

Performance of dairy cows offered isonitrogenous diets containing urea or fishmeal in early and in mid-lactation

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1. Sixteen Friesian cows were used in Expt 1 to measure the effect of substituting urea-N with fishmeal-N either in early lactation (Part 1) or in mid-lactation (Part 2).
2. In Part 1 (days 15–84 of lactation) the major N constituent of the concentrate was urea (U), urea-N:fishmeal-N in the ratio 2:1 (UF) or 1:2 (FU), or fishmeal (F). In Part 2 (days 84–175 of lactation) only urea (UM) and fishmeal (FM) were used.
3. Replacement of urea-N with fishmeal-N significantly ($P < 0.05$) increased yield of milk protein both in early and in mid-lactation. At both stages of lactation the cows were, by calculation, in positive energy balance. In mid-lactation replacement of urea-N by fishmeal-N significantly depressed ($P < 0.001$) the concentration of fat in milk.
4. Blood urea concentration decreased with increasing fishmeal inclusion ($P < 0.05$) from U to FU.
5. In Expt 2 the diets used in Expt 1, Part 1, were offered at a maintenance level of feeding to non-pregnant, non-lactating heifers in a 4×4 Latin square design experiment. Digestibility of dry matter, organic matter and cell-wall constituents increased progressively ($P < 0.05$) with the first two increments of fishmeal inclusion.
6. A major effect of replacing urea-N with fishmeal-N was to increase digestible organic matter intake (DOMI) and differences in DOMI between treatments in Expt 1, Part 1, accounted for observed differences in performance.

It has been suggested that rumen microbial protein production alone is insufficient to meet the needs of the high-yielding dairy cow (Agricultural Research Council, 1980). It follows that, for the high-yielding cow, the source of dietary N is likely to be an important consideration; only those food proteins which escape rumen degradation can contribute directly to the supply of amino acids to the tissues and it can be postulated that dietary protein sources may be selected to supplement microbial amino acid supply so as to meet precisely the needs of the cow for total amino acids (Kaufmann & Hagemester, 1975; Satter & Roffler, 1975; Verite *et al.* 1979; Agricultural Research Council, 1980).

The experimental background against which to test these proposals is still surprisingly weak. The majority of relevant reports come from North America where the comparison has usually been between soya-bean meal and urea (Huber, 1975; Polan *et al.* 1976; Wohlt & Clark, 1978). For rations containing more than 120 g crude protein (CP; $N \times 6.25$)/kg dry matter (DM), soya-bean meal generally promoted higher rates of milk production than did urea. Van Horn *et al.* (1979) reported that soya-bean meal also promoted higher rates of milk production than did cottonseed meal in isonitrogenous diets.

As fishmeal has been reported to be a protein source which escapes rumen degradation to a substantial extent (Agricultural Research Council, 1980), it might be expected that this could be a particularly effective source of dietary protein with which to manipulate amino acid supply to the intestines. Supplementation of maize-silage diets with fishmeal has been found to increase amino acid supply to the duodenum in growing cattle (Cottrill *et al.* 1982) and replacement of urea-N with fishmeal-N improved growth rate of young lambs (Ørskov *et al.* 1974) and young cattle (Oldham & Smith, 1982).

The aim of the experiments reported here was to investigate the effects of replacing urea-N

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Table 1. *The composition (g/kg) and mean concentration of dry matter (DM), organic matter (OM; g/kg DM), cell-wall constituents (CWC; g/kg DM), crude protein (CP; g/kg DM) and crude fibre (CF; g/kg DM) of the experimental concentrates U, UF, FU and F, Viton cubes and maize silage used in Expts 1 and 2*

Ration constituent...	U	UF	FU	F	Viton cubes*	Maize silage
Ingredient (g/kg concentrate):						
Barley	849	817	786	752	—	—
Ground maize	116	116	116	116	—	—
Urea	24	16	8	—	—	—
Fishmeal	—	40	79	121	—	—
Mineral/vitamin supplement	11	11	11	11	—	—
DM (g/kg)	854	856	858	863	874	214
OM (g/kg DM)	969	964	962	960	893	949
CWC (g/kg DM)	151	180	235	262	695	608
CP (g/kg DM)	181	186	177	192	38	70
CF (g/kg DM)	46	44	42	43	393	268

* Viton-NIS alkali-treated straw cubes (Unitriton Ltd).

with fishmeal-N in the diet of cows early in lactation and in mid-lactation, to assess the response of the cows to an increase in amino acid supplied to the intestines at these times.

A preliminary report of this work has been published (Oldham *et al.* 1979).

METHODS

Expt 1

Animals and management. Sixteen mature Friesian cows were used in the feeding trial. They were individually housed on rubber mats in concrete standings. The rations consisted of maize silage, alkali-treated straw cubes and concentrates (Table 1). Half of the daily ration of each constituent was offered at 06.00, the other half at 14.00 hours. These constituents were added separately to the same feed trough. Refusals of food were collected and weighed before the feed at 14.00 hours. Immediately after each feed the cows were milked in the standings. The cows were weighed on Monday and Friday each week.

Part 1

Experimental design and rations. This part of the experiment was designed to measure, in early lactation, the production response of cows offered isonitrogenous rations at a fixed level, designed to be below *ad lib.* intake, containing urea (U), urea + fishmeal (UF; ratio 2:1, urea-N:fishmeal-N), urea + fishmeal (FU; ratio 1:2, urea-N:fishmeal-N) or fishmeal (F) as the major N supplement. The proportions of all ration ingredients are given in Table 1 together with chemical analysis of the diets.

Before parturition all cows were offered, per day, 5 kg UF cubes, 2 kg Viton alkali-treated straw cubes and 10 kg maize silage (fresh weight basis).

For 14 d after parturition all cows received the same ration. This period was used for adjustment of data for differences between cows by covariance analysis. The level of feeding in this period rose by increments so that on day 14 post partum each cow received, on a fresh weight basis, 9 kg UF cubes, 2 kg Viton alkali-treated straw cubes and 15 kg maize silage/d. On day 15 post partum the cows were grouped in blocks of four according to live weight and milk yield, as measured in the period 8–14 d, and assigned to one of the experimental treatments: U, UF, FU or F. From days 15 to 84 post partum the daily feed

allocation was held constant at 11.5 kg experimental concentrate plus 2 kg Viton alkali-treated straw cubes plus 20 kg maize silage (equivalent to 15.9 kg DM/d). One cow (treatment U) refused to eat maize silage and was withdrawn from the experiment.

In the statistical analysis of the data, multiple comparisons between treatment means using analysis of variance were preferred to regression analysis, even though the treatments were clearly structured with uniform increases in fishmeal content of the concentrate. The reason for this choice was that the production responses measured should properly have been related to supply of nutrients absorbed from the gastrointestinal tract and not to dietary intake. Thus use of a dietary factor to define the response variable could be misleading and it was not possible to measure nutrient uptake from the gut so as to define the correct response variable.

Measurements and analysis. Milk yield was recorded daily. Samples of milk were taken for four consecutive milkings from each cow on days 5 and 6, 8 and 9, and 13 and 14 post partum, and bulked, for each 2-d period, in proportion to yield. Analysis of these samples was used for covariance adjustment of milk composition data.

Thereafter a sample, bulked in proportion to milk yield, was taken for four consecutive milkings every week. Milk samples were analysed for fat, protein and lactose concentration using IRMA (Mark II; Grubb Parsons Ltd, Newcastle upon Tyne). Live weights recorded on Monday and Friday each week were averaged for weekly means. Samples of jugular venous blood were taken from each cow by venepuncture (Vacutainer system; Becton-Dickinson, Cowley, Oxford) on days 5, 9 and 14 and on Monday mornings in weeks 3, 4, 5, 6, 8, 10 and 12 after calving. All blood samples were collected between 09.30 and 10.30 hours. These samples were analysed for urea, glucose, haemoglobin, albumin, total protein content and packed-cell volume using methods described by Rowlands *et al.* (1974).

Samples of all foods were taken each week, bulked on a calendar monthly basis and analysed for cell-wall constituents (CWC; Van Soest & Wine, 1967) and DM, N, organic matter (OM) and crude fibre (CF) by standard methods.

Part 2

This part of the experiment was designed to compare the performance of cows offered isonitrogenous rations containing either urea (UM) or fishmeal (FM) as the major N supplement in mid-lactation.

The same cows were used as in Part 1. On day 85 post partum they were re-randomized, balancing within treatments U, UF, FU and F to treatments UM or FM. This part of the experiment lasted from day 85 to day 175 post partum. Throughout this period the diet (/d) was held constant at 7 kg of either concentrate U (UM) or F (FM) plus 2 kg Viton alkali-treated straw cubes and 28 kg maize silage (equivalent to 13.8 kg DM).

The level of feeding and proportion of concentrates in the ration were changed from Expt 1 so as to reduce the overall plane of nutrition to be appropriate for cows yielding 20 kg milk/d and to lower the concentration of CP in the whole ration while still using the same concentrate formulations as in Expt 1.

Management of the cows was as in Part 1 except that no blood samples were taken.

All analyses of milk and food were as described for Part 1.

Because of the removal of one cow from the experiment in Part 1, only fifteen cows started this part of the trial (seven on treatment FM). A second cow had to be withdrawn with mastitis from the FM group during the trial.

Expt 2

The aim of this experiment was to measure the digestibility of rations used in Expt 1, Part 1.

Table 2. Intakes of dry matter (DM) and crude protein (CP) of cows offered treatments U, UF, FU and F in Expt 1, Part 1, the digestibility of DM, organic matter (OM), cell-wall constituents (CWC) and crude fibre (CF) of these treatments by heifers in Expt 2, and calculated intakes of digestible DM (DDM) and digestible OM (DOM) of cows in Expt 1, Part 1

Treatment	Intake (kg/d)		Digestibility				Calculated intake (kg/d)	
	DM	CP	DM	OM	CWC	CF	DDM	DOM
U	15.75 ^a	2.131 ^{a, b}	0.71 ^c	0.73 ^c	0.42 ^c	0.45 ^c	11.18	10.95
UF	15.71 ^a	2.175 ^a	0.76 ^b	0.78 ^b	0.56 ^b	0.56 ^b	11.94	11.67
FU	15.63 ^a	2.077 ^b	0.81 ^a	0.83 ^a	0.71 ^a	0.67 ^a	12.66	12.34
F	15.03 ^b	2.140 ^a	0.78 ^b	0.80 ^{a, b}	0.67 ^a	0.62 ^{a, b}	11.72	11.42
SED*	0.165	0.0232	0.012	0.011	0.027	0.029	—	—

^{a, b, c} Means in the same column which do not share a common superscript differ significantly ($P < 0.05$).

* SE of difference between two treatment means each of four cows with 8 df for DM and CP intakes (there was one cow missing for treatment U) and between two means of four heifers with 6 df for digestibility coefficients.

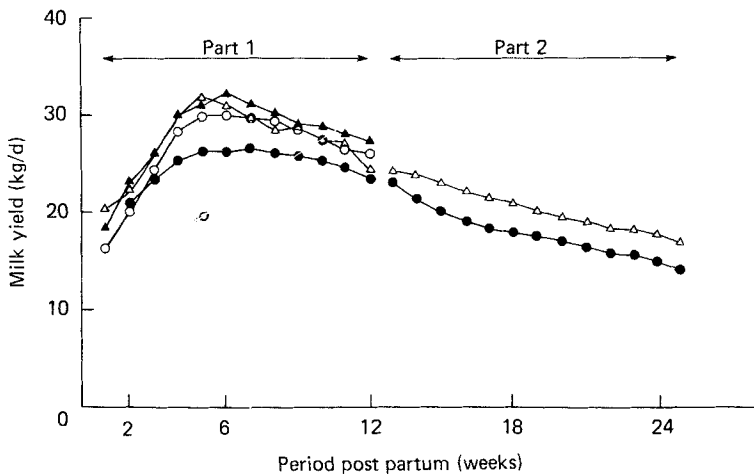


Fig. 1. Milk production (kg/d) of cows offered treatments (●) U, (○) UF, (▲) FU and (△) F in Expt 1, Part 1, and (●) UM and (△) FM in Expt 1, Part 2. For details, see pp. 338–339.

Four non-pregnant, non-lactating heifers were offered rations of the same composition as those used in Part 1 but offered at maintenance level of intake (equivalent to 5.9 kg DM/d). A 4 × 4 Latin square design was used in the trial with 4-week periods. A total collection of faeces was made for the last 8 d of each period as described by Smith (1979). Samples of food and faeces were analysed for DM, OM, CF and CWC as described above.

RESULTS

Expt 1, Part 1

Food intake. Cows offered treatment F consistently refused small amounts of food, with the result that DM intake for this treatment was lower ($P < 0.05$) than that for the other

Table 3. Milk production, milk composition and live-weight change of cows offered treatments U, UF, FU and F in Expt 1, Part 1

Treatment	Yield (kg/d)				Milk composition (g/kg)			Live-weight change (kg/d)
	Milk	Milk fat	Milk protein	Milk lactose	Fat	Protein	Lactose	
U	26.1	0.640 ^b	0.786 ^b	1.249	24.7 ^b	30.2	48.1	+0.39 ^{a, b}
UF	28.2	0.641 ^b	0.893 ^a	1.386	22.1 ^c	31.2	49.0	+0.53 ^a
FU	29.4	0.718 ^{a, b}	0.894 ^a	1.438	24.5 ^{b, c}	31.0	48.6	+0.18 ^b
F	28.7	0.785 ^a	0.851 ^{a, b}	1.391	27.4 ^a	29.8	48.4	+0.13 ^b
SED*	1.76	0.0372	0.0290	0.0382	1.01	0.80	0.91	0.134

^{a, b, c} Means in the same column which do not share a common superscript differ significantly ($P < 0.05$).

* SE of difference between two treatment means each of four cows with 7 df for milk-protein yield, adjusted by covariance on the values for days 7–14 of lactation, and with 8 df for the other variates. There was one cow missing for treatment U.

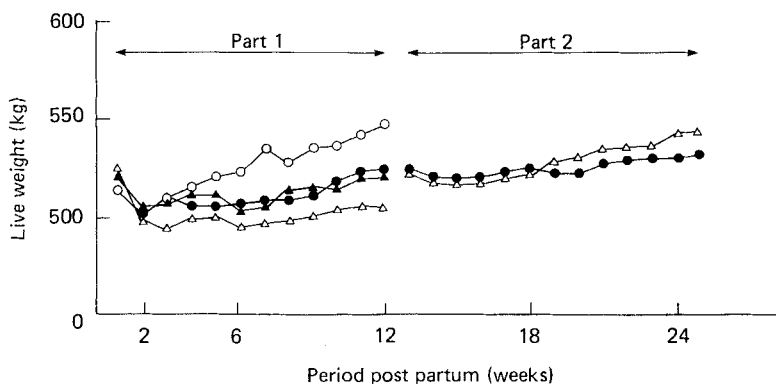


Fig. 2. Pattern of live-weight change of cows offered treatments (●) U, (○) UF, (▲) FU and (△) F in Expt 1, Part 1, and (●) UM and (△) FM in Expt 1, Part 2. For details, see pp. 338–339.

treatments (Table 2). Associated differences in CP intake, although statistically significant, were very small.

Intakes of digestible dry matter (DDM) and digestible organic matter (DOM) were calculated by multiplying mean measured DM and OM intakes in this experiment by digestibility coefficients for DM and OM measured in Expt 2 (see below and Table 2). Intake of DOM was greatest for treatment FU and least for treatment U, but the difference between these values could not be tested statistically.

Milk yield, milk composition and live-weight change. Lactation curves for the treatment groups are shown in Fig. 1. Mean milk yield was lowest with treatment U (Table 3).

The first two levels of fishmeal inclusion (UF and FU) significantly ($P < 0.05$) increased milk protein yield in comparison with that for treatment U. There was no effect on protein concentration in the milk. The highest level of fishmeal inclusion (F) had no further effect on protein yield but increased milk-fat yield and the concentration of fat in the milk. These effects on milk-fat yield and milk-fat concentration should, however, be viewed with caution as milk-fat concentrations were uniformly low for the four treatments (Table 3).

Table 4. Concentrations of urea, albumin, total protein, glucose, packed-cell volume (PCV) and haemoglobin in jugular blood of cows offered treatments U, UF, FU and F in Expt 1, Part 1

Treatment	Urea (mmol/l)	Albumin (g/l)	Total protein (g/l)	Glucose (mmol/l)	PCV (%)	Haemoglobin (g/l)
U	4.45 ^a	36	77 ^b	2.09 ^{a, b}	29	116
UF	3.06 ^b	35	83 ^{a, b}	2.35 ^a	32	120
FU	2.53 ^c	36	84 ^a	2.03 ^b	30	117
F	2.61 ^{b, c}	36	77 ^b	2.10 ^{a, b}	32	121
SED*	0.223	0.8	2.5	0.124	1.6	2.9

^{a, b, c} Means in the same column which do not share a common superscript differ significantly ($P < 0.05$).

* SE of difference between two treatment means each of four cows with 8 df. There was one cow missing for treatment U.

Table 5. Milk production, milk composition and live-weight change of cows offered treatments UM and FM in Expt 1, Part 2

Treatment	No. of cows	Yield (kg/d)				Milk composition (g/kg)			Live-weight change (kg/d)
		Milk	Milk fat	Milk protein	Milk lactose	Fat	Protein	Lactose	
UM	8	17.0	0.649	0.515	0.787	37.8	30.6	47.5	+0.18
FM	6	19.6	0.564	0.619	0.937	28.7	31.1	47.9	+0.48
Difference		2.6	-0.085	0.104*	0.150	-9.1***	0.5	0.4	0.30*
SED†		1.92	0.0557	0.0339	0.1082	1.43	0.61	1.38	0.112

* $P < 0.05$, *** $P < 0.001$.

† SE of difference with 5 df for milk-protein yield, adjusted by covariance on the values for days 7-14 of lactation, and with 6 df for the other variates.

There were no significant effects on milk lactose concentration or on milk lactose yield.

All cows gained in weight from the start of the experimental period (Fig. 2). Mean weight gains were highest for cows offered UF and lowest for those offered F.

Blood composition. Replacement of urea-N with fishmeal-N significantly reduced blood urea concentration (Table 4). There were no other consistent effects on blood composition, although blood glucose concentration was significantly ($P < 0.05$) higher with UF than with FU.

Expt 1, Part 2

Food intake. Intakes of DM for cows offered the urea (UM) or fishmeal (FM) treatments in mid-lactation were very similar (13.53 and 13.58 kg/d respectively, SE of difference 0.05). Intakes of CP were slightly lower for UM than FM (1542 and 1622 g/d respectively, SE of difference 18.3).

Milk yield and composition. Cows offered UM yielded significantly ($P < 0.05$) less milk protein than those offered FM (Table 5). Milk-fat yields were not affected by diet because the fat concentration in milk produced by cows offered UM was significantly ($P < 0.001$) higher than that for cows offered FM.

Live-weight change. Cows offered FM gained more weight than those offered UM.

Expt 2

The digestibilities of OM, CWC and CF were all increased by replacing urea-N with fishmeal-N (Table 2). Only the highest level of fishmeal inclusion had no further influence on digestibility.

DISCUSSION

In these experiments inclusion of some fishmeal in place of urea in the ration had a beneficial effect on yield of milk protein, both in early and in mid-lactation.

Ekern (1982) has summarized the results of a number of experiments done in Norway in which herring meal was compared with oil-seed meals as a source of N for lactating ruminants. In cows yielding 17–19 kg milk/d he reported no advantage in milk yield from inclusion of herring meal rather than oil-seed meal, but an increase of 3–7% in milk-protein concentration. In goats both milk yield and milk-protein concentration were increased by replacing soya-bean meal with herring meal (Ekern, 1982). In all of these studies the major forage source was grass silage, rather than maize silage as used in the present experiments.

The results presented here show that the main effect of fishmeal inclusion was to increase the yield of milk protein with little change in the concentration of protein in milk. Similar results were found when fishmeal was given as a protein supplement to cows on commercial farms (Miller *et al.* 1982) where the predominant forage was grass silage. In these trials fishmeal inclusion increased the daily yield of protein in milk, a result to be expected if fishmeal increased the supply of total protein to the intestines and the performance of the cows was limited by total protein supply.

In general there was little effect of treatment on the composition of milk in Part 1 of the trial, although milk fat concentrations were uniformly low. It has previously been found (Phipps, 1978) that milk-fat content may be low with maize-silage rations if no long hay or straw is included. Clearly the small amount of ground and pelleted alkali-treated straw included here was insufficient to prevent the milk-fat depression. The proportion of concentrates used in the ration seemed also to play a part. In early lactation (Part 1) concentrate DM represented 0.62 of total DM intake. When this was reduced to 0.44 in mid-lactation (Part 2), milk-fat content in general was increased, but the effect was small with fishmeal and large with the urea treatment.

The difference in milk-fat content between urea and fishmeal in this part of the experiment was large and highly significant in comparison with the other observed treatment effects. It is possible that the presence of fish oils inhibited the restoration of more normal milk-fat levels (treatment UM) when the ratio forage:concentrate was adjusted for Part 2 of the experiment (Nicholson & Sutton, 1971). The generally low milk-fat levels in Part 1 may have overshadowed such an effect. Alternatively, the presence of fishmeal in the ration in Part 2 may have influenced the rate of digestion of carbohydrates or cell walls in the rumen (McAllan & Smith, 1983) for which the results from Expt 2 give some support. Such an effect could be expected to influence milk-fat content (Sutton, 1982) but no definitive explanation of the observed effects on milk fat can be given as the appropriate measurements were not made. The magnitude of the difference in milk-fat content between treatments UM and FM is, however, noteworthy and was large enough to have important practical consequences.

The scale of the response to fishmeal inclusion, in daily yield of milk protein (14% increase comparing U and UF in Expt 1, Part 1; 17% increase comparing UM and FM in Expt 1, Part 2), was similar to the scale of response achieved when cows have been supplemented with casein by infusion into the abomasum (Oldham, 1981). It appeared that the response

in milk-protein yield to replacement of urea-N by fishmeal-N was as great in mid-lactation, when milk yields were only 17–20 kg/d, as in early lactation, when milk yields were 26–29 kg/d.

The effect on digestibility of replacing urea-N with fishmeal-N suggests that not only dietary concentration of N (Oldham & Smith, 1982) but also the nature of dietary N can affect digestibility under some circumstances. Since the main effect was on digestion of fibre it seems likely that the main influence was on digestion in the rumen. It has been suggested (Hespell & Bryant, 1979) that low-degradability protein sources, like fishmeal, exert their influence on animal performance not just by supplying extra undegraded dietary protein to the intestines but possibly also by acting as slow-release N sources in the rumen. There are some results which show a specific stimulating effect on rumen microbial growth *in vitro* when pre-formed amino acids, as opposed to ammonia, are supplied as N substrates (Maeng *et al.* 1976; Cotta & Russell, 1982). Replacing urea-N with fishmeal-N has been found to increase rumen microbial growth *in vivo* (Cottrill *et al.* 1982) and to maintain higher rates of cellulose digestion in the rumen in comparison with isonitrogenous amounts of urea (McAllan & Smith, 1983).

Substitution of urea-N with protein-N does not always result in improved digestion of food (Poos *et al.* 1979; Redman *et al.* 1980; Oldham *et al.* 1981) so factors other than the nature of the major N source play a part. It is not possible to identify what these may be from the results of the present work. However, it is pertinent to note the importance of conducting a digestibility trial in experiments such as this so that some account can be taken of the influence of dietary treatment on the supply of energy-yielding nutrients. It would have been preferable to have made the measurements of digestibility in cows at the same level of feeding as those in Expt 1 because level of feeding is known to be an important factor influencing the effect of dietary protein on ration digestibility (Oldham, 1984); unfortunately, however, this was not feasible in this study. Nonetheless, the effects of treatment on estimated DOM intake played a large part in determining the observed responses. Productive protein output (milk-protein yield plus protein content of live-weight change plus estimated maintenance protein need, calculated according to the Agricultural Research Council (1980)) was 84 (SE 3.6) g/kg DOM intake for the four treatments in Part 1. This consistent relation between performance and DOM intake accounted for differences in performance equal to predictions of duodenal protein supply based on information in Agricultural Research Council (1980).

It has been suggested that protein sources with a low-degradable protein concentration, like fishmeal, will exert their greatest influence on production in cows which are in substantial negative energy balance (Ørskov *et al.* 1981). In the present study the calculated average energy balance [(metabolizable energy (ME) intake) minus (gross energy of milk/ k_l) minus (ME required for maintenance)] of cows in Expt 1, Part 1 was (MJ ME/d) U, +10; UF, +13; FU, +24; F, +5. For these calculations it was assumed that maintenance needs for ME were 0.51 MJ/kg body-weight^{0.75} per d, k_l (the partial efficiency with which ME is used for milk synthesis) = 0.63, and gross energy concentrations (MJ/kg) in milk fat, protein and lactose were 38.1, 24.5 and 16.5, respectively.

For the mid-lactation part of the experiment, ration digestibilities were not measured, so precise calculations of ME intake cannot be made. However, on the basis of estimated ME intake (Ministry of Agriculture, Fisheries and Food, 1975) average energy balance in mid-lactation was +14 and +10 MJ ME/d for UM and FM respectively.

These calculations show that replacing a high rumen-degradable protein ingredient with a high undegraded dietary protein feed such as fishmeal, can improve performance even though the cows are not initially in negative energy balance. However, the causal effects of this response remain to be elucidated.

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REFERENCES

- Agricultural Research Council (1980). *The Nutrient Requirements of Ruminant Livestock*. Commonwealth Agricultural Bureaux, Farnham Royal, England.
- Cotta, M. A. & Russell, J. B. (1982). *Journal of Dairy Science* **65**, 226–234.
- Cottrill, B. R., Beaver, D. E., Austin, A. R. & Osbourn, D. F. (1982). *British Journal of Nutrition* **48**, 527–541.
- Ekern, A. (1982). In *Protein Contribution of Feedstuffs for Ruminants*, pp. 86–102 [E. L. Miller, I. H. Pike and A. J. H. Van Es, editors]. London: Butterworths.
- Hespell, R. B. & Bryant, M. P. (1979). *Journal of Animal Science* **49**, 1640–1659.
- Huber, J. T. (1975). *Journal of Animal Science* **41**, 954–961.
- Kaufmann, W. & Hagemester, H. (1975). *Übersichten zur Tierernährung* **3**, 33–65.
- McAllan, A. B. & Smith, R. H. (1983). *British Journal of Nutrition* **50**, 445–454.
- Maeng, W. J., Van Nevel, C. J., Baldwin, R. L. & Morris, J. G. (1976). *Journal of Dairy Science* **59**, 68–79.
- Miller, E. L., Galway, N. W., Newman, G. & Pike, I. H. (1982). In *Protein Contribution of Feedstuffs for Ruminants*, pp. 131–141 [E. L. Miller, I. H. Pike and A. J. H. Van Es, editors]. London: Butterworths.
- Ministry of Agriculture, Fisheries and Food (1975). *Energy Allowances and Feeding Systems for Ruminants*. Technical Bulletin no. 33. London: H.M. Stationery Office.
- Nicholson, J. W. G. & Sutton, J. D. (1971). *Journal of Dairy Research* **38**, 363–372.
- Oldham, J. D. (1981). In *Recent Advances in Animal Nutrition—1980*, pp. 33–65 [W. Haresign, editor]. London: Butterworths.
- Oldham, J. D. (1984). *Journal of Dairy Science* **67**, 1090–1114.
- Oldham, J. D., Broster, W. H., Napper, D. J. & Smith, T. (1979). *Proceedings of the Nutrition Society* **38**, 128A.
- Oldham, J. D., Fulford, R. J. & Napper, D. J. (1981). *Proceedings of the Nutrition Society* **40**, 30A.
- Oldham, J. D. & Smith, T. (1982). In *Protein Contribution of Feedstuffs for Ruminants* pp. 103–130 [E. L. Miller, I. H. Pike and A. J. H. Van Es, editors]. London: Butterworths.
- Ørskov, E. R., Fraser, C., McDonald, I. & Smart, R. I. (1974). *British Journal of Nutrition* **31**, 89–98.
- Ørskov, E. R., Reid, G. W. & McDonald, I. (1981). *British Journal of Nutrition* **45**, 547–555.
- Phipps, R. H. (1978). In *Forage Maize. Production and Utilisation*, pp. 263–295 [E. S. Bunting, B. F. Pain, R. H. Phipps, J. M. Wilkinson, R. E. Gunn, editors]. London: Agricultural Research Council.
- Polan, C. W., Miller, C. N. & McGilliard, N. L. (1976). *Journal of Dairy Science* **59**, 1910–1914.
- Poos, M. I., Bull, L. S. & Hemken, R. W. (1979). *Journal of Animal Science* **49**, 1417–1426.
- Redman, R. G., Kellaway, R. C. & Leibholz, J. (1980). *British Journal of Nutrition* **44**, 343–354.
- Rowlands, G. J., Little, W., Manston, R. & Dew, S. M. (1974). *Journal of Agricultural Science, Cambridge* **83**, 27–35.
- Satter, L. D. & Roffler, R. E. (1975). *Journal of Dairy Science* **58**, 1219–1237.
- Smith, T. (1979). *Journal of the Science of Food and Agriculture* **30**, 215–217.
- Sutton, J. D. (1982). In *Recent Advances in Animal Nutrition—1981*, pp. 35–48 [W. Haresign, editor]. London: Butterworths.
- Van Horn, H. H., Zometa, C. A., Wilcox, C. J., Marshall, S. P. & Harris, B. (1979). *Journal of Dairy Science* **62**, 1086–1093.
- Van Soest, P. J. & Wine, R. H. (1967). *Journal of the Association of Official Analytical Chemists* **50**, 50–55.
- Verite, R., Journet, M. & Jarrige, R. (1979). *Livestock Production Science* **6**, 349–367.
- Wohlt, J. E. & Clark, J. H. (1978). *Journal of Dairy Science* **61**, 916–931.