DM/ha); mean pasture organic matter digestibility was 775 g/kg DM. Average residency period per paddock was 3.4 days and herbage disappearance was measured. Ultrasonic fat depth was measured pre-slaughter, and post-slaughter, carcass weight and carcass conformation and fat score were determined. At 48 h post-mortem, lightness (L*), redness (a*) and yellowness (b*) of subcutaneous fat, and pH, colour (after 1 h bloom) and area of *M. Longissimus* muscle were recorded and, the 6-10 ribs joint was dissected. Data were statistically analysed using ANOVA. The model contained terms for treatment and block.

**Results** Mean estimated herbage DM intake (kg/day) for bulls was 7.4 and for steers, 6.5. Bulls had a higher (P<0.05) daily live weight gain, slaughter weight, carcass weight, kill-out proportion, carcass conformation and *M. Longissimus* area (89.0 v. 69.1 $cm^2$), and lower ultrasonic measures of body fatness and carcass fat score than steers (Table 1). Ribs joint lean proportion was higher (705 v. 628 g/kg) and fat proportion was lower (76 v. 146 g/kg) for bulls than steers (P<0.001). Ultimate muscle pH did not differ (5.63 v. 5.62, P>0.05) between gender, whereas subcutaneous fat 'L' (65.1 v. 70.2) and 'b' (15.0 v. 16.8), and muscle 'L' (28.9 v. 30.8) and 'b' (11.4 v. 11.9) values were lower (P<0.05) for bulls than steers.

**Table 1** Growth and carcass traits of early-maturing suckler-bred bulls and steers finished at pasture at 19 months

| | Bull | Steer | s.e.m. | P-value |
|---|---|---|---|---|
| Turn-out to pasture weight (kg) | 398 | 395 | 8.1 | 0.845 |
| Slaughter weight (kg) | 613 | 571 | 11.0 | 0.015 |
| Daily live weight gain (kg) | 1.13 | 0.92 | 0.034 | <0.001 |
| Carcass weight (kg) | 342 | 307 | 7.7 | 0.006 |
| Kill out proportion (g/kg) | 558 | 539 | 5.5 | 0.029 |
| Carcass conformation score (1-15) | 8.1 | 6.4 | 0.36 | 0.004 |
| Carcass fat score (1-15) | 6.3 | 8.4 | 0.22 | <0.001 |

**Conclusion** Steer carcasses were lighter but adequately finished, whereas heavier bull carcasses were only marginally finished. Gender had relatively minor effects on subcutaneous fat and muscle colour.

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**References**

Finneran, E., Crosson, P., O'Kiely, P., Shalloo, L., Forristal, D. and Wallace, M. 2012. Journal of Agricultural Science (Cambridge) 150, 123-139.
O'Riordan, E.G., Crosson, P. and McGee, M. 2011. Irish Grassland Association Journal 45, 131-146.

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# The prevalence of pelvic asymmetry in lame and non-lame Holstein cows

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**Application** Pelvic asymmetry can be linked to hindlimb lameness and lactation stage. All cows with hindlimb lameness (n=8) had right pelvic asymmetry (tuber coxae right greater than tuber coxae left)

**Introduction** Lameness studies and surveys imply that lameness is an increasing hindrance on the dairy industry incurring large scale losses as a result of reduced productivity amongst affected animals; UK prevalence alone ranging from 17-37% in the last ten years (Archer *et al.,* 2010). Earlier detection of lameness could be the solution to a reduction in afflicted bovines (Blackie *et al.,* 2013). Detection of static pelvic asymmetry could be a useful indicator of lameness that can be transferred from equines where it has proven highly successful (Pfau *et al.,* 2013).

**Material and methods** 28 Holstein dairy cows where selected from a herd of 450. All 450 cows were subjected to a locomotion score by an experienced professional using the DairyCo 0-3 scoring system and identified affected limbs. Cows were randomly selected from locomotion score groups 0 (n=14) and 2 (n=14). All cows were aged between 2-7yrs, lactation groups varied 1-6, DIM ranged 33-402. Bosch Laser measure was taped onto a 20cm dowel with a spirit level which was placed against a sticker located on the cranial aspect of the left and right tuber coxae. Three paired measurements were taken with the cows placed in an insemination stall in accordance with health and safety- taking measurements when the cow was standing square/equally.

**Results** Data was analysed using a paired T-test. No significant difference was found between locomotion score and pelvic asymmetry (P=0.119) however pelvic asymmetry may be an early indicator of hindlimb lameness (left hind P=0.036, right hind P=0.038). All incidences of hindlimb lameness (left hind n=4, right hind n=4) resulted in a right pelvic asymmetry.

**Table 1** The link between pelvic asymmetry and locomotion score

| Parameter | Locomotion score 0 (n=14) | Locomotion score 2 (n=14) | S.E.M. | P. Value |
|---|---|---|---|---|
| Average left | 141.23 | 140.22 | 1.203 | 0.41 |
| Average Right | 141.08 | 139.64 | 1.294 | 0.276 |
| Difference between L/R tuber coxae height | 0.72 | 1.15 | 0.268 | 0.119 |
| Lactation no. | 2.14 | 2.07 | 0.558 | 0.899 |
| DIM | 179 | 228 | 38.4 | 0.215 |

**Conclusion** Concurring with previous studies, muscular weight bearing compensations that can be seen in lame dairy cows would lead to anatomical adaptations not solely in the hindlimbs but eventually in the pelvis, as muscles adopt the role of stabilisers and reduce their activity in limb protraction and retraction. As seen in multiparous dairy cows, weakened suspensory apparatus and abdominal muscles increase the likelihood of pelvic asymmetry the greater the DIM. Further research into three dimensional static and dynamic pelvic asymmetries is required to support our initial findings.

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**References**

Blackie, N., Bleach, E., Amory, J. and Scaife, J. 2013. Journal of Dairy Science 96, 3564-3572.
Pfau, T., Starke, S., Tröster, S. and Roepstorff, L. 2013. The Veterinary Journal 198, 498-503.
Archer, S., Bell, N., Huxley, J. 2010. In Practice 32, 492-504.

# Effect of spatial location within clamp on maize silage density, quality and mycotoxin concentration

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**Application** Understanding the impact of silage compaction on silage quality can help farmers & contractors improve feeding value.

**Introduction** Mycotoxins are toxic secondary metabolites of fungi (Jouany and Diaz, 2005) derived from both fungal activity in the field and during storage. Field fungi include *Fusarium, Alternaria* and *Diplodia*, whereas Aspergillus and *Penicillium sps*. are storage fungi (Santin, 2005). Auerbach *et al*. (1998) report that Penicillium roqueforti is the most common species found in maize silage. However, González Pereyra *et al*. (2007) found isolation frequencies of 78% and 62% of *Aspergillus* and *Fusarium* species, respectively, in maize silage, with *Penicillium* species being less frequent at 33%. Driehuis *et al*. (2008) reported that 50% of the maize silage samples collected from core, surface and moulded areas of maize silage clamps were contaminated with mycotoxins: mycophenolic acid and roquefortine C.

**Material and methods**
Core samples of maize silage were collected from three maize silage clamps located in Staffordshire and Shropshire, West Mids, England, on 6 occasions between May and August 2016. Maize silage samples (9) were taken in a 3X3 grid across the front of clamp from top left (TL) to right bottom (RB). Silage density was calculated according to sample weight/core volume. Core samples from one farm were frozen to await analysis for mycotoxin by LS/MS/MS (Alltech, Dunboyne). Silage quality measures were statistically appraised as a factorial design (SiteXDateXSpatial location) and the limited mycotoxin concentration data was regressed against silage density using Genstat 18.

**Results**
Maize silage density, lactic acid concentration and DM was found to be greatest at the bottom and centre of the clamp and least dense on the top and periphery (Table 1). Silage pH was not different at the different sampling locations (P>0.05). Regression of mycotoxin concentration against silage density found a tendency (P>0.05) for field based mycotoxins to either increase or remain constant and clamp based mycotoxins to decrease with increased silage density (Table 1).

**Table 1** Effect of clamp face sample site on maize silage density, lactate, pH and selected mycotoxin concentration

| | Table of Means | | | | | | | | | Sample Site | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | L T | L M | L B | C T | C M | C B | R T | R M | R B | *P* value | S.E.M |
| DM g/kg | 247 | 249 | 226 | 277 | 277 | 275 | 218 | 234 | 220 | <0.001 | 0.815 |
| Density kg/m$^3$ | 394.1 | 532.4 | 551.6 | 704.6 | 764.2 | 865.0 | 369.7 | 542.7 | 603.9 | <0.001 | 25.09 |
| pH | 3.98 | 3.92 | 3.96 | 3.99 | 3.94 | 3.92 | 3.98 | 3.94 | 3.95 | 0.004 | 1.39 |
| Lactic acid g/kgDM | 16 | 20.4 | 21.4 | 28.3 | 27.0 | 24.9 | 21.3 | 22.6 | 21.3 | <0.001 | 0.158 |
| **R**egression analysis: Relationship between silage density and mycotoxin concentration | | | | | | | | | | | |
| DON (µg/kg) | 941.6 | 975.9 | 0 | 9306.2 | 1134.1 | 1184.9 | 846.9 | 1343.6 | 0 | p=0.459; $r^2$=0.081 | |
| FA (µg/kg) | 45.38 | 35.64 | 0 | 203.94 | 192.68 | 150.32 | 115.18 | 100.82 | 15.89 | p=0.116; $r^2$=0.315 | |
| PA (µg/kg) | 675.26 | 470.03 | 215.84 | 619.39 | 308.96 | 200.98 | 455.92 | 450.16 | 229.82 | p=0.174; $r^2$=0.246 | |

Where: L=Left; C=Centre; R=Right; T=Top; M=Middle; B=Bottom; DON= Deoxydivalenol; PA=Penicillic acid; FA=Fusaric acid

**Conclusion** The spatial location of the samples within a clamp resulted in differences in silage density. Low density silage can lead to reduced lactic acid concentration. Mycotoxin concentration did not vary at different spatial location however, reduced silage density may result in increased mycotoxin concentrations associated with storage fungi such as PA in the ensiled product.

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**References**
Auerbach, H., Oldenburg, E. and Weissbach, F. 1998. Journal of the Science of Food and Agriculture 76 (4), 565-572.
Driehuis, F., Spanjer, M.C., Scholten, J.M. and Te Giffel, M.C. 2008. Journal of Dairy science 91(11) 4261-4271.
Jouany, J.P. and Diaz, D.E., 2005. The mycotoxin blue book (Ch. 14). Nottingham University Press. 0229.82
Santin, E. 2005. The mycotoxin blue book (Ch. 9). Nottingham University Press.

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# Temporal changes of forage fatty acid profiles under three common pasture systems in the UK

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**Application** Enhanced understanding of dynamic changes in pasture quality contributes to improved resource use efficiency, liveweight gain and meat quality, all of which ultimately result in better livelihoods of livestock producers.

**Introduction** Although it is widely acknowledged that cattle raised on pasture produce beef higher in omega-3 polyunsaturated fatty acids (PUFA) than those grown under feedlot systems (Warren *et al*., 2008), forage quality resulting from different management systems is less understood, particularly regarding its changes over time. Using a whole-farm time-series dataset from the North Wyke Farm Platform (NWFP), this study compares fatty acid (FA) composition of forages cultivated under three common pasture systems in the UK: permanent pasture (PP; unsown for 20+ years); white clover/high sugar perennial ryegrass sward (WC; akin with organic systems) and high sugar perennial ryegrass monoculture (HS; akin with regularly reseeded systems).

**Material and methods** At the NWFP, 30 Charolais x Hereford-Friesian cattle are randomly allocated to each of the three aforementioned systems annually (Orr *et al*., 2016). For 2016 and 2017 grazing seasons (May–October), pasture snip samples were collected for each system every two to four weeks from the fields currently being grazed, with forages subsequently bulked into monthly samples. Forage fatty acid content was determined using the direct saponification method (Demirel *et al*., 2004) and measured using GC. Repeated measures ANOVA was used to analyse differences in fatty acid profiles between treatments and years.

**Results** Across years, FA contents under identical treatments were comparable at the beginning of the grazing season; by the end of the season, however, all 2017 samples demonstrated higher FA contents than 2016 samples, suggesting that growing conditions and grazing patterns are likely to have affected the forage quality. The main FA groups assessed in this study showed no significant difference between treatments or sampling period, although the contents of C18:2n-6 tended to be higher from August onwards (both 2016 and 2017) and C18:3n-3 in September/October (2017 only) (Figure 1).

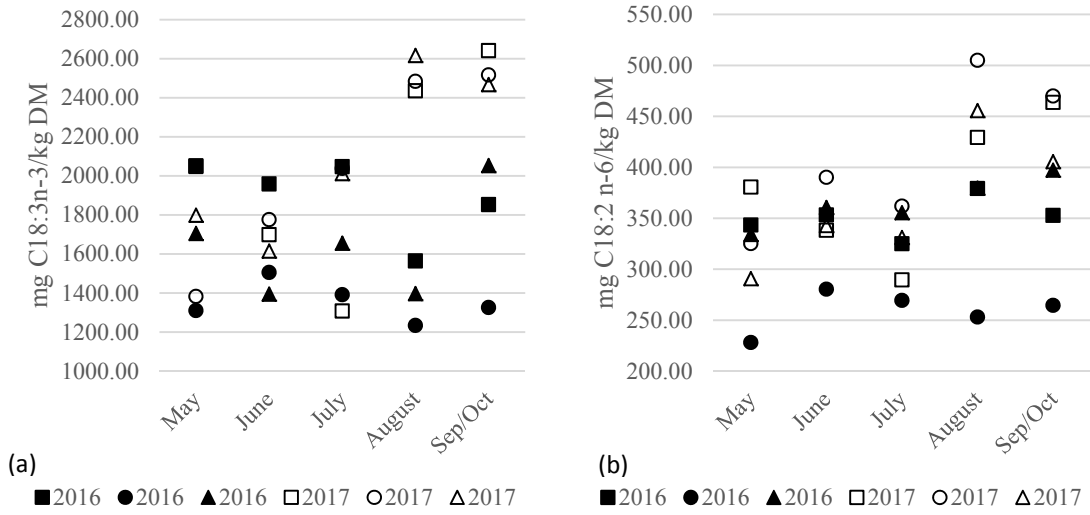


**Figure 2** Temporal changes in: (a) omega-3 FA and (b) omega-6 FA.

**Conclusion** The fatty acid composition of pasture plays an important role in grazing animals' diet, yet its differences across pasture types and time of the season are not frequently considered by commercial producers as part of farm management. The above results indicated that the three swards behaved comparably within each year, with temporal effects within and across seasons outweighing treatment effects in most cases. Impact of these swards on meat quality is reported by McAuliffe *et al*. (2018)

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**References**

Demirel, G., Wachira, A., Sinclair, L., Wilkinson, R., Wood, J. *et al*., 2004. British Journal of Nutrition 91 551-65.
McAuliffe, G.A., Takahashi, T., White, S., Hallett, K., Lee, M.R.F. 2018. Advances in Animal Biosciences 9 (1), 72.
Orr, R.J., Murray, P.J., Eyles, C.J., Blackwell, M.S.A., Cardenas, L.M., *et al*. 2016. European Journal of Soil Science 67, 374-385.
Warren, HE, Scollan, ND, Enser, M, Hughes, SI, Richardson RI, *et al*. 2008. Meat Science 78 (3), 256-269.

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# New developments in pain detection in dairy cows: Facial grimace scoring and infra-red thermography

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**Application** Observed increases in facial grimace score combined with Infrared Thermography (IRT) of the bovine foot may be sensitive and practically useful tools to advance non-invasive detection of foot problems and their associated pain levels in cows. These methods could aid efficacious treatment through objective measurement of site temperature and consequently increase herd health and welfare.

**Introduction** Intensive farming practices have resulted in an increase in acute and chronic disorders in livestock, impacting welfare and production. Lameness is a common condition in dairy cattle presenting in a variety of forms with an average prevalence of 36% within UK herds (Barker *et al*., 2010). The use of IRT for detection of physiological changes in animals has developed progressively over the last decade becoming an important tool in remote, non-invasive pain and health-status assessment in cattle alongside other physiological and behavioural measures. Increases in the temperature of individual claws in the bovine foot can take place up to six weeks before outward signs of lameness are seen making the ability to detect these changes before the onset of significant lameness and associated welfare and production effects are encountered significant. Lameness related pain is usually scored at penside using simple descriptive, numerical or visual analogue scales; this study has worked to validate the use of a facial grimace scoring system for cattle building on the work of Leach *et al.* (2012), Sotocinal *et al.* (2012), Miller *et al*. (2016) and Dalla Costa *et al.* (2016) in rats, mice and horses who developed facial grimace scoring systems based on Facial Action Coding Systems (FACS) used in humans. Action Units (AU's) for around 6 discrete facial areas/actions are described and scored using a 0 (not present), 1 (moderately present) and 2 (prominently present) scoring system. This methodology has interesting potential for use in cattle renowned for stoic behavioural mannerisms. Descovich *et al*. (2017) highlight that Facial Grimace Scales may have advantages over other proxy measures of pain in mammals as they are comprised of a few key indicators, presenting a more practical scale for real-time implementation. AHDB (2017) has identified cattle lameness as one of the most significant welfare and productivity issues in dairy farming. This study assesses the use of IRT as a non-invasive detection tool for foot problems in cattle for use in prophylactic and treatment based foot trimming to enable directed investigation thereby avoiding unnecessary invasive entry into large areas of the bovine foot. Paired with facial grimace scoring this method gives a more holistic view of the welfare state of each dairy cow presented.

**Material and methods** Data was collected fortnightly from 24 dairy cattle using an opportunistic sampling strategy in collaboration with a professional foot trimmer. Each cow was videoed walking 10 m on a level surface for mobility scoring and facial expression recording (action units recorded are ear position, orbital tightening, cheek muscle tension and nostril shape). Each cow was then restrained in a foot-trimming stall and thermographic images of each foot were taken to identify areas of potential inflammation. Each foot was then trimmed and/or treated by the foot trimmer with cause of lameness determined via this process. Still images were taken of any lesions discovered. Data collected included mobility score (using AHDB's 0-3 scale), facial expression scoring (0,1 or 2 for each action unit) while walking, head position and movement while walking, thermographic images of individual claws, photographic images of lesions if present, assessment by commercial foot trimmer of any cause of lameness. Data was then analysed using Minitab 17.

**Results** The preliminary findings are promising. Analysis of the correlation between lameness score and facial action units within the facial grimace score were highly significant (Pearson's rank correlation for lameness score and ear position 0.826, $P<0.01$, lameness score and orbital tightening 0.958, $P<0.01$, lameness score and cheek muscle tension 0.958, $P<0.01$, lameness score and nostril shape 0.958, $P<0.01$) indicating that these action units are indicative of pain. Thermographic images and foot problems revealed by examination of the foot appear to be closely correlated with lameness score (Spearman Rho 0.685, $P<0.01$). Early results indicate that IRT is sensitive to detect areas of altered blood flow in the feet of cattle indicative of sole ulcers, digital dermatitis and slurry heel. In addition thermographic imaging appears to be able to reveal problems in the foot such as developing sole ulcers, which are not yet visually apparent.

**Conclusion** Preliminary results of this ongoing study are promising in terms of the sensitivity to detect true foot problems and the specificity to avoid false positives meaning that IRT may be a valuable tool for herdsmen, dairymen and foot trimmers alike to detect mild foot problems and initiate treatment before they progress or become chronic and to assess the extent of more obvious foot problems and monitor treatment efficacy. Facial Grimace Scoring results indicate that this methodology is sensitive to detect pain in lame cattle and specific for cattle in pain versus those in no pain meaning it may have significant practical application in the agricultural industry.

**Acknowledgements** With thanks to Askham Bryan College Farm where the trial is being undertaken.

**References**

AHDB Dairy. 2017. Lameness. Available at https://dairy.ahdb.org.uk
Barker, Z.E., Leach, K.A., Whay, H.R., Bell, N.J., Main, D.C. 2010. Journal of Dairy Science 93, 932-41.

# Impact of alternative diets on the growth, development and nutrient composition of the desert locust, *Schistocerca gregaria*

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**Application** Rearing *Schistocerca gregaria* (desert locust) on feeds containing the relatively cheaper hydroponically grown barley seedlings, rather than their standard commercial diet of brassicas, does not have a detrimental effect on their growth characteristics but is more cost effective and produces insects with more desirable nutritional profiles.

**Introduction** Entomophagy, the eating of insects, is normal to approximately 2 billion people in 183 countries usually on a seasonal basis but has not been widely practiced in western societies (van Huis et al., 2017). Interest has grown in the large-scale farming of insects for human consumption as a way to help ensure future food security. *Schistocerca gregaria* is highlighted as one of the species most acceptable to consumers and is also amenable to large-scale farming (Bednarova et al., 2010; van Huis et al., 2013). One of the major costs in *Schistocerca gregaria* production is the apparent requirement for fresh vegetables, accounting for 83% (±9.5SD) of their diet on a dry matter basis (C.D.M. unpublished data), met commercially with brassicas. The aim of this study was to determine whether substitution of brassicas with hydroponically grown barley seedlings, a cheap alternative feed, affected *Schistocerca gregaria* growth, development and composition.

**Material and methods** *Schistocerca gregaria* were collected from a commercial rearing facility within 24 hours of hatching and, in cohorts of 50, randomly allocated to three feeding regimes consisting of commercial chicken feed (crude protein 160g/kg, crude fat 30g/kg) and one of the following: brassicas (control) or barley seedlings or alternating 12 hour availability of brassicas or barley seedlings (50:50). Barley seedlings were grown hydroponically without light for 12 days before being used as feed. Locusts were housed in 12L plastic containers (n=10 per feed regime, split evenly across two experimental repeats) lined with egg card to provide crawl space and to assist moulting. There was *ad libitum* access to feed with vegetable matter replaced every 12 hours. Locusts were reared at 34°C, 37.5% relative humidity until moulting to their winged adult form. Adults were removed daily, euthanized and weighed. Once all the locusts had been collected from each cohort they were oven-dried over 25 hours at 100°C for proximate nutrient analysis. Multiple cohorts were homogenised to produce enough material for proximate analysis: crude fat (Soxhlet ether extraction, n=2 per treatment) and crude protein (nitrogen auto-analyser, n=3 per treatment). Treatment groups were compared using Generalised Linear Mixed Models and Cox's regression (using the statistical package *Genstat*), significance was accepted at $P<0.05$.

**Results** There was no significant difference in rates of survival to adulthood (quasi-binomial logit-linked GLMM, $P>0.05$), developmental rate (Cox ZPH $P>0.05$), or adult mass (GLMM, $P>0.05$). Feed influenced nutritional composition: the 50:50 treatment provided the highest protein content and lowest fat deposition (Table 1).

**Table 1** *Schistocerca gregaria* nutrient composition fed on hydroponically grown barley or brassicas

| | Brassica (control) | Barley | 50:50 |
|---|---|---|---|
| Mean Fat (g/kg) | 159.6 | 203.1 | 135.3 |
| Mean Protein (g/kg) | 634.0 | 686.7 | 733.2 |

**Conclusion** Barley is a cheap and suitable alternative to partly or completely replace brassicas for commercial *S. gregaria* feeds. Partial replacement of brassicas with barley seedlings provides relatively higher protein and lower fat content which is a positive food ingredient characteristic

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**References**

Bednarova, M., Borkovcova, M., Zornikova, G., Zeman, L. 2010. Mendelnet 61 674-682.
van Huis, A., Tomberlin, J. 2017. Insects as Food and Feed. Wageningen Academic Publishers, Wageningen.
van Huis, A., van Itterbeeck, J., Klunder, H., Mertens, E., Halloran, A., Muir, G. and Vantomme, P. 2013. *FAO*, Rome.

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# *Culicoides* species composition and abundance, and new geographical species identification, on Irish cattle farms

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**Application** Putative *Culicoides* vector species of bluetongue virus (BTV) and Schmallenberg virus (SBV) may exist in abundance on Irish cattle farms. Their presence and abundance in this study demonstrates the potential for future transmission of current and novel arboviruses among livestock in Ireland.

**Introduction** *Culicoides* biting midges have been implicated in the transmission of over 50 arthropod-borne viruses worldwide including BTV and SBV. The recent unprecedented emergence of arboviruses in northern Europe, such as SBV and multiple serotypes of BTV has highlighted Europe's susceptibility to exotic arboviruses transmitted by biting midges from distant geographic regions. The abundance of suitable *Culicoides* vector species in northern Europe facilitated the rapid spread of SBV across the continent in 2012 (EFSA, 2013). There have been no studies on the *Culicoides* species composition and abundance on Irish cattle farms since the outbreak of SBV in 2012. Hence, an in-depth *Culicoides* entomology survey was conducted on sentinel farms in 2014.

**Material and methods** Ten herds in the south of Ireland which were part of a Schmallenberg virus sentinel herd surveillance study (Collins *et al*., 2016) were selected to cover as great an area of the south of Ireland as possible. Onderstepoort Veterinary Institute design ultraviolet light suction traps were used to collect insects; one trap was run overnight in the vicinity of livestock on each farm. Each site was sampled fortnightly over a period of 16 weeks (21$^{st}$ July – 5$^{th}$ November 2014) during the 2014 vector season, corresponding to eight catch collections per farm and a total of 68 night collections during the study period. Following collection, insects were frozen (-20°C) then stored in 75% ethanol. Collections were initially sorted into *Culicoides* and non-*Culicoides* spp. using a stereomicroscope. *Culicoides* were identified morphologically to species level using the keys of Campbell and Pelham-Clinton (1960) and reference wing images (The Pirbright Institute, UK). Female *Culicoides* were further classified according to physiological status into unpigmented, pigmented, gravid and blood-fed individuals.

**Results** A total of 23,929 *Culicoides* were collected. *Culicoides* were found ubiquitously across all sites; however, there was a large variation in the number of *Culicoides* collected on each farm (257 to 4,285 *Culicoides*). A total of twenty-one species of *Culicoides* were identified, including the first confirmed report of *C. clastrieri* and *C. cameroni* in Ireland, constituting new Irish records. The most abundant species identified were members of the *Culicoides obsoletus* (*C. obsoletus/scoticus*; 38%, *C. dewulfi*; 36% and *C. chiopterus*; 5%) and *Culicoides pulicaris* groups (*C. pulicaris*; 9% and *C. punctatus*; 5%) comprising 93% of all *Culicoides* collected. The remaining *Culicoides* were principally *C. achrayi* (5.1%) and *C. festivipennis* (0.8%). The number of species identified at each site varied from 10 to 15 species (mean 13). The six major *Culicoides* arbovirus vector species from the *Culicoides obsoletus* and *Culicoides pulicaris* groups were present on all ten farms. The physiological status was determined for 98% (n = 19,458) of all female *Culicoides* collected. The majority of the female arbovirus vector species collected were unpigmented (46%) and pigmented (33%), followed by gravid (12%) and blood-fed (5%). For non-vector species, gravid *Culicoides* (33%) were the most abundant, followed by unpigmented (28%), pigmented (28%) and blood-fed (10%).

**Conclusion** The most abundant *Culicoides* species identified in this study are the putative vectors of a number of arboviruses in Northern Europe. The presence and abundance of these species highlight that disease transmission could (re-)occur following a new incursion of SBV or other exotic *Culicoides*-transmitted arboviruses into Ireland.

**Acknowledgements** We acknowledge the contribution of John Heffernan (Teagasc) in assisting with sample collection. We thank the farmers for access to their farms and their data.

**References**

Campbell, J. and Pelham-Clinton, E. 1960. Proceedings of the Royal Society of Edinburgh 67, 181-203.
Collins, Á. B., Barrett, D, Doherty, M. L., Larska, M., Mee, J. F. 2016. BMC Veterinary Research 12, 234
EFSA, (European Food Safety Authority) 2013. EN-429 [22 pp.] Retrieved from http://www.efsa.europe.eu/publications. (Accessed 7 November 2017).

# Effect of anaerobic fungal treatment on the chemical composition of selected matured forages for their use in ruminant diet

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**Application** Anaerobic fungal treatment of matured tropical forages is a promising strategy to improve their nutritional value for sustained animal production during the dry season.

**Introduction** In tropical regions the available feed during the dry season is of low quality due to pronounced effects of season and maturity and this in turn affects ruminant performance and productivity. Several strategies are used to improve the nutritive value of these low-quality feeds by stimulating the rate and efficiency of the rumen microbiota. Manipulation of the rumen fermentation pathway by biological pre-treatment, especially fungal treatment, is a promising option. Anaerobic fungi have been found to play a crucial role in the degradation of lignocellulose and cellulose through enzymatic function and mechanical action weakening structural plant tissues, thus increasing the accessible site for other microbes to act upon, whereby better feed digestibility is obtained. This experiment determined the ability of two isolated anaerobic fungi from sheep rumen fluid to improve the chemical composition of selected matured forages for their use in ruminant diet.

**Material and methods** Two anaerobic fungi (*Neocallimastix* and *Orpinomyces*) strains were isolated in our laboratory from sheep rumen fluid and were cultivated on three grasses (*Brachiaria decubems*, *Andropogon gayanus* and *Lolium perenne)* and one straw (*Triticum aestivum)* for 28 days using a 4 X 3 factorial arrangement. The treatments of the forages with the fungi was done alongside the controls (i.e. untreated) and replicated four times. About 5g of each forage was weighed individually into 250 ml serum bottles and rehydrated with 10ml of the Orpin's medium. The bottles were sealed, autoclaved and those designated for fungal treatments were inoculated with 5ml of a 5 days old culture of each fungus, while those designated to serve as control were inoculated with additional 5ml of the autoclaved medium. The bottles were incubated at $39^0$C for 28 days. After incubation, the contents of each flask were filtered through tared What-man filter paper no. 1 and the residue on the filter paper was dried at $60^0$C overnight and sub-samples of each forage were then analysed in triplicate for dry matter (DM), organic matter (OM), ether extract (EE), ash, crude protein (CP), neutral detergent fibre (NDF), total phenolics (TP), total tannins (TT) and total antioxidant (TA) contents by using standard methods. The data were then statistically analysed by using two-way analysis of variance in Minitab16 software and means were separated using Tukey's post hoc test at $P<0.05$. Only the main effect of forage and fungi were presented.

**Results** The fungus reduced the DM, OM, NDF, TP and increased the ash and CP contents of the forages after 28 days of inoculation but had no significant influence on the TT and TA contents of the forages. *T. aestivum* recorded the highest DM, Ash and NDF content; *A. gayanus* recorded the highest OM and TT contents; *L. perenne* recorded the highest CP, TP, and TA contents.

**Table 1** The forage and fungal effects on the nutrient and secondary metabolite contents

| Factors | Chemical composition (g/kg DM) | | | | | Secondary Metabolites (g /kg DM) | | |
|---|---|---|---|---|---|---|---|---|
| | DM | OM | CP | Ash | NDF | TP | TT | TA |
| Forages | | | | | | | | |
| *A. gayanus* | $971.5^b$ | $962.8^a$ | $115.6^b$ | $37.2^b$ | $836.3^b$ | $10.1^b$ | $5.5^a$ | $4.6^b$ |
| *B. decumbens* | $971.5^b$ | $961.3^a$ | $59.6^c$ | $38.7^b$ | $838.6^b$ | $8.5^c$ | $4.2^b$ | $4.0^c$ |
| *L. perenne* | $970.8^b$ | $955.9^b$ | $157.5^a$ | $44.1^a$ | $752.2^c$ | $11.2^a$ | $5.1^a$ | $6.5^a$ |
| *T. aestivum* | $978.5^a$ | $954.1^b$ | $32.2^d$ | $45.9^a$ | $857.9^a$ | $7.2^d$ | $3.7^c$ | $2.7^d$ |
| SEM | 0.88 | 1.36 | 8.25 | 1.36 | 7.06 | 0.24 | 0.11 | 0.19 |
| Fungi | | | | | | | | |
| Control | $978.4^a$ | $967.5^a$ | $87.7^b$ | $32.5^b$ | $831.3^a$ | $9.9^a$ | 4.7 | 4.4 |
| *Neocallimastix* | $971.6^b$ | $953.7^b$ | $93.1^a$ | $46.3^a$ | $818.4^b$ | $8.7^b$ | 4.5 | 4.7 |
| *Orpinomyces* | $969.3^b$ | $954.3^b$ | $92.8^a$ | $45.7^a$ | $816.0^b$ | $9.2^b$ | 4.8 | 4.2 |
| SEM | 0.88 | 1.36 | 8.25 | 1.36 | 7.06 | 0.24 | 0.11 | 0.19 |

**Conclusion** The fungal treatment increased the soluble fraction (i.e. protein content) and decreased the fibre content of the mature forages over the selected inoculation time. The *Orpinomyces* appeared to have worked better for fibre reduction and increased crude protein fraction than *Neocallimastix*.

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# The inclusion of 25-hydroxyvitamin D and phytase inclusion in finisher pig diets on pig performance, bone parameters and pork quality

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**Application** A combination of phytase and 25-OH-$D_3$ in pig production has no additional benefits for improving pig performance, bone parameters or pork quality compared to offering these feed additives individually.

**Introduction** Leg weakness is a significant problem in fast-growing swine production systems and can affect the thriftiness of fattening pigs (O'Doherty *et al*., 2010). Cereal based pig diets require supplementation of inorganic phosphorus (P), as almost two thirds of the total P is present in the form of phytate, which is poorly utilised by monogastric animals. The inclusion of phytase enzymes in pig diets can reduce the anti-nutritional effect of phytate and improve the digestibility of minerals and nutrients. Another major factor affecting efficient calcium (Ca) and P absorption level for bone mineralisation is adequate vitamin D status. The supplementation of pig diets with 25-hydroxyvitamin $D_3$ (25-OH-$D_3$) has a major advantage over vitamin $D_3$ as it provides five times the biological value of vitamin $D_3$. The supplementation of phytase and 25-OH-$D_3$ has also been shown to improve Ca and P retention, which in turn may increase muscle Ca concentrations and improve post-mortem proteolysis, thereby increasing pork tenderness. Therefore the objective of this experiment was to examine the effect of phytase and 25-OH-$D_3$ in low P finisher diets on pig performance, nutrient and mineral digestibility, bone mineralisation and pork quality.

**Material and methods** The experimental design was a 2 × 2 factorial design and approved under University College Dublin Animal Ethics Committee. One hundred and twenty pigs were blocked on the basis of live weight and sex and allocated to 4 dietary treatments: (1) low P (4.8 g total P/kg, 5.1g Ca/kg ) diet (basal); (2) low P diet + phytase; (3) low P diet + 25-OH-$D_3$; (4) low P diet + phytase + 25-OH-$D_3$. Diets were formulated to be isoenergetic and isonitrogenous. Phytase (Ronozyme®) was included at concentrations of, 0 and 500 FYT (phytase units)/kg. A unit is defined as the quantity of enzyme that liberates 1 µmol of inorganic P/min from 1.5 mmol/L of sodium phytase at pH 5.5 at 37°C. The 25-OH-$D_3$ was added at an inclusion rate of 50 µg/kg/feed. Pigs were grouped in mixed gender (50:50) groups of ten/pen. Pens were equipped with single space computerised feeders to record individual feed intakes. Pigs were weighed at the beginning of the experiment day 0, 28 and day 55 (slaughter). Feed and faecal samples were analysed for dry matter (DM), ash, nitrogen (N), gross energy, Ca, P and acid insoluble ash to determine coefficient of apparent total tract digestibility (CATTD). Post slaughter metacarpal bones from the pig's front right foot were collected for bone ash, P, Ca and bone density. Following overnight chilling of the carcass, the *Longissimus thoracis* (LT) was excised for Warner Bratzler shear force (WBSF) values, cook and drip loss. Statistical analysis was carried out using PROC MIXED procedure of SAS.

**Results** Pigs offered phytase had a lower daily feed intake (2.45 kg v. 2.59 kg; $P<0.05$) and lower feed conversion ratio (2.74 kg/kg v. 2.85 kg/kg; $P<0.05$) compared to the non phytase fed pigs. There was no effect ($P > 0.05$) of phytase or 25-OH-$D_3$ inclusion on average daily gain, final body weight, carcass weight, kill-out %, back-fat depth, muscle depth or lean meat percentage. Pigs offered the phytase diets had an increased ($P<0.01$) CATTD of ash, P and Ca compared to pigs offered non-phytase diets. Pigs offered the 25-OH-$D_3$ diets had increased ($P<0.05$) CATTD of N and ash compared to pigs offered the non-25-OH-$D_3$ diets. Pigs offered the phytase diets had an increased ($P<0.05$) bone DM, ash, Ca, P and density compared to the pigs offered the non-phytase diets. There was no effect of 25-OH-$D_3$ on bone parameters. There was a significant interaction between phytase and 25-OH-$D_3$ on cook loss ($P<0.05$) and WBSF ($P<0.05$) (Table 1), Muscle from pigs offered 25-OH-$D_3$ had increased cook loss % and WBSF values compared to pigs offered the basal diet, however, there was no effect when phytase and 25-OH-$D_3$ were offered in combination.

**Table 1** Effect of dietary treatment on pork quality parameters

| | Treatments | | | | | Significance | | |
|---|---|---|---|---|---|---|---|---|
| Phytase | No | Yes | No | Yes | SEM | 25-OH-$D_3$ | Phytase | 25-OH-$D_3$ × Phytase |
| 25-OH-$D_3$ | No | No | Yes | Yes | | | | |
| Drip loss (%) | 11.7 | 10.3 | 11.4 | 11.1 | 0.672 | 0.782 | 0.216 | 0.415 |
| Cook loss (%) | 27.5 | 28.7 | 30.6 | 27.6 | 1.020 | 0.342 | 0.356 | 0.047 |
| WBSF (NT) | 39.4 | 42.7 | 45.0 | 43.4 | 1.160 | 0.011 | 0.477 | 0.043 |

**Conclusion** Overall there was no advantage of offering a combination of phytase and 25-OH-$D_3$ on pig performance, bone parameters or pork quality compared to offering these feed additives individually.

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**References**

O'Doherty J. V, Gahan D. A, O'Shea C, Callan J. J and Pierce K. M 2010. Animal 4, 1634-40.

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# The effect of litter birth order on piglet body weight and vitality measures

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**Application** Piglets born later during farrowing were more likely to have a lower vitality score but suckle sooner after birth, however this did not improve colostrum intake or piglet weight at day 1.

**Introduction** Investigating piglet birth order in litters is necessary to better understand the interactions between litter mates and its influence on performance. With increased litter size, piglets born late in farrowing have a higher risk of being stillborn or suffering hypoxia and as a result may take longer to suckle resulting in reduced colostrum intake and subsequent immunity (Herpin *et al* 1996, Cabrera *et al* 2012). If these piglets survive their performance is likely compromised. Indeed previous research suggests a higher birth order (i.e. pigs born later in the farrowing) is an indicator of reduced ability of piglets to survive one week after birth (Panzardi *et al* 2012). The objective of this study was to investigate piglet birth order, vitality and weight, as a tool to identify vulnerable piglets, with the aim to improve piglet survivability and performance.

**Material and methods** Data originated from a sow gestation trial that investigated the use of salmon oil and vitamin $D_3$ inclusion level. The study was conducted in AFBI Hillsborough between April 2016 and May 2017. Vitality measures were collected from litters from sows whose farrowings were attended (n=80). For each piglet (n=1143) time of birth, birth interval (mins) and vitality score was noted. Vitality score was assigned immediately after birth using a categorical scale as per the method of Baxter *et al* (2008), with 0 being no movement or breathing and 3 being good movement, breathing and piglet attempts to stand. Piglet sex and birth weight (kg) were also recorded, after which each piglet was marked with their birth order, placed back in their pen and observed for time to first suckle. Piglets were weighed again on day 1 to allow colostrum intake (g) to be estimated as per the method of Theil *et al* (2014). Data were analysed using Genstat (18th edition). Birth interval, birth weight, day 1 weight, time to first suckle and colostrum intake were analysed using linear mixed model methodology using REML estimation. Piglet sex was analysed as binomial distribution using generalised liner mixed model methodology. Piglet vitality score was analysed using a multilevel mixed-effects ordered logistic regression. In all analyses, sow and rep were included as random effects while birth order was included as a fixed effect.

**Results** Piglet sex did not influence birth order and birth order did not significantly affect birth interval or birth weight (P>0.05) but there was a tendency for piglets born later in the farrowing process to be gilts (P=0.091) and to be heavier (P=0.061), with an increase in birth weight of 0.004kg per piglet as birth order increased. Vitality score was affected by birth order, with an increased probability of receiving a better vitality score if born earlier during farrowing (P<0.001). Although, piglets born later during the farrowing process had a reduced time to first suckle (P<0.001; Figure 1) this had no effect on colostrum intake or day 1 weight (P>0.05).

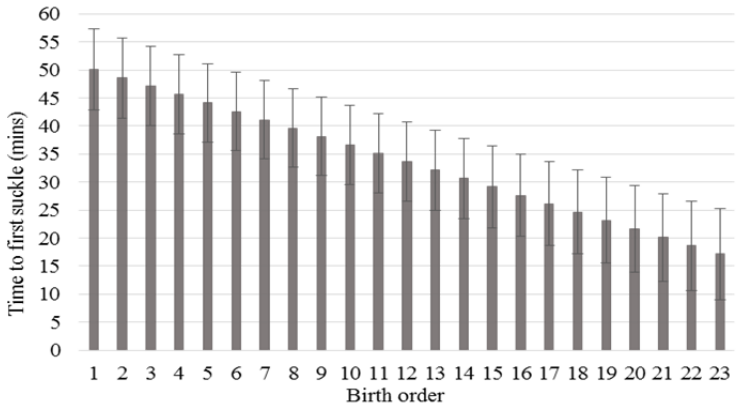


**Figure 1** Piglet birth order and mean time to first suckle (mins)

**Conclusion** Birth order significantly affected piglet vitality score and time to first suckle but this did not influence colostrum intake or day 1 weight. However, with a tendency for lower birth weight piglets to be born earlier in farrowing, they possibly benefited from access to better quality colostrum with reduced litter competition. This study shows the effect of birth order on piglet performance is unclear as although it impacts on initial piglet vitality score, later vitality measures were unaffected.

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**References**

Baxter E, *et al*. 2008 Theriogenology 69, 773-783
Cabrera, R.A, *et al* 2012 Journal of Animal Science and Biotechnology 3, 42.
Herpin P, *et al*. 1996 Journal of Animal Science 74, 2067-2075.
Panzardi A, *et al*. 2012 Preventative Veterinary Medicine 110, 206-213.
Theil PK, Flummer C, Hurley W, Kirstensen NB, Labouriau R, Sorensen MT 2014 Journal of Animal Science 92, 5507-5519.

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# Performance and nutrient utilization of White Fulani x Muturu calves fed diets containing agro-industrial by-products and browse *enterolobium cyclocarpum* leaves

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**Application** Large quantities of agro-industrial by-products are generated in Nigeria and many of them have potentials as non-convectional feedstuffs. Efficient supplementation of locally available grains or protein foliage has been demonstrated to improve rumen ecology, nutrient utilization and livestock performance.

**Introduction** Alternative feed resources and crop residues are locally available for use to increase livestock production in Nigeria. This study was conducted to evaluate the performance and nutrient utilization of yearling calves fed diets containing agro - industrial by-products and a browse plant, *Enterolobium cyclocarpum* leaves.

**Material and methods** The experiment was carried out at the Cattle Unit of the Teaching and Research Farm of the Federal University of Agriculture, Abeokuta, Nigeria. *E. cyclocarpum* leaves were harvested within the premises of the University, sun-dried and milled to 2mm sieve size. The ground leaves were included in concentrate rations at different levels. Agro-industrial by-products (wheat offal, palm kernel meal and cassava peels) were used to formulate the concentrate diets. Sixteen male cross-bred, yearling White Fulani X Muturu yearling calves, 10-11 months of age, weighing 75-80 kg were used for the 112 days study. The animals were randomly assigned to four treatment groups of *Enterolobium cyclocarpum* leaf meal (ECLM)-based concentrate diets in a completely randomised design arrangement. The treatment diets 1, 2, 3 and 4 contained 0, 5, 10 and 15 g/100g ECLM respectively. Proximate and fibre fractions' compositions of the diets were determined. The animals were each served 3kg concentrate at 09.00hours daily with *Panicum maximum* as basal diet and water available *ad-libitum*. Feed refusals were weighed each morning. Each calf was weighed at the commencement of the experiment and thereafter fortnightly to monitor weight changes during the trial. Faecal samples voided were collected during the last seven days of the digestibility trial, weighed, dried at 60$^{o}$C for 48 hours and were used in calculating the digestibility of each nutrient. Data collected included feed intake, weight gain and feed conversion ratio. The data were subjected to one-way analysis of variance (ANOVA).

**Results** Proximate composition (g/100g) of the experimental diets 1, 2, 3 and 4 indicated high dietary crude protein (17.08-17.78 CP), low ether extract (4.54-5.92) and crude fibre (15.06-17.05). Diet 4 (15g ECLM inclusion) had the highest dry matter content (90.94 g/100g) and diet 1 (0g ECLM inclusion) had the least value (90.31). Diet 1 (0 ECLM inclusion) had the highest CP value (17.78) and diet 4 (15g ECLM inclusion) the least CP (17.08). Diet 4 had the highest values for CF, EE, Ash, NDF, ADF, ADL and Cellulose while diet 1 had the lowest values. The highest weight gain ($P<0.05$) and the least feed intake occurred at 10g/100g ECLM inclusion level. Digestibility coefficients increased with increasing dietary levels of ECLM. The feed conversion ratio decreased ($p<0.05$) with increasing ECLM levels up to 10g/100g.

**Table 1** Performance characteristics of calves fed ECLM-based concentrate diets

| | 1 | 2 | 3 | 4 | SEM |
|---|---|---|---|---|---|
| Parameters | 0 ECLM | 5 | 10 | 15 | |
| Initial Weight (Kg) | 75.00 | 78.33 | 76.00 | 76.67 | 1.16 |
| Final Weight (kg) | 90.13 | 96.90 | 95.43 | 92.23 | 1.54 |
| Weight gain (kg/day) | 0.18$^{c}$ | 0.22$^{ab}$ | 0.23$^{a}$ | 0.19$^{bc}$ | 0.01 |
| Feed Intake (kg/day) | 4.24 | 4.34 | 4.18 | 4.23 | 0.03 |
| FCR | 23.67$^{a}$ | 19.73$^{bc}$ | 18.01$^{c}$ | 22.84$^{ab}$ | 0.82 |

$^{abc}$ means in the same row with different superscripts are significantly different ($P<0.05$).
Treatment 1= 0, 2= 5, 3=10 and 4= 15 g/100g ECLM inclusion levels respectively.

**Conclusion** ECLM inclusion up to 10 g/100g in concentrate diets had a positive effect on performance of yearling calves. In conclusion, *Enterolobium cyclocarpum* leaves could be used up to 10 g/100g level of inclusion as supplement with other cheap agro-industrial by-products in concentrate diets.

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**References**

Adams, M. C., Luo, J., Rayward, D., King, S., Gibson, R. and Moghaddam, G. H. 2008. Animal Feed Science and Technology 145, 41-52.
AOAC 2000. 20th ed. AOAC International, Arlington, VA, USA.

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# Effect of coconut oil on *in vitro* total gas and methane production of *Panicum maximum*

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**Application** Inclusion of coconut oil did not radically reduce gas production however it considerably lowered methane, and it will qualitatively boost the utilization of *Panicum maximum* as a major forage resource with lower fermentation losses.

**Introduction** *Panicum maximum* is a predominant forage species used for ruminant nutrition in tropical Africa due to its abundance and easy regeneration. Forages however generate more methane than concentrates during fermentation leading to loss of dietary energy (Johnson and Johnson, 1995). Coconut oil (CNO) is highly rated for methane ($CH_4$) abatement because of its rich in medium-chain fatty acids and has been found to induce reductions in methanogenesis (Machmüller and Kreuzer, 1999). The aim of this study was to assess the effect of coconut oil on the gas production of Panicum.

**Material and methods** Coconut oil was incubated at four (4) inclusion levels (0 ml/g, 1ml/g, 2ml/g and 3ml/g) with 200mg of *Panicum maximum* (substrate) as treatments for an experiment set out in a completely randomized design. The *in vitro* gas production technique employed was that described by Menke and Steingass (1988). Rumen liquor was collected from a White Fulani cattle maintained daily on *ad libitum Panicum maximum*, into warm insulated flasks, filtered through layers of cheesecloth and used as the source of inoculum. The inoculum was then mixed with sodium and ammonium bicarbonate buffer (35g $NaHCO_3$ plus 4g $NH_4HCO_3$ per litre) at a ratio of 1:2 (v/v) to prevent lowering the pH of the rumen fluid which could result in decreased activities of the microbes. 200mg of substrate, replicated six times (n=6) for each treatment and were placed into 100ml calibrated glass syringes fitted with plungers. 2 ml syringes fitted with needles were used to draw coconut oil according to the treatment allocations and infused into the glass syringes through its mouth. 30ml of the buffered inoculums was then added to each syringe containing the ground. The syringes were positioned vertically in a water bath and kept at 40°C. Blank syringes containing 30ml of the buffered inoculums only was included as control. Gas production was recorded at 0, 3, 6, 9, 12, 15, 18, 21 and 24 hours of incubation. Methane gas was determined by introducing 4ml of NaOH solution into the glass syringes and the difference in plunger levels recorded. Data obtained were subjected to one-way analysis of variance and significant means separated at $P<0.05$.

**Results** Coconut oil lowered (P=0.01) gas production during incubation, however samples incubated with 1ml/g CNO recorded gas production trend similar to 0ml/g as shown in Figure 1. Methane production was greatly lowered (P=0.01) by inclusion of CNO when the single dose 1ml/g CNO is compared to 0ml/g CNO as shown in Figure 2.

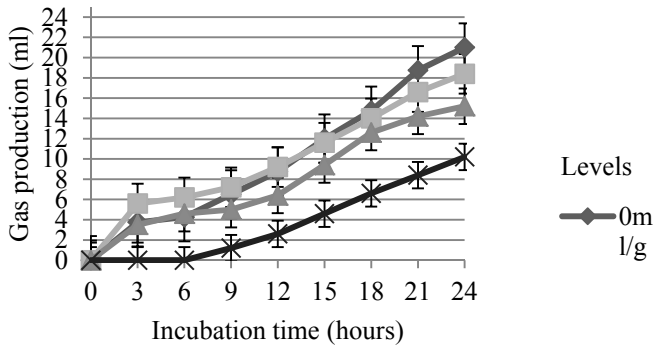


**Figure 1** Effect of coconut oil on gas production of *Panicum maximum*

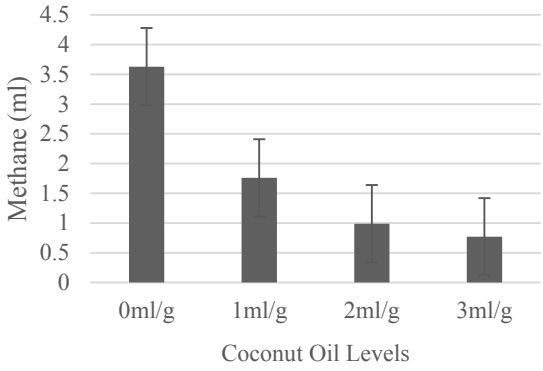


**Figure 2** Methane production of *Panicum maximum* incubated with coconut oil

**Conclusion** The addition of coconut oil reduced *in vitro* gas production from *Panicum maximum*, a mark of decreased rumen fermentation of the substrate. The study also recorded over 50% reduction in methane at the lowest level 1ml/g CNO, with further reductions as the level of CNO increased.

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**References**

Johnson, K. A. and Johnson, D. E. 1995. Journal of Animal Science 73, 2483.
Machmüller, A. and Kreuzer, M. 1999. Canadian Journal of Animal Science 79, 65-72.
Menke, K. H. and Steingass, H. 1988. Animal Research and Development 28, 7-55.

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# Nutritive value and effect of processing on microbiological quality and digestibility of black soldier fly larvae

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**Application** Black soldier fly larvae have potential as an alternative protein source for animal diets.

**Introduction** The EU is moving towards considering insects as a source of alternative protein for animal feed. In 2017, the EU made a momentous decision to legislate for the use of insects, including Black Soldier Fly Larvae (BSFL) (*Hermetia illucens*), in fish feed. However, scientific knowledge on the suitability of insects as a feed ingredient in ruminant diets is scarce (Makkar *et al*., 2014). It is also necessary to explore effective processing methods to certify insect feed as a safe ingredient in animal diets. BSFL are practical to rear and have a high protein and lipid content. This study investigated the nutritive value of BSFL and the effect of two processing techniques, High-Pressure Processing (HPP) and boiling, on microbial contamination and *in vitro* true dry matter digestibility (IVDMD).

**Material and methods** Representative samples (n=9) of BSFL were obtained from Hexafly (Co.Meath, Ireland). On arrival, the larvae were fasted for 24h before being frozen at -20°C. They were then subjected to the following treatments: Control (untreated), HPP (400Mpa for 90s/10min; 600Mpa for 90s/10min) and boiling (90°C for 10/15min). Microbiological analyses considered Total Viable Counts (TVC), Enterobacteria, Lactic acid bacteria and yeast and moulds. IVDMD was determined by incubating the larvae in buffered rumen fluid for 48h according to the DaisyII incubation method, followed by NDF digestion in an ANKOM 200 Fibre Analyser. Prior to the chemical analyses of untreated samples, the larvae were lyophilized and ground to 1mm. Samples were analysed for DM (930.15), EE (954.02), gross energy and CP, according to AOAC procedures (1990) and NDF according to Van Soest and Mason (1991). The mineral composition was measured using an energy dispersive X-ray fluorescence spectrometer. All samples were analysed in triplicate. Statistical analyses were performed using the MIXED procedure of SAS (version 9.4) (SAS Institute, 2016).

**Table 1** Chemical composition of BSFL (mean of triplicates ± standard deviation)

| | DM (g/kg as fed) | Gross energy (MJ/kg) | NDF (g/kg DM) | CP (g/kg DM) | EE (g/kg DM) | Ca (g/kg DM) |
|---|---|---|---|---|---|---|
| BSFL | 341.8 ± 0.3 | 23.7 ± 0.1 | 376.7 ± 1.1 | 441.5 ± 0.5 | 258.3 ± 0.8 | 43.6 ± 5.1 |

**Table 2** Effect of high pressure and thermal treatments of BSFL on microbial load and on IVDMD (n=9)

| | BSFL treatments | | | | |
|---|---|---|---|---|---|
| Item | Control | HPP[1] | Boiled[2] | SEM | P value |
| Microbial load | | | | | |
| TVC | 7.972 [a] | 6.911[b] | 5.624[c] | 0.1584 | <0.0001 |
| Enterobacteria | 7.654[a] | 3.512 [b] | 1.222 [c] | 0.3229 | <0.0001 |
| Lactic acid bacteria | 6.504[a] | 4.212[b] | 1.472[c] | 0.4151 | <0.0001 |
| Yeast and Moulds | 5.068[a] | 3.956[b] | 2.000[c] | 0.1303 | <0.0001 |
| Digestibility | | | | | |
| IVDMD (%) | 87.812[b] | 93.558[a] | 89.443[b] | 0.5721 | <.0001 |

SEM= Standard error of mean; [a, b, c] Means with different letters within the same row differ (P < 0.05)
[1]Average over all pressure and duration treatments; [2] average over all duration treatments

**Conclusion** BSFL have a favourable nutritive value which is rich in protein and high in calcium. Boiling was the most effective processing method at reducing microbial load whilst IVDMD improved after HHP treatment. The results of this study highlight BSFL as a potentially novel protein source for inclusion in animal diets, including ruminants, that can be sustainably and locally sourced, thus reducing dependence on imported protein crops.

**Acknowledgements** 'HEXAFLY' for providing the insect larvae for this research work

**References**

Association of Official Analytical Chemists (AOAC), Official Methods of Analysis. AOAC Washington, DC 1990.
Makkar, H. P. S., Tran, G., Heuzé, V., & Ankers, P. 2014. Animal Feed Science and Technology 197, 1–33
Van Soest, P. J. and Mason, V. C. 1992. Animal Feed Science and Technology 32, 45-53.

# Anti-inflammatory potential of *Ascophyllum nodosum* extract in an *in-vitro* colonic epithelial cell model

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**Application** An extract of *Ascophyllum nodosum* with anti-inflammatory characteristics *in vitro* has been identified. This extract has the potential to reduce inflammation in the gastrointestinal tract during stressful situations such as the post-weaning period in piglets.

**Introduction** Since the European Union implemented a ban of antibiotics as growth promoters in 2006, and more recently the phasing out of zinc oxide at medicinal levels by 2022, there is a need to identify alternatives to overcome issues faced during pig production, for example post-weaning induced inflammation and diarrhoea. In addition, concerns regarding emergence of multidrug resistant bacteria has led to a greater interest and demand for natural alternatives in agri-food production. Marine macro-algae have been considered as potential alternative supplements in swine nutrition to help maintain swine health and performance. Brown algae, such as *A. nodosum,* are a rich source of polysaccharides such as laminarin and fucoidan, which possess a variety of bioactivities including anti-inflammatory, anti-microbial and anti-oxidative qualities. Crude extracts of several marine macro-algae have been investigated in both *in-vitro* and *in-vivo* studies (Sweeney & O'Doherty, 2016). Caco-2 cells have been extensively applied as a model for initial screening of bioactives to determine potential application as an anti-inflammatory supplement for pigs. The aim of this study was to investigate the anti-inflammatory activity of an *A. nodosum* extract at various concentrations in a Caco-2 cell model.

**Material and methods** The *A. nodosum* extract contained fucoidan (27.44%) and laminarin (1.26%). Caco-2 cells were cultured in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% (v/v) fetal bovine serum, 1% (v/v) non-essential amino acids, 1% sodium pyruvate and 1% penicillin + streptomycin. Cells were maintained in vented 75 $cm^2$ flasks in a humidified cell culture incubator with 5% $CO_2$ at 37 °C. For the anti-inflammation assay, $10^5$cells/ml were seeded in a 24-well cell culture plate and maintained for 8–10 days. To induce a pro-inflammatory response, Caco-2 cells were treated with TNF-α (10 nmol/L) and used as negative control for this study. Caco-2 cells challenged with TNFα were simultaneously co-treated with the commercially available anti-inflammatory steroid, dexamethasone (10 nmol/L), as a positive control. The anti-inflammatory bioactivity was tested through a co-treatment of *A. nodosum* extracts at a final concentration of 0.5, 1.0 or 2.0 mg/ml in the presence of TNF-α. Following 24 h incubation, the media was harvested and the concentration of IL-8 in the supernatants was measured by ELISA. This experiment was carried out with technical replicates on three independent occasions (n=3). The means were compared using the PROC GLM procedure of SAS (9.4).

**Results** Compared to TNF-α stimulated cells, dexamethasone reduced IL-8 concentration by 51.92% ($P<0.05$) as expected. The greatest effect was observed following co-treatment with 2.0mg/ml of *A. nodosum* extract, which resulted in 73.66 % ($P<0.001$) reduction in IL-8 compared to TNF-α stimulated cells. Co-treatment with lower concentrations of *A. nodosum* (1.0 and 0.5mg/ml) also reduced IL-8 concentration by 57.23 and 55.86% ($P<0.05$), respectively (Table 1).

**Table 1** Effect on IL-8 production in Caco-2 cells stimulated with TNFα and co-treated with dexamethasone, *A. nodosum* for 24 h

| Treatment | IL-8 conc (pg/ml) | SD | % reduction | Significance |
|---|---|---|---|---|
| TNFα | 101.21 | ±6.90 | | |
| TNFα + Dexamethasone | 51.30 | ±4.20 | 51.92 | * |
| TNFα + *A. nodosum* 2.0mg/ml | 26.02 | ±4.88 | 73.66 | ** |
| TNFα + *A. nodosum* 1.0mg/ml | 42.25 | ±14.21 | 57.23 | * |
| TNFα + *A. nodosum* 0.5mg/ml | 43.61 | ±19.22 | 55.86 | * |

The % reduction values are relative to control (TNFα stimulated) Caco-2 cells. *P < 0.05, **P < 0.01, n=3.

**Conclusion** *A. nodosum* exhibits anti-inflammatory activity in a Caco-2 cell model. This study demonstrates its potential application as a dietary supplementation for piglets post-weaning, although further *in-vivo* research is necessary.

**Acknowledgements** Science Foundation Ireland; Project entitled: The Macroalgal Fibre Initiative: 'natural molecules naturally' (14/1A/2548).

**References**

Sweeney, T. and O'Doherty, J. V. 2016. Domestic Animal Endocrinology 56, S84-S89.

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# Overcoming challenges for Geofencing of real-time monitored grazing livestock

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**Application** Real time monitoring offers opportunities to alert managers of livestock problems, but the resolution/accuracy of GNSS poses challenges to identify the location of stock within grazing paddocks and in boundary breaches.

**Introduction** GNSS provides opportunities for tracking livestock, and new real-time communication methods provide opportunities to turn this technology from research into a powerful management aid. In larger scale wildlife contexts, Geofencing is a useful tool using a range of statistical and cartographic techniques (Wall *et al* 2014), but virtual boundaries are often defined in hundreds of metres or even kilometres. For farmed livestock, physical fences may only be millimetres in cartographic scale. Using low cost GNSS modules with low resolution accuracy, identifying when animals have breached a boundary and simply identifying correct grazing paddock are simple but important practical challenges.

**Material and methods** Prototype sheep collars containing GNSS technology and communicating to the 'cloud' via LoRaWAN (Long Range Wide Area Network) were placed on two non pregnant ewes within a flock of 12 ewes in a small field (1.85 Ha) referred to in results as the target field. The field centre was 625 metres from the nearest LoRaWAN gateway, connecting via Ethernet to the internet. The field was not in the optimum 'line of sight' for the LoRaWAN antenna, with some buildings and topography creating potential barriers. The two collars were configured to require a minimum of six GNSS satellites for location triangulation with data transmission frequency set to either one or five minute intervals over a 14-day trial period. Prior to deployment on sheep, the '1 minute' collar was deployed on the top of each corner fencepost for periods of c.1 hour. In parallel studies, a smartphone with inclusive GNSS unit using Endomondo, a leisure app for tracking runners and cyclists, was used to collect the tracks of a person covering either a circuitous 12 km route around fields (when measuring sward heights for other studies) or climbing a nearby mountain (10.1km line of sight away from the gateway and 850m above sea level). A sheep collar transmitting live via LoRaWAN on a 1 minute cycle was carried at the same time. Data was logged in Zoatracks (http://zoatrack.org/), an on-line analysis site for wildlife tracking, and mapped and analysed in terms of both spatial and temporal patterns.

**Results** Spread of mapped points from the collars at static points were in excess of 20m (10m from the centre), as a result of standard GNSS error. Sheep location 'hotspots' linked to both grazing and camping areas for the two collared sheep were in close proximity to the fence line with a high proportion of locations on the outside of the fenceline and simplistically would be allocated to another field rather than the target field. Simple analytical methods were assessed to reduce GNSS spread and reduce the potential for incorrect identification of field location. For Sheep 1, with 15976 locations at 1 minute location cycles over 14 days, it was found that 29% of locations were outside the best assessment of the fence line boundary (itself an issue). Using a 10 location rolling average visibly reduced the spread of apparent locations beyond the fence line, but 20% of locations were still outside the fence line. Adding a 10m buffer line outside the physical fence barrier line for the raw unadjusted locations reduced the proportion of 'outside' locations to 1.5%. But combining a rolling average and a 10m buffer zone reduced the number of locations not allocated to the target field to just 9 locations or 0.05% (1 in 1775) and the numbers of time-consecutive locations outside the buffered target field was zero. For Sheep 2 with a GNSS cycle of 5 minutes, with 1543 locations over 11 days, 21.3% of raw data points were outside the fenceline. By adding a 10m buffer zone, for this collar, the number of locations not allocated to target field was reduced to 1.6%. Using a 10 location rolling average alone reduced the number outside the fence line to 1.9% and a combination of rolling average and 10 metre external buffer zone effectively placed every location within the field. Communication data losses, in the absence of full line of sight, are expected to be under 10% (i.e. 9 out of 10 messages are received by the gateway). The sheep trial demonstrated that 83% of locations were made live in the ascribed one minute cycle, 90% of locations were within a 2 minute cycle. Trials by people acting as sheep surrogates provided no grossly inaccurate location data, with similar communication loss.

**Conclusion** Live sheep location can be achieved using low cost GNSS modules communicating via LoRaWAN. Data from these low resolution modules can allocate sheep to the required field but need multiple locations and some element of data smoothing and/or buffer areas. Real time location requires a run of preceding data points included in any audit. Alerts of fence line breach based on single data points would be premature and require multiple locations to avoid false negatives. To ensure that real time location is useful, these must be frequent. Application using current technology and resolution of location can support improved security, welfare, management and labour use for grazed livestock.

**Acknowledgements** SRUC and Hoofprints are grateful to Innovate UK for funding this work.

**References**

Wall, J., Wittemyer, G., Klinkenberg, B. and Douglas-Hamilton, I. 2014. Ecological Applications 24 (4), 593–601.

# Inflammatory IL-1β expression by bovine endometrial stromal and epithelial cells is mediated via the NLRP3 inflammasome

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**Application** Bovine endometritis is a pathological inflammatory immune response that occurs in a subset of cows postpartum and has major implications for both fertility and production costs. The mechanism behind the switch to a pathological inflammatory immune response is poorly understood, resulting in a lack of effective treatments for the disease. Understanding the complex immune mechanisms underlying the pathology may lead to targeted therapies and improved disease outcomes.

**Introduction** Following calving, all cows experience massive inflammatory activity in the uterus caused by the influx of bacteria and mechanisms of tissue repair. Major mediators of the inflammatory response are the endometrial epithelial and stromal cells which respond by releasing pro-inflammatory cytokines. This is a healthy inflammatory response allowing the uterus to return to a state of homeostasis (Chapwanya, A, *et al*., 2012). However, 30% of cows develop endometritis, pathological uterine inflammation that compromises fertility. Previous work by our group has identified higher levels of the pro-inflammatory cytokine IL-1β in cows that develop endometritis (Foley, C., *et al*, 2015). In the present study, we investigated roles for endometrial cells in regulating the switch from healthy to pathological inflammation and hypothesized that regulation of inflammasome activity (the complex controlling the release of the potent pro-inflammatory cytokine IL-1β) is key.

**Material and methods** Bovine endometrial tissue was isolated from non-pregnant bovine uteri at stage I of the oestrus cycle, immediately post-mortem. Epithelial and stromal cells were isolated from the uterine horn ipsilateral to the corpus luteum. Pure populations of cells were confirmed based on morphology and on positive staining for cytokeratin (epithelial) and vimentin (stromal). To investigate IL-1 production, cells were stimulated with lipopolysaccharide (LPS) for 3, 6, 12 and 24 hours, followed by nigericin (an inflammasome stimulator) for 1 hour. Levels of IL-1α, IL-1β and IL-18 mRNA were measured using qPCR and protein levels of IL-1β were analysed using western blotting and ELISA. A Kruskall – Wallis test with Dunns multiple comparison post-hoc test was used to analyse statistical differences between 2 or more groups.

**Results** We have identified that stimulation of uterine epithelial and stromal cells with nigericin in combination with LPS resulted in IL-1β expression in a time dependent manner, with protein levels peaking after 6 hours in stromal cells (>4600 pg/ml). IL-1β expression was higher in basolaterally stimulated polarized epithelial cells at 6 hours compared to apically stimulated cells (529pg/ml *vs* 385pg/ml). Bioinformatic and qPCR analysis identified that inflammasome components are conserved within the bovine genome and expressed within endometrial cells with the exception of caspase-1 of which no expression was identified within endometrial cells. Validation was performed by treatment of both endometrial stromal cells and polarized epithelial cells with an inhibitor of the NLRP3 inflammasome (MCC950) and a pan-caspase inhibitor (Z-Vad-FMK) blocked IL-1β production by stromal cells, indicating that IL-1β production is inflammasome dependent within these cells.

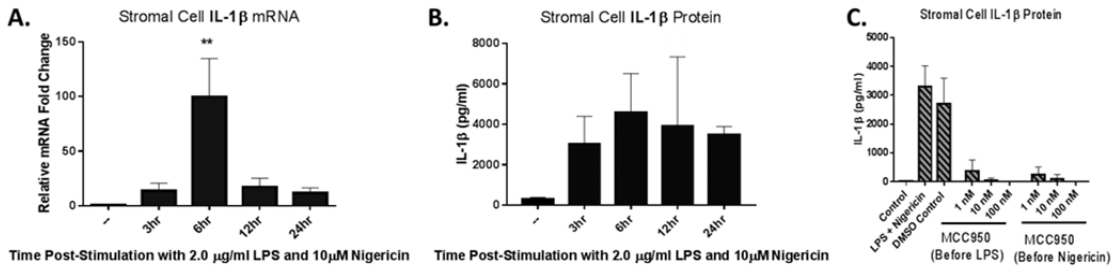


**Figure 1** (A) IL-1B mRNA levels and (B) IL-1 protein levels in stromal cells following stimulation with LPS and nigericin. (C) IL-1 levels following pre-treatment with the NLRP3 inhibitor MCC950 and subsequent treatment with LPS and nigericin in endometrial stromal cells.

**Conclusion** The data suggests a critical role for inflammasome-activated IL-1β in endometrial cell mediated inflammation. Inhibition of IL-1β by the NLRP3 inhibitor MCC950 denotes a novel target for therapeutic intervention in the treatment of the pathological inflammation associated with post-partum endometritis and warrants further investigation.

**Acknowledgements** We would like to thank the staff at Kildare Chilling Company for facilitating sample collection. This work was funded by a DAFM stimulus grant, number 13/S/472.

**References**

Chapwanya, A., *et al*., Reproduction Fertility and Development 2012.
Foley, C., *et al*., BMC Genomics, 2015

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# Bio-conversion of cereal stover by oyster mushroom (*Pleurotus* spp) affects nutritive value and rumen fermentative dynamics of complete diets

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**Application** Bio-conversion increased CP of treated sorghum and maize stover and reduced fibre components, resulting in increase in rumen fermentation.

**Introduction** Cereal stover contribute immensely to ruminant nutrition but are harvested when they are low in nutritive value. This affect rumen fermentation and productivity. Corrective strategies involve ammoniation or legume supplementation. Low cost oyster mushroom for converting stover may increase its nutritive value and the sale of mushroom would add to farm income. Therefore, the objective was to test the treatment of cereal stover with Pleurotus spp on nutritive value and rumen fermentation of stover.

**Material and methods** Sorghum (SS), millet (ML) and maize (MS) stover was randomly sampled from five smallholder farms and each cereal material pooled. A 2 x 3 arranged study was carried out. The dried material was chopped and half hydrated for 24-36 hours while the other half was used as control. The hydrated material was steamed, cooled and then inoculated with Pleurotus spp spawn. After harvesting the mushroom, spend stover was sun-dried. Complete diets were formulated in duplicates and comprised of 0.55 treated stovers (Millet = MLP; Maize = MSP; Sorghum = SSP), untreated stovers and 0.45 concentrate. Standard AOAC procedures were used for chemical composition in duplicates. Rumen DM fermentation was estimated (Ørskov and McDonald 1979) and data fitted into equation $P = a + b\ (1 - e^{-ct})$. Effective degradability (ED=bc/(c+k); Ørskov, 1992) was estimated at outflow rate of 0.03/hr ($ED_3$) or 0.05/hr ($ED_5$). *In vitro* DM and OM digestibility after 96hrs of incubation in syringes were done in duplicates after two runs. Differences due to stover, mushroom treatment (not shown) and their interactions were estimated using GLM of SAS (2002-08) at $P<0.05$ and results reported as LSD means ± sem.

**Results** Bio-conversion reduced fibre and increased OM digestibility (Table 1). Bio-conversion also increased soluble fraction and effective degradability at the two outflow rates. The greatest response for these parameters was for maize stover.

**Table 1** Nutritive value, OM and *in vitro* digestibility (g/kg DM) and *in vitro* fermentative characteristics of bio-converted cereal stover

| Forage | Ash | CP | ADF | ADL | NDF | OMD | IVDMD |
|---|---|---|---|---|---|---|---|
| Nutritive Value | | | | | | | |
| ML | 58.9$^{b}$ | 156.4$^{ab}$ | 184.2$^{c}$ | 105.9$^{d}$ | 615.9$^{d}$ | 846.8$^{c}$ | 792.5$^{c}$ |
| MLP | 48.8$^{a}$ | 150.8$^{b}$ | 173.0$^{bc}$ | 90.4$^{c}$ | 617.9$^{d}$ | 874.4$^{b}$ | 838.7$^{b}$ |
| MS | 49.6$^{a}$ | 152.0$^{b}$ | 185.0$^{c}$ | 79.1$^{b}$ | 503.4$^{a}$ | 874.9$^{b}$ | 808.4$^{bc}$ |
| MSP | 48.3$^{a}$ | 156.7$^{ab}$ | 158.3$^{ab}$ | 63.0$^{a}$ | 583.9$^{b}$ | 900.6$^{a}$ | 888.7$^{a}$ |
| SS | 53.1$^{ab}$ | 154.0$^{ab}$ | 174.1$^{bc}$ | 74.6$^{b}$ | 612.5$^{cd}$ | 890.1$^{a}$ | 790.8$^{c}$ |
| SSP | 52.9$^{ab}$ | 159.2$^{a}$ | 145.2$^{a}$ | 61.6$^{a}$ | 601.7$^{c}$ | 896.8$^{a}$ | 839.8$^{b}$ |
| SEM | 2.98 | 1.93 | 5.93 | 3.17 | 4.67 | 4.09 | 14.31 |
| | | | | | | | |
| Fermentation | | | | | | | |
| | a (%) | b (%) | c (%/hr) | $ED_3$ (%) | $ED_5$ (%) | | |
| ML | 12.2$^{c}$ | 60.5$^{b}$ | 0.020 | 36.0$^{c}$ | 29.2$^{c}$ | | |
| MLP | 20.0$^{b}$ | 60.6$^{b}$ | 0.016 | 41.5$^{b}$ | 35.0$^{bc}$ | | |
| MS | 13.9$^{b}$ | 60.3$^{b}$ | 0.028 | 42.5$^{bc}$ | 35.2$^{bc}$ | | |
| MSP | 26.3$^{a}$ | 65.4$^{a}$ | 0.020 | 52.2$^{a}$ | 44.8$^{a}$ | | |
| SS | 15.5$^{c}$ | 60.7$^{b}$ | 0.022 | 39.7$^{bc}$ | 32.9$^{c}$ | | |
| SSP | 23.7$^{ab}$ | 61.8$^{b}$ | 0.018 | 45.8$^{ab}$ | 39.2$^{ab}$ | | |
| SEM | 1.30 | 0.59 | 0.0055 | 2.12 | 1.78 | | |

**Conclusion** Bio-conversion of stover reduced fibre components and increased fermentative characteristics and *in vitro* digestibility. This is likely to increase the supply of nutrients to the animal and improve productivity.

**Acknowledgements** Funds were from The Regional Universities Forum for Capacity Building in Agriculture (RUFORUM)

**References.**

Ørskov, E. R. 1992. Protein Nutrition in Ruminants. 2nd Edition Academic Press. London.
Ørskov, E. R. and McDonald, I. 1979. Journal of Agricultural Science 92, 499-503.

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# Growth performance and *Eimeria* count of West African dwarf (WAD) goat does fed algal-biomass supplemented diets

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**Application** Innovative feeding of algal-biomass to West African dwarf (WAD) goat does could be a nutritional strategy to enhance their growth performance and health status. This improvement in turn results in increased food production.

**Introduction** Low productivity of goat does has been linked with inadequate nutrition and disease challenge (Aina *et al.* 2002). Concentrate supplementation to goats at weaning to adult stages enhanced growth performance particularly in the early and late dry season (Kawas *et al*., 2010) but the reproductive performance of WAD goats was low. No elaborate study exists on the performance of WAD goats fed an algal containing diet and how it improves their health and production performance. Therefore, this study sought to explore the effects of feeding rations with varying levels of algal biomass with a high level of docosahexaeonic acid (DHA) to WAD goat does at the growing phase before being mated.

**Materials and methods** The experiment was carried out at the Goat Unit of the Small Ruminant Section of Teaching and Research Farm, College of Veterinary Medicine, Federal University of Agriculture, Abeokuta, Nigeria during the dry season (October-December, 2016). Twelve WAD does aged 6 months (average body weight 8.5±1.5kg) were raised under an intensive management system in a completely randomized experiment. The does were divided into four treatments with three replicates each. The treatments were T0- Control, T1- 15%, T2- 30%, T3- 45% algal biomass (DHA Gold, DSM company, Netherlands). The crude protein (%) content of the diet was T0- 16.5%, T1- 16.5%, T2- 16.4%, T3- 16.4% while the energy (MJ/kg dry matter) was 12.21, 12.34, 12.40 and 12.44. The WAD does were housed in wooden floored, well ventilated, open sided individual pens covered with corrugated aluminium sheets. The animals were quarantined for 2 months and vaccination and medication was administered against viral and bacterial diseases. The animals were fed 1.5kg each of Panicum grass and Gmelina tree leaves as basal diet in the morning and 500g of concentrate diet in the afternoon (12.00GMT). Water was given *ad-libitum*. Data were collected weekly for dry matter intake, water intake, weight changes and *Eimeria* oocyst count. All data collected were subjected to a one-way analysis of variance and means were separated using Tukey test in Minitab 16 statistical software. The experimental duration (data collection) was three months.

**Table 1** Nutrient intake and Eimeria count of WAD does fed dietary algae biomass supplemented diets

| Parameters | T0 | T1 | T2 | T3 | P-value | SEM |
|---|---|---|---|---|---|---|
| Weight gain g/doe/day | $51.4^{b}$ | $52.9^{b}$ | $60.7^{a}$ | $63.1^{a}$ | 0.02 | 12.42 |
| ConcentrateDMintake g/doe/day | $99.0^{c}$ | $122.14^{b}$ | $106.71^{c}$ | $182.57^{a}$ | 0.001 | 0.03 |
| Forage DM intake g/doe/day | $582.40^{b}$ | $563.2^{b}$ | $620.8^{b}$ | $815.2^{a}$ | 0.00 | 0.58 |
| Total DM intake g/doe/day | $681.40^{c}$ | $685.34^{b}$ | $727.51^{b}$ | $997.77^{a}$ | 0.03 | 80.64 |
| Protein intake g/doe/day | $12.65^{b}$ | $14.25^{b}$ | $16.36^{b}$ | $32.46^{a}$ | 0.02 | 17.90 |
| Fat intake g/doe/day | $13.2^{b}$ | $12.89^{b}$ | $7.71^{c}$ | $20.29^{a}$ | 0.03 | 12.47 |
| Water intake litre/doe/day | $1.3^{b}$ | $2.2^{a}$ | $1.5^{b}$ | $2.5^{a}$ | 0.00 | 0.08 |
| Rectal temperature ($^{0}$C) | 38.77 | 38.93 | 38.75 | 39.90 | 0.30 | 0.88 |
| Eimeria reduction (oocyst/g) | $8900.0^{b}$ | $7600.0^{b}$ | $8200.0^{b}$ | $13000.0^{a}$ | 0.02 | 4500.12 |

$^{ab}$ means in the same rows with different superscripts are significantly different $P<0.05$, T0- Control, T1- 15%, T2- 30%, T3- 45% inclusion respectively.

The reduction in *Eimeria* count (population) for the goat does on the 45% algal biomass diet was higher ($P<0.05$) at the end of the third month of the experiment. The feed intake and weight gain of goats fed the highest level of algal biomass inclusion was highest ($P<0.05$).

**Conclusion** The growth performance and *Eimeria* parasite load of WAD goat does was improved with an increase in the level of algal-biomass supplementation.

**Acknowledgement** Prof S.A. Edwards, DSM company, Algae Research team members Federal University of Agriculture, Abeokuta, Nigeria.

**References**

Aina, A. B., Yusuf, A. O., Sogbade, L. A. and Sowande, O. S. 2002. Nigerian Journal of Animal Production 29, 89-194.
Kawas, J. R., Andrade-Montemayor, H. and Lu, C. D. 2010. Small Ruminant Research 89, 234-243.

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# Effects of by-product and linseed oil supplementation to a pasture based diet on methane production, diet utilisation and ruminal fermentation

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**Application** On a grass based diet a by-product and linseed oil concentrate can reduce methane emissions.

**Introduction** The efficacy of lipids in reducing methanogenesis is highly dependent on the chain length and degree of unsaturation (Beauchemin *et al*. 2007). Linseed oil (LO) is a polyunsaturated fatty acid (PUFA), rich in linolenic acid (C18:3) and highly efficacious in reducing methane emissions in ruminants (Martin *et al*., 2010). By-products (BP) differ from cereal grains, as usually contain lower levels of starch and higher levels of fibre and lipids. High lipid content in BP may have $CH_4$ abatement potential (Benchaar *et al*. 2013). Therefore, our experiment was designed to evaluate the effects of BP inclusion and LO on $CH_4$ production with perennial ryegrass pasture as the main dietary forage source.

**Material and methods** Dietary treatment consisted of a supplementary concentrate (factor I) containing either 35% BP (BP35) or 95% BP (BP95) with and without 6% LO inclusion (factor II), thereby culminating in a 2 (concentrate; BP 35 or BP95) x 2 (Oil; + or-) factorial arrangement. Dietary *in vitro* treatments consisted of a 50:50 perennial ryegrass based pasture (PRG) and one the four supplementary concentrates. Treatments were randomly allocated to sixteen fermentation vessels in the rumen simulation technique (Rusitec). The *in vitro* incubation trial involved of a single incubation period lasting 14 days consisting of 10 days adaption and four day sampling period. Dry matter degradation, gas production and outflow of fermentation products were measured on days 11, 12, 13 and 14. Methane production was analysed using a GC100 portable $CH_4$ reader (ADC Gas Analysis; Hoddeston, UK). In-vitro data were analysed using a mixed model ANOVA (SAS version 9.1.3).Fixed effects in the model included (*i*) BP concentrate (BP35 or BP95) and (*ii*) Oil (+ or-) and their interaction. Fermentation vessel was included as a random effect in the statistical model.

**Results** The dry matter digestibility of the concentrate treatments were: BP35 (68.9%); BP35+oil (68.7%); BP95 (57.4%) and BP 95+oil (54.4%). In general, concentrate that contained lower levels of BP had a higher digestibility parameters (DM, OM, CP, NDF and ADF) than the concentrates that contained a higher BP inclusion ($P < 0.0001$). Production of volatile fatty acids (VFA) was highest for the low BP diets (BP35; $P<0.05$) relative to the BP95. Acetic acid proportion ($P<0.05$) and acetate:proprionate ratio ($P<0.05$) was greatest in the BP95 concentrate compared to the BP35 diet. Methane production (when expressed as production per day or in terms of production per organic matter digested) was affected ($P<0.05$) by BP concentrate type and LO supplementation. Addition, of LO reduced $CH_4$ for both BP concentrate types with BP95+oil having the lowest $CH_4$ production, BP35+oil and BP95 intermediate and BP35 having the highest $CH_4$ production, respectively.

**Table 1** Effects of by-product inclusion and linseed oil supplementation on gas production; methane production; pH and fermentation pattern in the rumen simulation technique (Rusitec) system

| | Concentrate composition | | | | | *P*-value | | |
|---|---|---|---|---|---|---|---|---|
| Parameter | BP35 | BP 35+oil | BP95 | BP 95+oil | SEM | Diet | Oil | Diet x oil |
| Methane (mmol/d) | 3.92[a] | 3.23[b] | 3.03[b] | 2.65[c] | 0.098 | <.0001 | <.0001 | 0.14 |
| Methane (mmol/omd) | 0.30[a] | 0.25[b] | 0.26[b] | 0.22[c] | 0.006 | <.0001 | <.0001 | 0.003 |
| **VFA production (mmol/day)** | | | | | | | | |
| Total volatile fatty acids | 40.52[a] | 36.20[b] | 34.91[b] | 36.55[b] | 1.11 | 0.03 | 0.24 | 0.01 |
| **VFA molar proportion (%)** | | | | | | | | |
| Acetic: Propionate ratio | 2.26[a] | 2.14[a] | 2.35[b] | 2.38[b] | 0.069 | 0.03 | 0.53 | 0.30 |
| Acetic | 58.87[a] | 58.75[a] | 59.26b | 60.74[b] | 0.005 | 0.03 | 0.20 | 0.13 |
| Propionic | 16.68[a] | 16.66[a] | 16.74[a] | 15.91[a] | 0.0032 | 0.29 | 0.20 | 0.22 |
| Butyric | 14.15[a] | 13.44[a] | 12.14[b] | 12.63[b] | 0.0035 | 0.001 | 0.76 | 0.11 |

**Conclusion** Under *in vitro* conditions, the inclusion of BP and addition of supplementary LO to a concentrate supplemented grass based diet of ruminants can effectively reduce $CH_4$ production.

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**References**

Beauchemin, K.A., McGinn, S.M. and Petit, H.V., 2007. Canadian Journal of Animal Science 87(3), 431.
Benchaar, C., Hassanat, F., Gervais, R., Chouinard, P.Y., Julien, C., Petit, H.V. and Massé, D.I., 2013. Journal of Dairy Science 96(4), 2413-2427.
Martin, C., Morgavi, D.P. and Doreau, M., 2010. Animal 4(03), 351-365.

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# *In vitro* gas production of cassava peel-*Leucaena* based concentrate diets for ruminant feeding

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**Application** Increased gas production associated with the inclusion of *Leucaena leucocephala* leaves in the cassava peel-based concentrate diets means that more nutrients are available for ruminants when by-products are fed during dry season.

**Introduction** Dry season is often characterised by low animal productivity due to scarcity of fresh forage and lignification of most pastures. The use of agro-industrial by-products (AIBPs) like cassava peels has been a viable alternative to help combat this problem of feed scarcity (Mirzaei-Aghsaghali and Maheri-Sis, 2008). However, there is need to improve the quality of AIBPs if the animal performance is to be sustained. Incorporation of browse plants like *Leucaena leucocephala* into the diets of ruminant is known to help their productivity (D' Mello, 1992). The aim of this study was to assess the gas production of cassava-Leucaena based concentrate diets for ruminant feeding.

**Material and methods** Cassava peel based concentrate diets were formulated with four (4) inclusion levels of ground *Leucaena* leaves at 0% (T1), 5% (T2), 10% (T3) and 15% (T4). Other ingredients included dried brewers grain, palm kernel cake, oyster shell and salt. The experiment was set out as a completely randomised design with four (4) treatments. The *in vitro* gas production technique employed was that described by Menke and Steingass (1988). Rumen liquor was collected from four (4) White Fulani cattle maintained daily on the diets into warm insulated flasks, filtered through layers of cheesecloth and used as the source of inoculum. The inoculum was then mixed with sodium and ammonium bicarbonate buffer (35g $NaHCO_3$ plus 4g $NH_4HCO_3$ per litre) at a ratio of 1:2 (v/v) to prevent lowering the pH of the rumen fluid which could result in decreased activities of the microbes. 200mg of substrate, replicated eight times (n=8) for each treatment and were placed into 100ml calibrated syringes fitted with plungers. 30ml of the buffered inoculums was then added to each syringe containing the ground samples and were then positioned in an incubator kept at 39$^{o}$C. Blank syringes containing 30ml of the buffered inoculums only was included as control. Gas production was recorded at 0, 4, 8, 12, 16, 20 and 24 hours of incubation. Data obtained were subjected to one-way analysis of variance and significant means separated at $p<0.05$.

**Results** Inclusion of *Leucaena* resulted in increased (P=0.00) gas production up to 24 hours period of incubation from the diets. The highest gas production was obtained 15% level of inclusion in concentrate diet while the least was obtained from the control (no *Leucaena*).

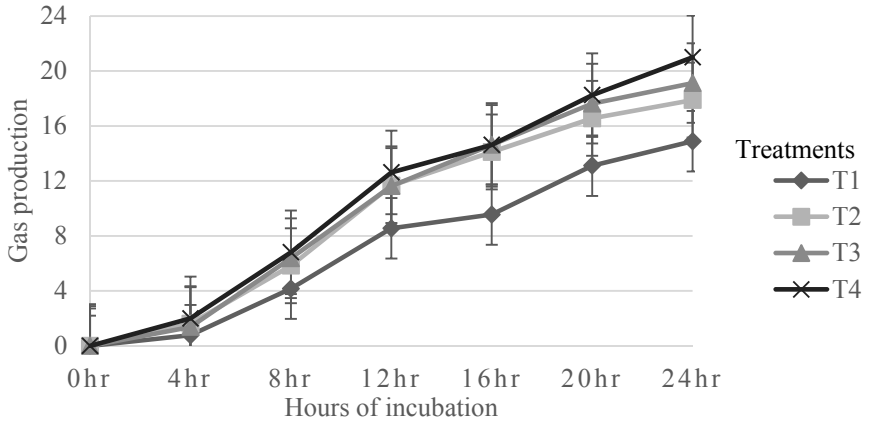


**Figure 1** *In vitro* gas production of cassava-Leucaena concentrate for ruminant feeding

**Conclusion** Inclusion of *Leucaena leucocephala* leaves increased the gas production of cassava peel based concentrate diets for ruminant feeding. The inclusion of Leucaena leaves will result in more nutrients being available for ruminants during the dry season.

**Acknowledgements** The authors gratefully acknowledge the support of ANN Department, FUNAAB, Nigeria.

**References**

D' Mello, J. P. F. 1992. Animal Feed Science and Technology 38, 237-61.
Menke, K. H. and Steingass, H. 1988. Animal Research and Development 28, 7-55.
Mirzaei-Aghsaghali, A. and Maheri-Sis, N. 2008.World Journal of Zoology 3(2), 40-46

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# No influence of prenatal treatment on plasma cortisol and rumen microbial community structure in sheep

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**Application** It is suspected from studies on other species (Rogers *et al*., 2016) that unfavourable management practices during the last part of gestation may lead to rumen microbial community alterations, disrupting physical and cognitive development, the efficient use of feed and potentially the overall health of sheep in a long term or permanent manner.

**Introduction** Prenatal stress (PNS) is known to have a long term programming effect determining physical, mental and health development of progeny (Rooke *et al*., 2017). Recent studies have also underlined the link between PNS and the colonisation/evolution of intestinal microbiota (Rogers *et al*., 2016), whereas very little is known on the effect of PNS on rumen microbiota colonisation. The aim of this project was to investigate the effect of PNS on the structure of the rumen microbiota community in the offspring, at a mature state.

**Material and methods** The samples for this project were collected from 64 lambs whose dams (Scottish Mules) were separated into control, stress and positive management groups for the last 8 weeks of gestation. The stress group was exposed to different stressful events and was allowed smaller living space, whereas the positive group was given more pen space and enhanced feed. The control group was reared under standard farming conditions. The female lambs were fed on a hay and concentrate diet for 2 weeks before slaughter, whereas the males had been on a grass and concentrate diet until slaughter. As diet is a determinant of microbial diversity in the rumen, analyses were done separately by gender. Samples of rumen liquid digesta and plasma for cortisol were collected at slaughter at 8 (± 9 days) months of age. Thirty-five rumen digesta samples were sequenced, representing the three treatment groups: controls (n=12), stressed (n=10) and positive treatment (n=13). Microbial DNA extraction was carried out (Yu and Morrison, 2004) and sequencing was performed for the V3-V4 region. The reads were processed using the Deblur tool (Amir *et al*., 2017) in Galaxy; cortisol in blood plasma was measured by ELISA. Statistical analyses were performed on the R statistical package (R Core Team, 2016) and using GenStat (one-way ANOVA for maternal treatments). In order to explore the relationship between cortisol and the microbial community ANOVA and regression analyses were performed, whereas when investigating the potential influence of cortisol on individual OTUs, a Constrained Principal Component Analysis (CPCA) was performed. The likelihood of specific OTUs being present with low and high cortisol was examined using Indicator Value analysis (IndVal). Significance was declared when $P < 0.05$.

**Results** No differences in plasma cortisol levels were observed between the treatment groups (ANOVA, females: $P = 0.491$, males: $P = 0.583$). The Shannon, Simpson, Evenness and Richness diversity Indices, investigated separately for both sexes, showed no significant differences according to treatment (Kruskal-Wallis for females and males respectively: Shannon P= 0.561/ 0.717, Simpson P = 0.692/ 0.911, Richness P = 0.804/ 0.184, Evenness P = 0.859/ 0.849). The CPCA analyses suggested that certain bacterial species do not cluster when plotted with cortisol as a constraining factor, therefore, it could be speculated that they are influenced by cortisol. The median cortisol concentration was used to classify high and low values. According to the IndVal, a number of species have a higher likelihood of being present with lower cortisol values (Females, n = 20 and Males, n = 3) or higher cortisol levels (Females, n = 6 and Males, n = 12), with Specificity or Fidelity equalling 1.

**Conclusion** No difference was observed in microbial diversity between the three maternal treatment groups, which is coherent with the cortisol analysis. Despite this, when data was investigated independent of group (median of cortisol), particular OTUs appear to have an increased likelihood of being present when cortisol levels are high, and others when cortisol levels are low. Further investigation would be needed to assess if these findings are reproducible and whether these species have a functional role within the rumen community or if their presence could be used as an indicator of stress.

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**References**

Amir, A., McDonald, D., Navas-Molina, J. *et al*. 2017. mSystems. 2, e00191-16.
R Core Team 2016. R Foundation for Statistical Computing, Vienna, Austria.
Rogers, G. B., Keating, D. J., Young, R. L., Wong, M-L., Licinio, J, Wesselingh, S. *et al*. 2016. Mol Psychiatry 21, 738–748
Rooke, J., Gareth, A., Dwyer, C., Rutherford, K. M. D. 2017. Animal Welfare 26, 403-415.
Yu, Z. and Morrison, M. 2004. BioTechniques 36, 808–812.

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# Comparison of rolled barley and oats as supplements to grass silage for finishing beef cattle

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**Application** Under the conditions of this experiment, rolled oats had a feeding value comparable to that of rolled barley.

**Introduction** Deficiencies in nutrient supply for cattle fed grass silage-based diets are usually rectified by concentrate supplementation (McGee, 2005). Cereals make up a substantial proportion of animal feedingstuffs used in Ireland (Drennan *et al*., 2006) and elsewhere. However, there is little published information on the relative performance of cattle offered rolled oats compared to barley, especially when offered as a supplement to grass silage (Huuskonen, 2009). The objective of this study was to determine the effects of replacing rolled barley with rolled oats in a supplement on intake, growth, carcass and selected meat quality traits of beef cattle offered grass silage.

**Material and methods** Late-maturing breed steers (initial live weight, 443 kg, SD 38.5) were blocked by sire breed and weight, and from within block randomly assigned to one of two (n=12) concentrate treatments: barley-based 'control' (862 g rolled barley, 60 g soya bean meal, 50 g molasses, and 28 g minerals and vitamins/kg fresh weight) and oats-based (853 g rolled oats, 70 g soya bean meal, 50 g molasses, and 27 g minerals and vitamins/kg fresh weight) concentrate rations. Concentrates were prepared as coarse mixtures. Steers were individually offered 4.0 kg dry matter (DM) of the respective concentrates, in two feeds daily, as a supplement to grass silage (DM digestibility, 713 g/kg; crude protein 102 g/kg DM) offered *ad libitum* during a 134 day finishing study. Animals were weighed at the beginning and end of the study, and every 14 days throughout. Ultrasonic fat and muscle depth were measured at the beginning and end of the study. Post-slaughter, carcass weight and, carcass conformation and fat score were determined. At 48 h *post-mortem*, subcutaneous fat depth and the lightness (L*), redness (a*) and yellowness (b*) of subcutaneous fat were measured. After 72 h *post-mortem* pH, drip loss and colour (L*, a*, b*, after 1 h bloom) of the *longissimus thoracis* muscle were determined. Data were statistically analysed using ANOVA with terms for treatment and block in the model. Initial weight was included as a covariate.

**Results** Replacement of barley with oats in the concentrate supplement had no effect (P>0.05) on average daily live weight gain, slaughter weight, silage DM intake, feed conversion ratio, carcass traits (Table 1) or ultrasonic measures of body fat and muscle gain. Muscle and subcutaneous fat 'L*', 'a*', 'b*', saturation or hue values did not differ (P>0.05) between the two concentrates. There was no difference (P>0.05) between treatments in muscle pH and drip loss.

**Table 1** Effect of replacement of rolled barley with rolled oats on dry matter (DM) intake, average daily live weight gain (ADG), feed conversion ratio (FCR) and carcass traits of finishing steers offered grass silage

| | Barley | Oats | s.e.m. | P-value |
|---|---|---|---|---|
| Slaughter weight (kg) | 570 | 571 | 7.0 | n.s. |
| ADG (kg) | 1.03 | 1.03 | 0.057 | n.s. |
| Silage DM intake (kg/day) | 5.1 | 5.4 | 0.15 | n.s. |
| FCR (kg DM/ kg ADG) | 8.9 | 9.2 | 0.39 | n.s. |
| Carcass weight (kg) | 328 | 325 | 5.7 | n.s. |
| Kill-out proportion (g/kg) | 564 | 560 | 7.2 | n.s. |
| Carcass conformation score (1-15) | 9.1 | 8.6 | 0.43 | n.s. |
| Carcass fat score (1-15) | 7.6 | 7.3 | 0.30 | n.s. |
| Subcutaneous fat depth (mm) | 5.6 | 4.7 | 0.57 | n.s. |

**Conclusion** Rolled oats can replace rolled barley in a concentrate supplement to high-digestibility grass silage without negatively affecting performance or selected meat quality traits of beef cattle.

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**References**

Drennan, M. J., McGee, M. and Moloney, A. P. 2006. Irish Journal of Agricultural and Food Research 45, 135-147.
Huuskonen, A. 2009. Livestock Science 122, 53-62.
McGee, M. 2005. In: Silage production and utilisation (ed. Park R S and Stronge M D). Proceedings of the XIVth International Silage Conference, Belfast, Northern Ireland. Wageningen Academic Publishers 51-64.

# Detection of differentially expressed genes in the *M. longissimus thoracis et lumborum* from full-sibling lambs divergent for fatness

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**Application** Functional polymorphisms in genes identified as differentially expressed and associated with fatness will be useful for genomic selection for carcass and meat quality traits in breeding programmes.

**Introduction** Increased lean meat yield and overall faster growth rate are deemed to be economically important traits with regard to lamb production. Long-term selective breeding for enhanced lean growth has reduced intramuscular fat levels with subsequent detrimental effects on eating quality (Hopkins *et al.,* 2005). One of the main factors affecting lamb-eating quality is fat content, which is positively correlated with juiciness, flavour, tenderness and overall acceptability (Fernandez *et al.,* 2009). Fat is also a source of energy, contributes to the absorption of fat-soluble vitamins (A, D, E, and K) and is a structural component of many hormones. The aim of this study was to identify a panel of functional candidate genes associated with lipid metabolism that are differentially expressed in the *M. longissimus thoracis et lumborum* (LTL), using a divergent full-sibling model.

**Material and methods** Sixty-four pairs of twin lambs were used;they were sired by rams from 1 of 3 terminal-sire breeds (Charollais (n = 20), Suffolk (n = 21) and Texel (n = 23)). All animals were slaughtered (at commercial slaughter weight) by electrical stunning followed by exsanguination; samples (2 g) of M. *longissimus thoracis et lumborum* were collected and stored in RNAlater (Ambion Inc., Austin, TX) within 15 min post-mortem. Total RNA extraction from the LTL, Quantity ($A_{260/280}$) and quality, cDNA synthesis, and quantitative real-time PCR were performed according to the methods described by Alam *et al*. (2012). All RNA samples with $A_{260/280}$ ratios ≥2.0 were used for further analysis. Carcasses were classified for fatness and conformation, and subcutaneous fat depth over the 12$^{th}$ rib was measured on the cold carcass. Data on carcass fat score and fat depth were adjusted for carcass weight, slaughter date and litterID. The adjusted data (residual fat depth and residual fat score) were combined (Proc PRINCOMP; SAS 2012) to yield an index of within-litter divergence for fatness. The most divergent pairs (n=7) of twin lambs were identified, each yielding 1 HIGH and 1 LOW lamb. The genes evaluated were chosen from a panel of 63 whose expression levels had been quantified using gene-specific primers and qPCR; the software package qbase+ (Biogazelle, Belgium) was to calculate normalized relative quantities (NRQ) of each gene. Gene expression values (mean and variance) for the LOW and HIGH groups were compared for genes (n = 10) identified as exhibiting significantly increased within-litter variation in expression level, based on the CV. Using an experimentwise error rate of 0.05.

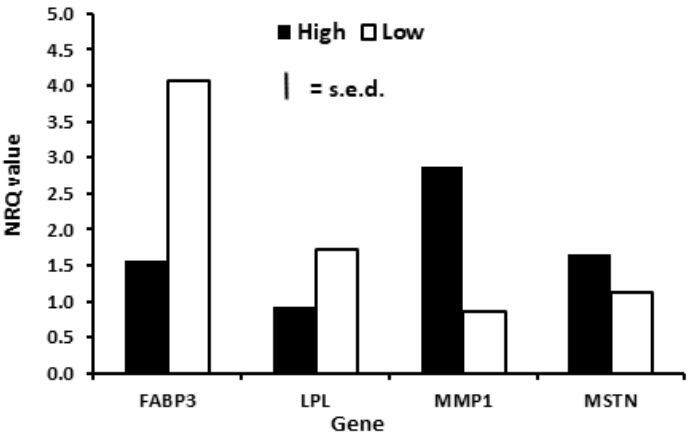


**Figure 1** Mean NRQ values for genes with DE for fatness (vertical bars represent s.e.d. between High and Low groups.

**Results** Four genes (Figure 1) were identified as differentially expressed between the HIGH and LOW fatness animals (*FABP3* and *MMP1*, P < 0.001; *LPL*, P < 0.01; MSTN, P =0.05). The expression levels of *MMP1 MSTN* were upregulated in the HIGH fatness animals, while expression levels of *FABP3* and *LPL* were higher in the LOW fatness animals.

**Conclusion** Four genes were differentially expressed between twin lambs divergent in fatness. Functional polymorphisms in the promoter region of these genes are currently being invested.

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**References**

Alam, T., Kenny, D. A., Sweeney, T., Buckley, F., Prendiville, R., McGee, M and Waters, S. M. 2012. Physiological Genomics 44, 198–209
Fernandez, X., Monin, G., Talmant, A., Mourot, J. and Lebret, B. 1999. Meat Science 53, 59-65.
Hopkins, D. L., Hegarty. R. S., Farrell, T. C. 2005. Australian Journal of Experimental Agriculture 45(5), 525–533.

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# Pigs that are divergent in feed efficiency do not differ in small intestinal architecture

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**Application** This study provides a better understanding of the mechanisms influencing differences in feed efficiency in pigs.

**Introduction** Improving feed efficiency is a key goal of the pig industry as feed costs make up over 70% of the total costs of production (Patience *et al*., 2015). Residual feed intake (RFI) is a measure of production efficiency defined as the difference between the observed and expected feed intake of an individual based on growth and backfat (Patience *et al*., 2015). However, it is a trait influenced by a variety of biological and physiological traits including digestion, metabolism, body composition, physical activity and thermoregulation (Herd and Arthur, 2009). Previously, more efficient low RFI (LRFI) pigs having an increased ileal digestibility of gross energy (GE), and an improved total tract digestibility of gross energy, nitrogen and dry matter compared to the inefficient high RFI (HRFI) pigs. These improvements were partly attributed to the LRFI pigs having higher relative gene expression levels of fatty acid binding transporter 2 (FABP2), the sodium/glucose co-transporter 1 (SGLT1), the glucose transporter GLUT2 and the enzyme sucrase–isomaltase (SI) in the jejunum (Vigors *et al*., 2016). However, while changes in nutrient transporter gene expression have been attributed to influencing changes in nutrient digestibility, our understanding of the influence of changes in small intestinal architecture are still not fully understood. Increased villus height or crypt depth would increase the surface area available for nutrient absorption potentially influencing the improvements in nutrient digestibility in LRFI pigs. Therefore, the objective of this study is to examine the small intestinal morphology of pigs divergent in RFI.

**Material and methods** Male pigs (Large white x Landrace x PIC337 boars; PIC Genetics) (92 days old (d.o.), body weight (BW) 41.35 kg (SD = 4.36)) were fed a standard finishing diet for a 43-day recording period prior to slaughter, to evaluate feed intake and growth to calculate residual feed intake. When pigs were 146 d.o., animals designated HRFI (n=12) and (LRFI) (n=12) (average weight 93.26 kg, SEM 2.37 kg) were slaughtered to collect tissue for histological analysis. On removal of the digestive tract, sections of the jejunum (60 cm from the stomach) were excised and fixed in 10 % phosphate-buffered formalin. The preserved segments were prepared using standard paraffin-embedding techniques. The samples were sectioned at a 5µm thickness and stained with haematoxylin and eosin. Villous height and crypt depth were measured on the stained sections (10 x objective) using a light microscope fitted with an image analyser (ImagePro Premier 9.2; Media Cybernetics). Measurements of fifteen well-oriented and intact villi and crypts were taken for each segment. Villous height was measured from the crypt–villous junction to the tip. Crypt depth was measured from the crypt–villous junction to the base. Results are expressed as mean villous height or crypt depth in µm. Data were analysed as a complete randomized design experiment with RFI as the main effect.

**Results** The RFI groups did not differ in villous height, crypt depth or the villous height: crypt depth ratio in the jejunum ($P > 0.10$).

**Table1** Measurements of jejunal architecture

| Measurement | High | Low | Std Err | *P* value |
|---|---|---|---|---|
| Villus height (µm) | 252.81 | 236.59 | 11.05 | 0.3113 |
| Crypt depth | 139.85 | 139.61 | 7.98 | 0.983 |
| Villus: crypt ratio | 1.84 | 1.73 | 0.088 | 0.3844 |

**Conclusion** The results from this study suggest that changes in the small intestinal architecture are not involved in differences in efficiency between RFI groups. While needing to be confirmed in a large cohort, the lack of effects in this study in combination with the results of Vigors *et al*. (2016), suggest increases in nutrient transporter gene expression are more important in influencing changes in nutrient digestibility than changes in small intestinal morphology in LRFI pigs.

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**References**

Herd, R. M., and Arthur, P. F. 2009. Journal of Animal Science 87, E64-E71.
Patience, J. F., Rossoni-Serao, M. C. and Gutierrez, N. A. 2015. Journal of Animal Science and Biotechnology 6, 33.
Vigors, S., Sweeney, T.,O'Shea, C. J., Kelly, A. K. and O'Doherty, J. V. 2016. Animal 10, 1848-1855.

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# Effect of *Acacia tortilis* leaf meal inclusion level on feed intake, digestibility and live weight gain of yearling male Boer goats fed *Avena sativa* grass hay-based diet

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**Application** *Acacia tortilis* leaf meal has potential of being used as a protein supplement during the dry season when goats depend on low-quality roughage such as *Avena sativa* hay. There is, also, some evidence that it reduces methane emission in ruminants.

**Introduction** Goats are economically, nutritionally and culturally very important to the people of Limpopo Province in South Africa. However, their productivity is limited by scarcity of high quality feed, especially during the long dry season. There is need to identify more nutritious feed to alleviate the prevailing nutritional problems during the dry season. Acacia species such as *Acacia tortilis* (*A. tortilis*) are well-adapted to drought and are identified as suitable protein supplements during the dry season (Brown and Ngambi, 2017). Utilization of *A. tortilis* is restricted by the presence of secondary plant compounds such as condensed tannins (Brown *et al*., 2016). Intake of condensed tannins by ruminants may depress feed intake and diet digestibility and hence adversely affect production. The *A. tortilis* leaf meal inclusion levels for optimal intake, digestibility and performance of Boer goats are not available. The aim of this study was, therefore, to determine the effect of *A. tortilis* leaf meal inclusion level on productivity of Boar goats fed *Avena sativa* grass hay-based diet.

**Material and methods** The study was conducted at the University of Limpopo Experimental farm in August 2016. Fresh leaves of *A. tortilis* were harvested at the farm in June, 2016. *Avena sativa* grass hay was used as a basal diet. The grass is well grazed during summer and is suitable for hay making. Twenty-four yearling male Boer goats with an average initial live weight of 23 ± 2 kg were ear-tagged and allocated, in a completely randomized design, to four dietary treatments containing *A. tortilis* leaf meal inclusion levels of 10, 15, 20 or 30% of the total diet. These inclusions include low and high tannin-levels as indicated in the literature. The animals were housed in individual metabolic cages during the study period. The goats were fed *ad libitum*, allowing a 15% refusal of each diet. The experiment lasted for 28 days. Feed intake was measured throughout the study period. Faeces were collected from each goat starting on day 21 before feeding for a period of seven successive days. From the food consumed and faecal matter secreted, apparent digestibility of the nutrients was calculated. Methane emissions were measured using a hand-held methane detector. All data on feed intake, *in vivo* digestibility, live weight gain and methane emission by goats were analysed using the GLM procedures of SAS (SAS, 2010) with diet as fixed effect. Fisher's least significant difference (LSD) test was used for mean separation where there were significant differences ($P < 0.05$).

**Results** *Acacia tortilis* leaf meal inclusion level had no effect ($P > 0.05$) on body weight of goats. However, *A. tortilis* leaf meal inclusion improved ($P < 0.05$) dry matter and crude protein intake and crude protein digestibility of goats. The fodder also reduced ($P < 0.05$) methane emission in goats.

**Table 1** Effect of *Acacia tortilis* inclusion on performance and methane emissions in yearling male Boer goats

| | Diet | | | |
|---|---|---|---|---|
| Variable | $AT_{10}$ | $AT_{15}$ | $AT_{20}$ | $AT_{30}$ |
| DM intake (g/d) | $402^{ab}$±18.623 | $353^{b}$±19.250 | $424^{ab}$±18.924 | $466^{a}$±18.767 |
| CP intake (g/d) | $30.98^{bc}$ ±1.573 | $28.51^{c}$ ±1.626 | $35.84^{b}$ ±1.599 | $42.93^{a}$ ±1.586 |
| CP digestibility (cm) | $0.28^{b}$ ±0.267 | $0.45^{a}$ ±0.096 | $0.13^{c}$ ±0.049 | $0.19^{bc}$ ±0.033 |
| BW (kg) | 23.50±1.146 | 22.49±1.758 | 24.88±1.148 | 24.00±1.363 |
| $CH_4$ emission (ppm-m) | | | | |
| Before | $22.00^{a}$ ±5.033 | $22.00^{a}$ ±4.000 | $21.67^{a}$ ±4.410 | $22.33^{a}$ ±2.333 |
| After | $11.50^{b}$ ±0.144 | $12.88^{b}$ ±1.516 | $12.42^{b}$ ±1.244 | $13.42^{b}$ ±0.333 |

[a, b]: Means with different letters in the same column are significantly different ($P < 0.05$)

**Conclusion** *A. tortilis* leaf meal inclusion improved crude protein intake and digestibility and methane emission by goats and thus has potential to be used as a protein supplement when low-quality roughage is fed as a basal diet.

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**References**

Brown, D., Ngambi, J. W. and Norris, D. 2016. Indian Journal of Animal Research 50, 41 – 47.
Brown, D. and Ngambi, J. W. 2017. Tropical Animal Health and Production 49, 1051 – 1057.

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# Temporal development of the rumen microbial community in beef calves from birth to post weaning

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**Application** The data generated in this study will enhance our knowledge on the temporal dynamics of the rumen microbiome in early life as well as identifying potential windows of opportunity for dietary manipulation in the young calf.

**Introduction** Establishment of the ruminal microbial community in the young calf is essential for development of rumen function, and a smooth transition from milk to solid diets. There is some evidence that modification of the rumen microbiota in early life may lead to changes which persist into adulthood, possibly allowing for the redirection of rumen microbial metabolism towards more energetically efficient pathways (Abecia *et al*., 2010, Yanez-Ruiz *et al*., 2010). However, the optimal "window of opportunity" for intervention remains unknown (Yanez-Ruis *et al*., 2015). Thus the objectives of this study were to assess taxonomic and functional progression of the rumen microbiome from birth to post weaning.

**Material and methods** Ninety-three Aberdeen Angus cross heifers were used in this study. Oestrous cycles were synchronised and the heifers were artificially inseminated with semen from a single Aberdeen Angus bull, yielding 66 viable pregnancies. Foetal sex was determined at day 100 of gestation. Heifers were managed similarly on two farms until parturition. Heifers were blocked into four calving replicates based on calf gender and calving date. Calves were assigned to one of 7 groups, with balanced numbers of males and females in each, of which 5 were used in the present study; D7; n = 8, D14; n = 9, D21; n = 9, D28; n = 9, or D96; n = 9. Calves remained with their dam for 48h and were penned individually thereafter. Calves were offered 5L (13.5% solids) milk replacer daily and had free access to calf starter from D7. Calves in the D96 group were offered hay and concentrates *ad libitum* post-weaning. Samples of rumen solid and liquid digesta and a section of the rumen wall were collected immediately following euthanasia for each treatment group. Amplicon libraries targeting the V4 hypervariable region of the bacterial 16S rRNA gene were prepared from genomic DNA extracts and sequenced using an Illumina MiSeq. Taxonomy was assigned using QIIME, and statistical analyses were performed in R.

**Results** Nonmetric Multidimensional scaling (NMDS) plots showed a clear separation of samples based on age, with sub-clustering based on location also evident (Fig. 1). Solid and liquid samples from each timepoint clustered closely together, indicating that there were no broad differences in bacterial composition among the two fractions in early life (Fig. 1). Age had a major effect on microbial structure from D7-D21 ($P < 0.05$), but there was no statistically significant difference between 3- and 4-week old calves, indicating a stabilisation of the microbial community (Fig 1). D96 animals had a different microbiota profile from the younger calves, probably due to increased consumption of forage ($P < 0.05$) post weaning.

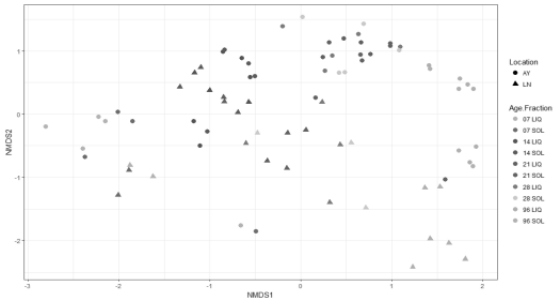


**Figure 1** NMDS based on Bray Curtis dissimilarity matrix.

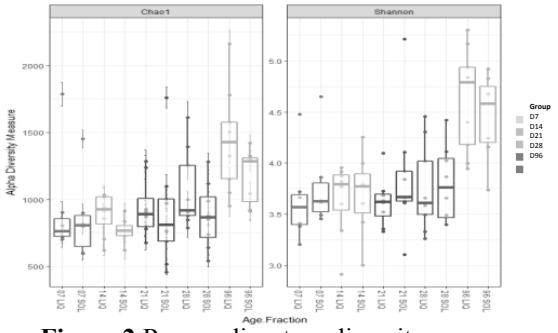


**Figure 2** Rumen digesta α-diversity measurements.

α-diversity indices were stable across age groups for the first four weeks of life (Fig. 2), but weaned animals (D96) had more diverse microbial communities than the younger animals ($P < 0.05$). Differential abundance analysis underlined these temporal dynamics in microbial composition, with large shifts in microbial composition throughout early life. Bacteroidetes, Firmicutes, Proteobacteria and Actinobacteria were the dominant bacterial phyla throughout life, but their abundances changed dramatically with age ($P < 0.05$). D7 animals had a large number of rare taxa, not associated with the normal rumen microbiota. Known fibre degraders were detected within the first week, and the introduction of solid feed induced significant increases in Short Chain Fatty Acid (SCFA) producing bacteria, potentially contributing to rumen epithelial development.

**Conclusion** These findings highlight the dynamic nature of the rumen microbiota in early life, and show that the rumen bacteriome becomes relatively settled by week 3-4 of life, but changes significantly again during the post-weaning period.

**References**

Abecia, L., *et al*. 2013. Journal of Animal Science 91(10),4832-4840
Yanez-Ruiz, D. R., *et al*. 2010. FEMS Microbial Ecology 72(2), 272-278
Yanez-Ruiz, D. R., *et al*. 2015. Frontiers in Microbiology 6, 1131.

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# Evaluation of linear type traits among lactating beef and dairy beef crossbred suckler cows

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**Application** Assessment of linear type traits of lactating beef and dairy x beef (DBX) crossbred suckler cows at pasture can be used to predict longevity as well as health and management issues such as locomotion, lameness and udder health.

**Introduction** Functionality is a major factor that must be considered when selecting suckler cows to retain within the breeding herd. The practice of linear type classification has been extensively used to assess an array of functional traits such as locomotion, lameness, docility, milkability and mastitis, or to predict longevity (Veerkamp *et al.* (2002). A divergent cow phenotype exists within the national suckler herd due to the two contrasting replacement strategies available to Irish beef producers – females sourced from within the suckler herd or sourced from the dairy herd (McGee, 2012). Identifying whether differences between beef and dairy x beef (DBX) cows exist in terms of linear scores and functionality may indicate the suitability of the replacement strategy to an Irish pasture based system. Consequently, the objective of this study was to determine if differences in linear type traits existed between lactating beef and DBX suckler cows.

**Material and methods** This study was carried out at Teagasc Grange, Dunsany, Co. Meath, Ireland in 2016. Data were available from 30 primiparous and 83 multiparous spring calving cows; 50 beef and 63 DBX cows. Animals were sired by either Aberdeen Angus or Limousin bulls. All cows calved for the first time at 24 months of age and the mean calving date was 22 March 2016. Cow live weight and body condition score (BCS; 0 to 5) were recorded along with an assessment of locomotion and a series of linear type traits by visual assessment of a trained technician on 12 October 2016. A list of the linear variables assessed and the scale used are listed in Table 1. Data were analysed using mixed models in PROC HPMIXED (SAS Inst. Inc., Cary, NC) with the fixed effects of cow type (beef versus DBX), parity (1, 2 or 3), sire breed (AA or LM) and days in milk included in all models.

**Results** Live weight was 707 kg and 624 kg ($P<0.001$), and BCS was 3.23 and 2.61 ($P<0.001$) for beef and DBX, respectively. The DBX were found to be more docile ($P<0.05$) than beef cows (7.79 and 7.23, respectively). Teat placement and udder suspension were greater ($P<0.05$) for DBX (3.24 and 3.15, respectively) compared to beef cows (2.73 and 2.67, respectively). The DBX scored greater ($P<0.001$) for milkability than beef cows at 3.84 and 2.82, respectively. This may be attributed to the greater milk yield associated with DBX (Murphy *et al.*, 2008). Pelvic width score was greater ($P<0.001$) for beef cows (6.68) than DBX (5.70) similar to Laster (1974) who found differences in pelvic size between beef and DBX cows. Beef cows also had a more ideal hind leg rear view (3.71) than DBX (3.28; $P<0.05$). For all other variables investigated no difference was found between beef and DBX cows.

**Table 1** Linear traits recorded plus scale used

| Linear trait | Scale scored | |
|---|---|---|
| Locomotion | 1 (low) | 10 (high) |
| Fore Legs | 1 (toes pointed out) | 10 (toes pointed in) |
| Hind Legs - Side View | 1 (straight) | 10 (sickled) |
| Hind Legs - Rear View | 1 (straight) | 10 (sickled) |
| Docility | 1 (aggressive) | 10 (docile) |
| Width of Pelvis | 1 (narrow) | 10 (wide) |
| Length of Pelvis | 1 (short) | 10 (long) |
| Rump angle | 1 (shallow) | 10 (deep) |
| Width at pins | 1 (narrow) | 10 (wide) |
| Condition score | 0 (emaciated) | 5 (fat) |
| Teat placement | 1 (Close teat) | 5 (very wide) |
| Teat size | 1 (very small) | 5 (balloon shaped) |
| Udder suspension | 1 (very tight) | 5 (very pendulous) |
| Milkability | 1 (difficult) | 5 (easy) |

**Conclusion** Results from the current study suggest that some differences exist in linear type traits between beef and DBX cows. While DBX were more docile and had better milk attributes, beef cows had better hind legs and greater pelvic width indicating potentially less lameness and calving difficulties; all traits which can effect longevity. However, as a subjective visual assessment, linear scoring should be combined with physical performance recording to provide a more accurate and detailed comparison of the two cow types.

**References**

Laster, D.B. 1974. Journal of Animal Science 38, 496-503.
McGee, M. 2012. Review of Irish suckler cow types: Research perspective. In Suckler Cow Breeding Conference, Tullamore.
Murphy, B., Drennan, M.J., O'Mara, F.P. and McGee, M. 2008. Irish Journal of Agricultural and Food Research 47, 13-25.
Veerkamp, R.F., Gerritsen, C.L.M., Koenen, E.P.C., Hamoen, A. and De Jong, G. 2002. Journal of Dairy Science 85, 976-983.

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# Effect of sheep breed on intramuscular fat percentage and fatty acid composition of *M. longissimus thoracis et lumborum*

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**Application** The sheep breeds examined differ in intramuscular fat (IMF) and fatty acid (FA) profiles. This suggests that different marketing strategies could be developed to meet consumer demand for both taste and leaner cuts of meat.

**Introduction** Consumers increasingly seek flavoursome cuts of meat with a drive towards lower fat/healthier food options. IMF concentration influences both the health profile of meat and important characteristics of sensory quality such as juiciness, flavour and tenderness perception (Pannier *et al*., 2014). Studies in pigs have identified differences between breeds in relation to IMF concentration and variation in IMF is also associated with changes in the fatty acid profile (Vehovsky *et al*., 2015). High levels of saturated fatty acid (SFA) consumption are associated with increased risk of cardiovascular disease, while certain MUFA and polyunsaturated fatty acids (PUFAs) are associated with human health benefits. The objective of this study is to identify differences between breeds for IMF and FA profile to understand how the meat of different breeds can meet consumer needs.

**Material and methods** Fatty acid composition and IMF were determined for *Longissmus thoracsis et lumborum* (LTL) from 64 twin lambs from mixed breed ewes, sired by Charollais (n = 40), Suffolk (n = 42) or Texel (n = 46) rams. Fatty acid composition was determined from 1 g of LTL muscle by gas chromatography (Clarus 580, PerkinElmer, MA, USA) using a capillary column (Zebron ZB-5MS). The statistical model included the effects of sire breed, slaughter date and IMF. Breed differences were assessed using Tukey-Kramer post-hoc tests. Proportions of FAs are expressed relative to IMF %.

**Results** Lambs sired by the Suffolk breed had higher IMF% than those sired by Charollais ($P < 0.05$) and tended to have higher IMF% than the Texel sired lambs ($P < 0.10$). Lambs sired by Suffolk had higher MUFA than both the progeny of Texel and Charollais ($P < 0.05$), and a higher proportion of the most abundant MUFA, oleic acid (C18:1ω9), than the Charollais. The Suffolk-sired lambs tended to have higher total FA and had higher SFAs (palmitic (C16:0) and stearic acid (C18:0)) than the lambs sired by the Charollais breed ($P < 0.10$). Texel-sired lambs tended to have higher IMF% and MUFA ($P < 0.10$) than the Charollais sired lambs. PUFA concentration did not differ significantly between the sire-breed groups.

**Table 1** Intramuscular fat & fatty acid composition (average ±SEM) of M. *longissmus thoracis et lumborum* (mg/g).

| | Breed of sire | | |
|---|---|---|---|
| | Charollais | Texel | Suffolk |
| Intramuscular fat % | $2.2^{axy} \pm 0.20$ | $2.3^{ay} \pm 0.19$ | $2.9^{bx} \pm 0.18$ |
| **Fatty acid** | | | |
| Total FA (mg/g) | $21.3^{x} \pm 1.82$ | $25.0^{xy} \pm 1.68$ | $26.9^{y} \pm 1.69$ |
| Total SFA | $10.7 \pm 0.97$ | $12.1 \pm 0.89$ | $13.2 \pm 0.89$ |
| Total MUFA | $9.2^{ax} \pm 0.87$ | $11.5^{aby} \pm 0.79$ | $12.3^{by} \pm 0.78$ |
| Total PUFA | $1.5 \pm 0.09$ | $1.4 \pm 0.08$ | $1.4 \pm 0.08$ |
| Long-chain UFA[†] | $0.7 \pm 0.04$ | $0.7 \pm 0.04$ | $0.7 \pm 0.04$ |
| C16:0 | $4.9^{x} \pm 0.48$ | $6.1^{xy} \pm 0.44$ | $6.4^{y} \pm 0.44$ |
| C18:0 | $4.6^{y} \pm 0.41$ | $4.8^{xy} \pm 0.37$ | $5.6^{x} \pm 0.37$ |
| C18:1ω9[‡] | $8.4^{a} \pm 0.78$ | $10.6^{ab} \pm 0.72$ | $11.4^{b} \pm 0.72$ |
| C18:2ω6 | $0.8 \pm 0.06$ | $0.7 \pm 0.05$ | $0.8 \pm 0.05$ |

[†]C20:1ω9 + C20:2ω6 + C20:3ω3 + C20:3ω6 + C20:5ω3 + C22:6ω3; [‡]Includes C18:2ω6 and 18:3ω3 (co-eluted)
[ab] Means: $P < 0.05$, [xy] Means: ($P < 0.10$)

**Conclusion** Lambs sired by Suffolk rams had the highest IMF% and highest FA concentration in IMF, suggesting improved eating quality. While total SFA was unaffected by sire breed, meat from lambs sired by Charollais was the leanest and had the lowest FA suggesting meat from this breed may be more suitable for consumers seeking healthier cuts of meat. Future work will assess the sensory quality of meat from these breeds.

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**References**

Pannier, L., Pethick, D.W., Geesink, G.H., Ball, A.J., Jacob, R.H. 2014. Meat Science 96, 1068-1075.
Vehovsky, K., Stupka, M., Okrouhlá, M., Brzobohatý, L., 2015. Journal of Central European Agriculture 16, 122-129.

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# Comparison between PLS and Bayesian models for the prediction of beef water holding traits using VisNIR spectra

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**Application** Drip-loss and cook-loss are economically important traits in beef production and the ability to predict such losses from carcasses using rapid methods would offer opportunities to improve this aspect of quality in the breeding population.

**Introduction** Some of the most critical quality and technological traits of meat are its moisture retention abilities, known as water holding capacity (WHC). Measurement of WHC is important to the beef industry because a loss in moisture leads to a loss in weight, and consequently a loss in yield (ElMasry *et al*., 2011). Reduced WHC also negatively impacts sensory quality traits such as juiciness and tenderness (Warriss, 2010). Visible-Near-infrared spectra (VisNIRS) has been proposed as a rapid, non-destructive technique with the potential to predict WHC values of beef. The aim of this study was to assess the relevance of VisNIR spectra for the prediction of beef WHC using and comparing different chemometric modelling approaches.

**Material and methods** Crossbred beef bulls and steers (18±4 month old, n= 485) finished under controlled feeding and environmental conditions were slaughtered in 7 batches in a commercial plant. ASD Labspec 5000 (ASD Inc., Boulder Colorado, USA) VisNIR spectrometer was used to collect spectra between 350-2500 nm with 1 nm intervals. Spectra were collected in triplicate and saved in absorbance mode [log (1/*R*)]. VisNIR spectra were recorded on the cut surface of the *LTL* muscle at 24 h *post mortem* (PM), immediately after cutting and after 1h blooming (25 h PM). This was repeated at 48 h and 49 h PM, respectively. Steaks with a thickness of 2.54 cm were removed from the *LTL* at 48 h PM. For the determination of cook-loss %, 14 day aged steaks were trimmed of external fat and weighed. Steaks were immersed in a water bath for cooking at 72°C until an internal core temperature of 70°C was reached. Once samples cooled to room temperature their weight was recorded and cook-loss was expressed as percentage of original weight. Drip-loss % was analysed according to the procedure of Honikel and Hamm (1994) and expressed as percentage of original weight. All of the computations were carried out in the R environment. The packages used were "pls" for the Partial Least Square (**PLS**) regression, "BGLR" for the Bayesian and "prospectr" for the spectral treatments. We have used 3 spectral treatments (raw, 1 derivative, standard normal variate and detrend), 2 approaches for selection of the optimal number of PLS components (PLS_a and PLS_b), and two Bayesian models, Bayes B and Bayes Lasso. Ten validation replicas were performed by slaughter day for each trait, dividing the data set in a validation and a calibration set in each replica. The coefficient of determination of calibration ($R^2_{CAL}$) and validation ($R^2_{VAL}$), and the root mean squared error of validation ($RMSE_{VAL}$) were calculated for each replica as model fitting statistics.

**Results** Low to moderate $R^2$ values were found for all of the studied traits (Table 1). Drip-loss was better predicted using the spectra recorded at 2 days post-mortem after 1 hour interval, while cook-loss was modelled best at the quartering stage, 1 day post-mortem. No significant difference was found among models, however, Bayesian models were better than PLS for prediction of drip-loss. For cook-loss, PLS was better using the snvd, in respect to Bayesian with raw spectra.

**Table 1** Fitting statistics of prediction models. Best results according to $RMSE_{VAL}$ are shown.

| Model/trait | Drip Loss | | | | Cook Loss | | | |
|---|---|---|---|---|---|---|---|---|
| | $R^2_{CAL}$ | $R^2_{VAL}$ | $RMSE_{VAL}$ | Treatment/d | $R^2_{CAL}$ | $R^2_{VAL}$ | $RMSE_{VAL}$ | Treatment/d |
| PLS_a | 0.11 | 0.09 | 1.20 | snvd, d2h1 | 0.11 | 0.10 | 2.79 | snvd, d1h0 |
| PLS_b | 0.10 | 0.09 | 1.20 | snvd, d2h1 | 0.13 | 0.09 | 2.81 | snvd, d1h0 |
| BayesB | 0.28 | 0.10 | 1.16 | snvd, d2h1 | 0.18 | 0.09 | 2.95 | raw, d1h0 |
| BayesL | 0.27 | 0.09 | 1.18 | snvd, d2h1 | 0.17 | 0.09 | 2.84 | raw, d1h0 |

$R^2_{CAL}$, $R^2_{VAL}$, $RMSE_{VAL}$: average of the 10 validation replicas. Treatment/d: standard normal variate and detrend (snvd); raw spectra (raw); day 1 and 2 post-mortem (d1, d2) respectively; time 0 and after one hour (h0, h1) respectively.

**Conclusion** Cook-loss and drip-loss are difficult traits to predict but Bayesian approaches seem to be more promising compared with PLS regression to develop more accurate prediction models in this analysis.

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**References**

ElMasry G, Sun D.W, Allen P 2011. Food Research International 44, 2624–2633.
Honikel K, Hamm R 1994. Advances in meat research. 9th edition 125–161.
Wariss PD 2000. Meat science: An introductory text. 2nd edition 229-251.

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# Epidemiology of morbidity and mortality in Irish suckler beef and dairy calves from birth to 6 months of age

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**Application** Morbidity and mortality estimates were determined for suckler beef and dairy calves under Irish field conditions; these baseline data can be used to benchmark and monitor changes in calf health over time.

**Introduction** To date, no observational studies have been conducted to characterise the incidence of calfhood morbidity and mortality in Ireland. Disease surveillance data are available through the All-Island Animal Disease Surveillance Programme, and are compiled from voluntary submissions, often from clinically ill calves or herds with recurring health problems. The study objective was to describe the epidemiology of morbidity and mortality in Irish calves from birth to 6 months of age.

**Material and methods** In total, 1,392 suckler (n = 111 farms) and 2,090 dairy calves (n = 84 farms) were born between July 2014 – June 2016 and followed until 6 months of age. Farmers recorded information on disease events, treatments, and calf death. Case definitions were provided to assist with classification of disease. Cumulative incidence and incidence rate of crude and cause-specific morbidity were determined using health data from 1,192 suckler (n = 84 farms) and 1,733 dairy calves (n = 55 farms). Cumulative incidence of mortality was determined using death data from all calves. Generalised linear mixed models were used to evaluate associations between calf type (suckler vs. dairy) and morbidity and mortality.

**Results** Crude and cause-specific morbidity for suckler and dairy calves is described in Table 1. Median age at first treatment in suckler calves for crude morbidity, diarrhoea, bovine respiratory disease (BRD), navel infection, and joint infection/lameness was 14 (Q1 = 8, Q3 = 43), 13 (Q1 = 8, Q3 = 23), 48 (Q1 = 31, Q3 = 96), 7 (Q1 = 5, Q3 = 12), and 24 (Q1 = 11, Q3 = 52) days, respectively. Median age at first treatment in dairy calves for crude morbidity, diarrhoea, BRD, navel infection, and joint infection/lameness was 13 (Q1 = 7, Q3 = 20), 12 (Q1 = 7, Q3 = 19), 20 (Q1 = 11, Q3 = 30), 18 (Q1 = 6, Q3 = 27), and 37 (Q1 = 20, Q3 = 91) days, respectively. Suckler calves had greater odds of BRD (OR, 95% confidence interval (CI): 2.8, 1.2 – 6.5, $P = 0.01$), navel infection (5.1, 1.9 – 13.2, $P < 0.001$), and joint infection/lameness (3.2, 1.3 – 7.8, $P = 0.01$) in the first 6 months of life compared to dairy calves. Suckler calves also had increased rates of navel infection by 6 months of age compared to dairy calves (incidence rate ratio (IRR), 95% CI: 3.3, 1.3 – 8.4, $P = 0.01$). Incidence rate of diarrhoea from birth to 6 months of age was greater in dairy vs. suckler calves (IRR, 95% CI: 1.1, 1.1 – 5.0, $P = 0.03$). Median age at death for suckler and dairy calves was 51 (Q1 = 30, Q3 = 74) and 27 (Q1 = 18, Q3 = 74) days, respectively. Suckler and dairy calves did not differ for the odds of mortality in the first 6 months of life ($P = 0.83$; Table 1).

**Table 1** Morbidity and mortality for suckler beef and dairy calves from birth to 6 mo. of age.

| | Cumulative incidence[1] | | | | Incidence rate[2] | | | |
|---|---|---|---|---|---|---|---|---|
| | 0 to 1 mo. | 1 to 3 mo. | 3 to 6 mo. | 0 to 6 mo. | 0 to 1 mo. | 1 to 3 mo. | 3 to 6 mo. | 0 to 6 mo. |
| Suckler calves | | | | | | | | |
| Crude morbidity[3] | 14.3 | 6.0 | 2.1 | 20.3 | 15.0 | 3.1 | 0.7 | 4.1 |
| Diarrhoea | 8.2 | 1.8 | 0 | 9.4 | 8.7 | 0.9 | 0 | 1.9 |
| BRD[4] | 1.3 | 3.0 | 1.5 | 5.6 | 1.3 | 1.6 | 0.5 | 1.0 |
| Navel infection | 3.4 | 0 | 0 | 3.4 | 3.4 | 0 | 0 | 0.6 |
| Joint infection | 1.2 | 0.6 | 0.3 | 2.0 | 1.3 | 0.3 | 0.1 | 0.4 |
| Mortality[5] | 0.7 | 1.6 | 0.4 | 2.7 | - | - | - | - |
| Dairy calves | | | | | | | | |
| Crude morbidity[3] | 27.6 | 3.5 | 1.2 | 30.1 | 30.6 | 2.3 | 0.4 | 8.7 |
| Diarrhoea | 23.3 | 1.4 | 0.2 | 24.3 | 25.5 | 0.9 | 0.1 | 6.7 |
| BRD | 2.7 | 1.1 | 0.1 | 3.5 | 3.1 | 0.6 | 0.04 | 1.0 |
| Navel infection | 1.2 | 0.3 | 0 | 1.4 | 1.2 | 0.2 | 0 | 0.4 |
| Joint infection | 0.2 | 0.2 | 0.3 | 0.5 | 0.2 | 0.2 | 0.1 | 0.2 |
| Mortality[5] | 1.7 | 0.9 | 0.8 | 3.3 | - | - | - | - |

[1]Calves treated for disease / population at risk, no. per 100 calves; [2]Disease events (all-occurrences) treated / animal-time at risk, no. per 100 calf-mo. at risk; [3]Treated for at least 1 disease event; [4] Bovine respiratory disease; [5]All-cause mortality

**Conclusion** Approximately 20% of suckler calves and 30% of dairy calves were treated for disease between birth and 6 months of age. Suckler calves were more frequently treated for BRD, navel and joint infections in the first 6 months of life, relative to dairy calves. The incidence rate of diarrhoea from birth to 6 months was greater in dairy than suckler calves.

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