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ABSTRACTS OF COMMUNICATIONS

The One Hundred and Forty-seventh Meeting of The Nutrition Society was held at the Hannah Dairy Research Institute, Kirkhill, Ayr, on Friday, 9 February 1962, at 1 p.m., when the following papers were read :

Metabolites of riboflavin. By E. C. OWEN, *Hannah Dairy Research Institute, Kirkhill, Ayr*

On certain diets (Crossland, Owen & Proudfoot, 1958*a*,*b*) British Saanen goats excrete only riboflavin but after an oral dose of riboflavin the urine acquires a greenish fluorescence even in daylight and in the lactating goat the milk changes from white to yellow. Studies in this laboratory have shown these colorations to be due to a series of metabolites (RM1, RM2 and RM3) of riboflavin. I have isolated the chief one of these, RM1, in very small amounts in pure crystalline form from concentrates in chloroform (Montgomery, Owen & Proudfoot, 1959). Crystalline concentrates of RM1 contain a colourless component, which has a strong absorption between 230 and 240 mμ, as their major constituent. RM1 in solution free from the colourless substance was separated by Montgomery by partition of the mixture between amyl alcohol and water and more recently I have made a similar separation by passing the chloroform concentrate through a column of alumina (Savory and Moore) which absorbed all the pigment in a narrow band at the top. Elution, with 2% acetic acid in chloroform, split the yellow-fluorescent band into a wide upper yellow-fluorescent band and a much smaller lower yellow-fluorescent band, between which, but distinct from each, was a very narrow blue-fluorescent band. During this development a blue-fluorescent material was eluted. The material forming the major yellow-fluorescent band was collected and had, as also had lumiflavin and the RM1 extracted by pentanol from water, the type of absorption spectrum shown by methylated derivatives of isoalloxazine (Kuhn & Weygand, 1934; Kuhn & Bär, 1934; Kuhn & Rudy, 1934*a*,*b*) and called 1c by Harbury, LaNoue, Loach & Amick (1959), with peaks at 270, 350 and 450 mμ. On evaporation the acid chloroform solution gave yellow crystals. In spite of many similarities lumiflavin and RM1 are different substances. Concentrates from urine, mixed with concentrates from three times as much milk, gave paper chromatograms which showed (1) that RM2 was not present in milk, (2) that free flavinadenine dinucleotide was not present in urine, (3) that RM1 and RM3 were matched by substances of the same R_F in milk, and (4) that a substance with the R_F of flavin-5′-phosphate was present in both. The appearances of chromatograms were unaffected by keeping the goats in complete darkness from dosing to collection of milk and urine.

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Riboflavin in human milk. By E. C. OWEN, *Hannah Dairy Research Institute, Kirkhill, Ayr*, and F. E. HYTTEN, *Obstetric Medicine Research Unit (Medical Research Council), University of Aberdeen*

Because paper chromatography reveals metabolites of riboflavin in the milk of the goat, after ingestion of the pure vitamin (Owen, 1962) it was used to study the effects of ingestion of riboflavin by lactating women. From two primiparous mothers one M.K. aged 21, the other V.R. aged 30, milk samples were taken on the 4th and 6th days of lactation respectively and phenol extracts from them (Crossland, Owen & Proudfoot, 1958*a*,*b*) were chromatographed on the one sheet of paper. The chromatogram from M.K. showed in u.v. light four yellow-fluorescent substances of which one was riboflavin. The ratios of the R_F of the other spots to the riboflavin were 0·10, 0·24 and 0·47. The chromatogram of V.R.'s milk however showed no riboflavin but did show the other three substances. Both the women then ate 6 mg pure riboflavin and 4 h later milk samples were again taken. There was an increase in the fluorescence of the riboflavin spot from M.K. and a riboflavin spot appeared on V.R.'s chromatogram but no new yellow-fluorescent substances appeared on either. Neither woman produced either RM1 or any other new substance after ingestion of the vitamin, but this could be attributed either to differences between human and ruminant metabolism or to the insufficient size of the dose (6 mg) compared with the 2 g given to the goats, for earlier experiments had shown that 100 mg but not 30 mg could cause RM1 to appear in the urine of the goat.

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The incidence of rickets and idiopathic hypercalcaemia in Dundee. By K. G. LOWE, R. G. MITCHELL, H. G. MORGAN, W. K. STEWART and J. THOMSON, *Queen's College, University of St. Andrews, Dundee*

The virtual elimination of nutritional rickets as a common disease in this country has had two unexpected consequences—the recognition of rickets due to genetically determined metabolic disease and the appearance or recognition of idiopathic hypercalcaemia in infants. The incidence of nutritional rickets in Dundee, as assessed by admissions to hospital, declined steeply from 1935 onwards. Between 1948 and 1958, at least eight cases of 'resistant' rickets were diagnosed in the city. Cases of idiopathic hypercalcaemia of infants were recognized from 1952 onwards,

the incidence reaching a peak in 1956. Since 1957, when the Joint Sub-Committee on Welfare Foods made its recommendations that the levels of vitamin D in national cod-liver oil, national dried milk and infant cereals be considerably reduced (Central and Scottish Health Services Councils: Standing Medical Advisory Committees, 1957), the incidence of idiopathic hypercalcaemia in Dundee has fallen sharply.

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The effect of selenium on lamb growth: co-operative experiments on Scottish farms. By K. L. BLAXTER, *Hannah Dairy Research Institute, Kirkhill, Ayr.* (ON BEHALF OF THE AGRICULTURAL RESEARCH COUNCIL COMMITTEE ON SELENIUM)

Over fifty members of the staffs of the three Agricultural Colleges in Scotland have collaborated with a small committee of the Agricultural Research Council in conducting experiments to determine whether the administration of selenium to growing lambs increases weight gain. A total of 4448 lambs on seventy-six farms were used as experimental animals. Half the lambs were given 3 mg Se as selenate by subcutaneous injection at intervals of about 4 weeks. Each lamb was identified by an ear tag and weighed at monthly intervals for 3 or 4 months.

Experimental treatment had no effect on mortality or morbidity of lambs. Those given Se gained an average of 0·81 lb/lamb more than the controls during the experiment. Each farm was classified either on the basis of soil surveys where these existed or of a knowledge of local geology into groups based on the presumptive Se content of the soil. The overall classification showed that the response to Se on thirty-one farms with a low presumptive Se content was 1·72 $\pm$ 0·32 lb/lamb, a figure which increased to 2·03 $\pm$ 0·44 when the sixteen farms in this group for which soil surveys existed were considered. On twenty-seven farms with a high presumptive Se content the response was $-0{\cdot}16 \pm 0{\cdot}34$ lb/lamb and for those thirteen farms in this group which had been surveyed $-0{\cdot}21 \pm 0{\cdot}49$ lb/lamb.

The experiments reveal the widespread presence of a deficiency of Se in Scottish soils. Judged from the relative magnitude of the growth responses of lambs to Se, however, the deficiency is mild compared with that noted in New Zealand (Hartley & Grant, 1961) or in Oregon, USA (Schubert, Muth, Oldfield & Remmert, 1961).

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The zinc contents of eggs of some inbred lines of Brown Leghorn fowls. By W. A. DEWAR, *Agricultural Research Council Poultry Research Centre, West Mains Road, Edinburgh, 9*

To determine which of the inbred lines of Brown Leghorn fowls maintained at this Centre was most suitable for studies in zinc metabolism, a survey of the eggs

laid by the lines was carried out. One egg from each of twelve hens in each line was taken, boiled hard and the shell and albumen discarded since their Zn contents are negligible. The Zn content of the defatted yolk was determined by a mixed colour dithizone method. The diet contained 27 mg Zn/kg food.

The mean Zn contents, with their standard errors, for eggs from five of the lines are set out in Table 1. It is apparent that the Zn contents do not differ between lines, therefore the abilities of the birds to transfer Zn from the food to the egg do not differ, although differences within the lines could be large, as shown by the standard errors for lines I and D.

Table 1. *Zinc contents of eggs from inbred lines of Brown Leghorn fowls*

Line	N	I	R	D	B
Egg weight (g)	45·6	51·2	38·3	44·4	55·7
Zn content (mg/egg)	0·50 ± 0·02	0·51 ± 0·07	0·44 ± 0·04	0·49 ± 0·08	0·48 ± 0·02

Effect of diet on spontaneous atherosclerosis in turkeys. By A. N. HOWARD*, G. A. GRESHAM and I. W. JENNINGS, *Department of Pathology, University of Cambridge*

Aortic rupture, which commonly occurs in 12–20 weeks old male turkeys given commercial rations, is associated with an atherosclerotic plaque at the site of rupture (Gresham & Howard, 1961). It is suggested that atherosclerosis leads to a weakening of the aortic wall which may then rupture. Another factor which may contribute is the high blood pressure of certain breeds.

In the present experimental series, the effect of dietary fat on the incidence and severity of aortic lesions has been investigated. Four groups of ten Broad Breasted Bronze poults, 1 day old, were placed on the following diets: (1) a commercial turkey starter ration (made and generously supplied by Spillers, Ltd, London), containing 25% protein, (7·5% animal protein), 2·5% fat, 3·5% fibre, 0·12% cholesterol; (2) diet 1 supplemented with 20% beef tallow; (3) diet 1 supplemented with 20% arachis oil; (4) a semi-synthetic diet containing 20% beef tallow which is known to produce atherosclerosis in rabbits (Howard & Gresham, 1961). Half the birds in each group were killed after 13 weeks and the remainder at 29 weeks.

Atherosclerosis (2–150 mm^2 area) of the aorta was seen in all birds examined, particularly in the mid-aortic region and including the portion of aorta between the sciatic and iliac arteries where rupture usually occurs. The lesions bore a close histological and histochemical similarity to those seen in man and were more extensive in the older birds. Although the mean area of diseased aorta was greater in groups (2) and (3) compared with group (1), the difference was not statistically significant since there was a very wide individual variation within each group. Extensive atherosclerosis was also seen with the semi-synthetic diet (group 4) and one animal in this group died of aortic rupture.

*Member of External Scientific Staff of the Medical Research Council.

Plasma cholesterol (150–250 mg/100 ml), which was the same in all groups, was relatively high compared with many other species. There was no difference in plasma phospholipids and glycerides between the groups.

It was concluded that the spontaneous atherosclerotic lesion occurring in turkeys provides a useful experimental model for the study of factors such as diet which may affect the disease.

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The digestibility of the carbohydrate constituents of S_{23} rye-grass cut at four stages of growth. By R. WAITE, M. J. JOHNSTON and D. G. ARMSTRONG, *Hannah Dairy Research Institute, Kirkhill, Ayr*

S_{23} rye-grass, cut at four stages of advancing maturity and dried on a commercial machine, was fed to sheep in a calorimetric experiment designed to determine net energy values (Armstrong, 1960). Analyses of the four grasses and of the faeces from one of each pair of sheep fed at three levels were made by methods described by Waite & Gorrod (1959*a*,*b*). The results are shown in the table together with the apparent digestibilities meaned over the three levels of feeding.

The carbohydrate constituents of four cuts of S_{23} rye-grass and their digestibility coefficients

	Cut 1 Young leafy		Cut 2 Late leafy		Cut 3 Flower emergence		Cut 4 Full seed	
	Percentage of dry matter	Percentage digestible	Percentage of dry matter	Percentage digestible	Percentage of dry matter	Percentage digestible	Percentage of dry matter	Percentage digestible
Hexoses	4·6		6·3		6·7		4·0	
Sucrose	3·2	100	2·7	100	2·8	100	2·8	100
Fructosan	6·0		2·8		1·8		3·8	
Cellulose	21·3	92	22·1	88	23·9	86	26·7	73
Hemicelluloses comprising:	15·8	91	18·9	85	19·4	82	25·7	57
Xylan	6·9	89	8·9	82	10·1	79	14·4	55
Araban	2·1	95	2·2	90	2·3	88	2·5	72
Glucan	1·9	96	2·2	95	2·0	93	1·7	74
Galactan	0·7	94	0·7	89	0·7	88	0·7	70
Aldobiouronics etc.	4·2	89	4·9	80	4·3	74	6·4	52
Pectin	2·4	72	2·1	56	2·2	50	2·2	36
Organic acids	4·2	94	4·9	88	4·6	89	2·9	68

The percentage of soluble carbohydrates varied only slightly in the four grasses. In the structural carbohydrates both the cellulose and xylan percentages increased with advancing maturity, the xylan content rising at a faster rate than that of cellulose.

The digestibility of all the structural carbohydrates was highest in the youngest grass (cut 1) and the most marked decline in digestibility occurred between cuts 3 and 4, after the grass had flowered. The fall in digestibility was greatest in the xylan, uronic acid and pectin fractions. Although the digestibility (73%) of the cellulose (here meaning the pure glucan) in cut 4 was considerably lower than in the other cuts, this compound in mature grass must still provide a major source of energy for the animal.

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The voluntary food intake of steers and wether sheep. By K. L. BLAXTER and R. S. WILSON, *Hannah Dairy Research Institute, Kirkhill, Ayr*

Previous experiments (Blaxter, Wainman & Wilson 1960, 1961) showed that the voluntary intake of food dry matter by adult sheep increased with increasing apparent digestibility of the fodders they were offered. Similar experiments were made with growing steers and the quantitative relationship between apparent digestibility of food and food intake was determined and compared with that found in sheep. Each of eight Aberdeen Angus-type steers initially weighing about 350 kg were given four roughages *ad lib.* for periods of 32–36 days. Food intake, apparent digestibility of the food and weight gain were measured. The results are shown in Fig. 1. It was concluded that for fodders of the same apparent digestibility no significant differences

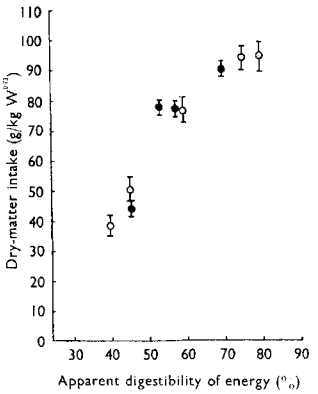

Fig. 1. Relation between the apparent digestibility of food energy and the voluntary intake of food by cattle (●) and by sheep (○). The ranges are the standard errors of means.

between adult sheep and growing cattle occurred in their mean voluntary intakes of food when these were expressed as g food/kg body-weight raised to the power 0·73 (g/kg $W^{0.73}$). The marked association between amount of food eaten and food quality was quantitatively similar in both species. The steers lost body-weight when given the poorest ration of oat straw and made appreciable gains (14·6 g/kg $W^{0.73}$) when given the best ration of dried grass.

Metabolism experiments in which maintenance requirements of energy have been determined show that sheep require less energy/kg $W^{0.73}$ to maintain them than do cattle (Blaxter & Wainman, 1961). Since the voluntary food intakes of the two species are closely comparable, sheep can attain a greater relative food level (Kleiber, 1935) than can cattle. It follows that when food is supplied *ad lib.* sheep are likely to be the more efficient converters of roughages into animal product.

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The calcium and phosphorus content of milk from hill ewes. By J. DAVIDSON and I. MCDONALD, *Rowett Research Institute, Bucksburn, Aberdeen*

Information on the calcium and phosphorus content of ewe's milk is sparse, especially for the Cheviot and Blackface breeds which are important in Scottish hill farming. Differences in mineral content among other breeds and changes with advancing lactation have been reported (Neidig & Iddings, 1919; Peirce, 1934, 1936; Godden & Puddy, 1935; Barnicoat, Logan & Grant, 1949).

Composite samples of milk taken periodically from ten Blackface and ten Cheviot sheep which lambed at the Rowett Institute in 1961 were analysed for Ca and P.

Calcium and phosphorus content of Cheviot and Blackface milk

Weeks after lambing	Total solids (g/100g)		Ca (g/kg milk)			P (g/kg milk)		
	Cheviot	Blackface	Cheviot	Blackface	Mean	Cheviot	Blackface	Mean
2	19·6	18·6	1·68	1·67	1·67	1·52	1·51	1·51
6	20·8	21·4	1·85	1·89	1·87	1·45	1·46	1·46
12	21·9	—	1·91	—	1·91	1·61	—	1·61
16	19·6	18·7	2·37	2·50	2·44	1·75	1·73	1·74
SE of mean (3 df)					±0·04			±0·01

Results in the table show that in both breeds there was a rise in mineral content of the milk at the end of lactation but there was no difference between breeds. The average concentrations for the whole lactation can be estimated as 1·84 g Ca and 1·53 g P/kg milk, by weighting the four mean figures in accordance with the normal trend in production during lactation (Wallace, 1948*a*,*b*,*c*; Thomson & Thomson, 1953; Hunter, 1957). Figures published previously by Godden & Puddy (1935) were of the same order but did not always show a rising trend through lactation.

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Digestion in the crop of the fowl. By W. BOLTON, *Agricultural Research Council Poultry Research Centre, West Mains Road, Edinburgh, 9*

Food in the crop may be acted upon by the bacterial flora, and by the enzymes secreted by the fowl and in the food.

Preliminary experiments had suggested that hydrolysis of starch to sugar, and the production of lactic acid, were the major changes taking place in the crop. In a study of these phenomena four adult male fowls were allowed access to food for 1 h, and then killed, 1, $2\frac{1}{2}$, $4\frac{1}{4}$ or 6 h after feeding, and the crop contents were analysed. The results, set out in Table 1, show that sugar content increased during the first $2\frac{1}{2}$ h, with little change in the other components studied. Thereafter the

Table 1. *Contents of the crop at various periods after feeding (dry-matter basis)*

		Time after feeding (h)			
	Mash	1	$2\frac{1}{2}$	$4\frac{1}{4}$	6
pH	5·9	6·1	6·1	5·2	4·5
Sugar (%)	1·3	4·0	3·7	3·3	1·6
Ethyl alcohol (mg/100 g)	9	13	6	18	109
Acetic acid (%)	0·01	0·02	0·01	0·10	0·13
Lactic acid (%)	0·2	0·2	0·2	1·7	3·2

sugar content and the pH fell, and the contents of alcohol, acetic and lactic acids increased. This is suggestive of a period wherein amylase activity predominates followed by a period when the multiplication of lactobacilli is the most important feature.

The One Hundred and Fiftieth Meeting of The Nutrition Society was held at the Zoological Society of London, Regent's Park, London, N.W.1, on Friday, 25 May 1962, when the following papers were read:

Arginine metabolism and nutrition in the chick. By G. H. SMITH and D. LEWIS, *University of Nottingham School of Agriculture, Sutton Bonington, Loughborough*

Arginine is an essential amino acid for the chick but the level required in the diet seems to vary with the overall amino acid composition of the diet. The apparent requirement ranges from 2·3% on diets mainly based on casein to around 0·9% on diets of a more usual commercial type. An examination of this problem has been made in terms of the metabolic interrelationships of arginine. Arginine has two major functions in the body; as a constituent of protein and as a donor of amidine groups for creatine synthesis. It is also attacked by arginase to yield urea and ornithine: as the urea cycle is not operative in avian species this may represent a wasteful pathway.