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ABSTRACTS OF COMMUNICATIONS

The One Hundred and Twenty-second Meeting of The Nutrition Society (Fiftyfourth of the Scottish Group) was held in the Physiology Lecture Theatre, Queen's College, Dundee, on Saturday, 7 February 1959, at 2 p.m., when the following papers were read:

Commercial breads as sources of vitamin E for rats as determined by the haemolysis test. By I. M. SHARMAN and PAMELA J. RICHARDS, Dunn Nutritional Laboratory, University of Cambridge and Medical Research Council

Various kinds of bread were compared in regard to their adequacy or deficiency as sources of vitamin E for rats, using a modification of the haemolysis test of György & Rose (1949). The rats were taken when about 90 g in weight, and were first fed upon bread 'Co', a commercial white loaf, supplemented only by halibut-liver oil to provide vitamin A. The animals grew at a moderate rate, and remained in apparent good health, on this diet. After about 7 weeks the haemolysis test was applied and was found to be positive in all the rats, with an average percentage haemolysis of 90, indicating a deficiency of vitamin E. The rats were then used repeatedly for testing other forms of bread. Thus, five rats were given bread 'Ho', reputed to include added wheat germ, for 14 days. The average percent haemolysis now fell to 0.5, indicating adequacy of vitamin E. The animals were then given bread 'Co' again until their erythrocytes once more exhibited a liability to haemolysis. Six rats were next divided into two groups of three animals each, as indicated in Table 1, and given alternately another bread 'Al', baked from wholemeal flour. The results indicated that this bread also was effective in protecting the erythrocytes against haemolysis.

Table 1.	Percentage i	haemolysis	of e	erythrocytes j	from rats _.	fed	on b	read	'Co'	and	'Al	,
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Rat		Haemoly Days o	rsis (%) n diet		Haemo Days	on diet
no.	Bread	o	14	Bread	14	34
I	ſ	100	90	٦	14	0
2	≻,Co,	100	90	'Al'	8	0
3	J	98	91	J	93	٥
4	ſ	96	2	ſ	96	82
5	'Al'	93	9	≻,Co,	90	69
6	J	93	3	J	78	66

Thus the haemolysis test provides a convenient method for distinguishing the adequacy or otherwise of commercial breads as sources of vitamin E.

REFERENCE

György, P. & Rose, C. S. (1949). Ann. N.Y. Acad. Sci. 52, 231,

Evaluation of faecal nitrogen as an index of herbage digestibility by means of a continuous digestibility trial. By J. F. D. GREENHALGH (DRUMMOND JUNIOR FELLOW), I. MCDONALD and J. L. CORBETT, Rowett Research Institute, Bucksburn, Aberdeen

Indirect estimates of herbage digestibility derived from faeces nitrogen concentrations, combined with estimates of faeces output, allow the calculation of the herbage consumption of grazing animals. Regression equations for predicting digestibility are obtained with animals given cut herbage and are most accurate if derived with, and applied to, one type of herbage (Raymond, Minson & Harris, 1956).

For 54 consecutive days herbage was cut from a grass-clover sward and given in amounts providing 16 lb of dry matter daily to each of three steers of 800 lb live weight. The animals were confined in metabolism stalls and fitted with faeces-collection equipment. For the last 45 days organic matter (O.M.) digestibility and percent nitrogen in faeces O.M. were measured for each animal for each 24 h period. During the trial digestibility declined from 80 to 66% and faecal-nitrogen concentration from 4.8 to 1.7%.

Eight regressions of digestibility on faecal-nitrogen concentration were calculated, from the unit 24 h observations on single animals and from averages over three animals, and similarly from averages over successive 2-, 3- and 5-day periods. The residual variances (R.V.) of these regressions would have varied inversely as the number of unit observations in each average, if these had been independent, but in fact averaging over three animals reduced the R.V. by a factor of 1.4 not 3. Averaging over different periods did have approximately the expected effect on R.V., except that there was an excessive reduction from 1-day to longer periods, suggesting that end-of-period errors may have been affecting 1-day observations appreciably.

The following regression of percent digestible O.M. (y) on % N in faeces O.M. (x) was calculated from fifteen averages, each over three animals and 3 days:

 $y = 14\cdot 3x - 1\cdot 6x^2 + 48\cdot 0$ (residual standard deviation $= \pm 0.85$ percentage units). The s.E. of a digestibility coefficient predicted by this equation from an average faeces nitrogen value for three animals over 3 days, and in the range $2\cdot 0-4\cdot 5\%$ N, was calculated to be $\pm 0\cdot 9$ units. From the observed behaviour of the R.V., s.E. of prediction for one animal over 3 days and for one and three animals over 9 days were calculated as $1\cdot 1$, $0\cdot 7$ and $0\cdot 6$ units respectively. Although a s.E. of $\pm 0\cdot 7$ units represented a percentage error of $\pm 1\%$ for mean digestibility (73%), the factor used in the estimation of herbage consumption is indigestibility (27%), for which the percentage error would be $\pm 2\cdot 6\%$. This value is smaller than any previously reported.

REFERENCE

Raymond, W. F., Minson, D. J. & Harris, C. E. (1956). Int. Grassld Congr. VII. Palmerston North, N.Z., p. 123.

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Micro-organisms in the rumen of goats eating an artificial diet deficient in riboflavin, lysine and tryptophan. By E. C. OWEN and R. PROUDFOOT, Hannah Dairy Research Institute, Kirkhill, Ayr

In the experiments of Crossland, Owen & Proudfoot (1958) samples of rumen contents were taken from four lactating goats on a diet of zein, potato starch, straw pulp, treacle, urea and a salt mixture with long oat straw as roughage. Such a diet is deficient in the amino acids, lysine and tryptophan and in the vitamins of the B group, but the lactating goat can obtain the missing nutrients from the symbiotic organisms in its rumen (Edwards & Darroch, 1956; Crossland et al. 1958). Our later experiments (Crossland et al. 1958) showed that riboflavin synthesis was occurring in the rumen of all four goats. The present report refers to the microscopic examination of the rumen contents of these latter four goats. Frequent previous examinations of the rumen contents of the goats while they were eating a winter diet or were grazing had shown the invariable presence of oligotrich and holotrich protozoa, of Selenomonas ruminantium, of Sarcina sp., of streptococci both free and adherent to cellulose and of Oscillospira guilliermondii. All these organisms stain readily with iodine. Of the four goats studied by Crossland et al. (1958) only one ate all its diet readily and would have eaten more had more been offered to it. Only in this goat were all the above organisms found. In the goat with the poorest appetite for the artificial diet sarcinae were hard to find and Selenomonas ruminantium could not be found. Sarcinae were readily found in the other three goats. Iodophilic bacteria attached to cellulose, and holotrich and oligotrich protozoa were found in the rumen contents of all the goats. Free iodophilic streptococci were present in all four animals though they were scarce in one of them.

Oscillospira guilliermondii was found only in the animal with the best appetite for the diet.

We wish to thank Mrs A. M. Nisbet for the photomicrographs shown at the meeting.

REFERENCES

Crossland, A. C., Owen, E. C. & Proudfoot, R. (1958). Brit. J. Nutr. 12, 312. Edwards, D. C. & Darroch, R. A. (1956). Brit. J. Nutr. 10, 286.

The assay of antirachitic activity in man. By H. G. MORGAN, Queen's College, Dundee, W. C. THOMAS, Jr., C. E. BILLS and J. E. HOWARD, Johns Hopkins Medical School and University, Baltimore, U.S.A. (introduced by J. M. STOWERS)

Method. The antirachitic potency of test material, given as a single oral dose, is compared with that of standard cholecalciferol in young rachitic rats by a line test as modified by Dr C. E. Bills.

Observations on human serum. Antirachitic activity shows high persistence in vitro when lyophilized or stored at 30°. There is no detectable loss after dialysis or ultrafiltration, implying 'binding' by protein. The activity is shown by starch-block

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electrophoresis to reside mainly in the α -globulin fraction. A similar distribution of activity is found also in idiopathic steatorrhoea (one patient), renal hypophosphataemia (one patient), and in normal serum after in vitro incubation with ergocalciferol.

Normal levels in man are 1-3 i.u./ml serum. An increased serum level is found 8 h after a first oral dose, and maximum levels are reached after some weeks. Raised serum levels persist for long periods; thus in a case of ergocalciferol overdosage, the serum level fell only from 30 to 6 i.u./ml in 7 months after stopping the intake of the vitamin. Much individual variation is found both with the serum levels for a given dosage, and with the degree of hypercalcaemia resulting.

Table 1. Antirachitic activity of serum from hypoparathyroid patients receivingergocalciferol

Patient	Daily dose of vitamin D ₂ (mg)	Duration Rx	Serum calcium (mg/100 ml)	Assay (i.u./ml)
E.P.	1.52	ı year	9.2	40
A.M.	1.22	2 years	8.4	40
L.L.	2.2	6 months	10.0	30
A.C.	2.2	5 years	10.0	48
M.R.	2.2	2 years	0.1	50
H.C.	5.0	15 months	9.6	18
L.C.	5.0	3 years	8.6	8

Observations in disease. Hypocalcaemia from idiopathic steatorrhoea: of three such patients examined, only one had an abnormally low level; the second had a normal, and the third a raised serum assay level. These findings suggest that an impaired absorption of vitamin D is alone not sufficient to explain the hypocalcaemia of idiopathic steatorrhoea. Normal serum levels were found in renal hypophosphataemia ('vitamin-D resistant rickets'), in hypercalcaemic states due to hyperparathyroidism, malignancy and sarcoidosis, and in two infants with the 'mild' form of idiopathic hypercalcaemia.

In the interpretation of these findings, we must remember that the test animal was the rat, a creature that differs greatly from man in its response to some antirachitic substances, such as AT 10 (dihydrotachysterol).

The effect of low-level feeding of chlortetracycline on rumen fermentation in early-weaned calves. By T. R. PRESTON, P. K. DINDA and N. A. MacLeod, Rowett Research Institute, Bucksburn, Aberdeen

Two pairs of Friesian bull calves were reared on the early weaning system described by Preston, McLeod & Dinda (1959). One member of each pair was fed a concentrate mixture containing 8 mg chlortetracycline/lb, the other was fed the same mixture without antibiotic. Samples of rumen liquor were obtained by stomach tube at weekly intervals from 2 weeks to 12 weeks. The calves were deprived of food from 5 p.m. on the night prior to sampling until 9 a.m. the following morning when they were fed an amount of concentrate which it was estimated they would eat up within

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1 h. Level of feeding was equalized within pairs according to the metabolic live weight $W^{0.73}$ of the calves.

The mean pH and the mean concentration of steam-volatile fatty acids in strained rumen liquor for the 10-week period are set out in Table 1.

Table 1. Mean pH of, and mean steam-volatile fatty-acid concentration (m-equiv. 100 ml) in, rumen liquor, before and 4 h after feeding, from control and chlortetracycline-supplemented calves sampled weekly between 2 and 12 weeks old

	pH				F	Fatty-acid concentration				
	Control calf (a)	Supple- mented calf† (b)	Difference Value S.E.		Control calf (c)	Supple- mented calf [†] (d)	Difference (c-d) Value S.E.			
Before feeding: Pair A	7.48	7.71				• •				
Pair B	7.46	7.66	0·23 0·20*	$_{\pm 0.12}^{\pm 0.12}$	32·9 49·3	24·3 30·6	8·6** 18·7**	$^{\pm 2.3}_{\pm 5.9}$		
4 h after feeding:										
Pair A Pair B	5.94 5.33	6·42 5·95	0·48* 0·62*	$_{\pm 0.15}^{\pm 0.15}$	89·9 129·5	85·1 97·8	4·8 31·7***	±3·3 ±7·2		
*Signif ***Signif	icant differe icant differe	nce, $P < 0.05$. nce, $P < 0.001$		**Significant †Given 8 m	difference, P < g chlortetracycli	: 0.01. ine/lb concen	trate.			

Both before and 4 h after feeding the rumen liquor from the calf which was fed concentrates supplemented with chlortetracycline had a higher pH and a lower concentration of steam-volatile fatty acids than the liquor from its control mate.

REFERENCE

Preston, T. R., McLeod, N. A. & Dinda, P. K. (1959). Anim. Prod. 1, 13.

Differences in yield and composition between first and second lactations. By F. E. HYTTEN, Obstetric Medicine Research Unit (M.R.C.), Maternity Hospital, Aberdeen

The yield and fat content of a 24 h collection of 7th-day milk have been measured in both the first and second lactations of 120 women. For fifty of these women the milk was also analysed for lactose and nitrogen.

Yield. The mean yield in the first lactation was 445 ml compared to 514 ml in the second lactation, a highly significant difference, and there was a high correlation between yields in the two lactations (r = 0.618). The variations in yield were considerably less in the second lactation than in the first.

Fat content. The mean fat contents were $3 \cdot 03$ and $3 \cdot 37$ g/100 ml; the rise of $0 \cdot 34$ g/100 ml in the second lactation was highly significant. The correlation between fat contents in the two lactations was low ($r = 0 \cdot 287$) and there was no difference in the variation.

Lactose content. The mean lactose content in the first lactation was 6.40 g/100 ml and 6.52 g/100 ml in the second, a statistically significant difference.

Total nitrogen. The mean nitrogen content in the first lactation was 296.9 mg/ 100 ml and 288.6 mg/100 ml in the second lactation; the fall was statistically significant. The higher sugar content and the lower nitrogen content suggest that 7th-day milk in the second lactation is more mature, equivalent to 9th- or 10th-day milk in the first lactation; this advanced maturity will explain some but by no means all the increase in yield and fat content.

The improved and less variable yield and the higher fat content, together with the somewhat more rapid maturation of the milk suggest an overall increase in the efficiency of lactation at the second attempt.

Effect of reduced intake of greenstuff on β -carotene and vitamin A in goat colostrum. By E. C. OWEN and R. PROUDFOOT, Hannah Dairy Research Institute, Kirkhill, Ayr

Chanda & Owen (1952) and Chanda (1953) found abundant β -carotene in the colostrum of British Saanen goats and reported that, in a goat which was a persistent milker, there was no carotene in the milk even 2 days before parturition at which time milk secretion ceased. When however 2 days later at parturition this goat came into milk again the colostral secretion was yellow and contained β -carotene (Chanda & Owen, 1952). In the present work colostrum samples were again examined after a period when the intake of greenstuff by the goats had been much restricted. The milk of several of the goats was analysed for some time before parturition and was found to contain vitamin A but no carotene. At parturition nine goats all showed a relatively large concentration of vitamin A in the first colostrum, and carotene was detected in the colostrum had been subjected to prepartal milking but before parturition carotene was not found in the milk and the prepartal milk was poorer in vitamin A than was the colostrum.

There was thus a contrast between the abundant carotene in the colostrum of the goats in 1952 and 1953 and the paucity of carotene in the colostrum in the present work though the goats now studied were the progeny of the earlier ones. It would appear that an abundant intake of pasture in the earlier experiments was responsible for higher outputs of carotene and this explanation is borne out by the fact that in the later experiments when the intake of greenstuff and consequently the intake of carotene was lower the concentration of vitamin A in the colostrum was also lower. In the 1952-3 experiments, samples of colostrum from eight goats averaged $_{385} \mu g$ vitamin A and $_{27} \mu g \beta$ -carotene per 100 ml. In the present experiments, samples of colostrum from eight goats averaged 112 μg vitamin A and 2 $\mu g \beta$ -carotene per 100 ml. Included in this latter average are figures for the colostrum from two goats in which the β -carotene was not measurable and in all of the samples in the present work carotene, when present, was measurable only in the initial colostrum whereas in 1952-3 carotene was measurable for over 3 days after parturition.

Thanks are due to Miss M. Lightbody for carotene and vitamin A analyses.

REFERENCES

Chanda, R. (1953). J. agric. Sci. 43, 54. Chanda, R. & Owen, E. C. (1952). Biochem. J. 51, 404. The One Hundred and Twenty-fifth Meeting of The Nutrition Society was held at Queen Elizabeth College, University of London, on Friday, 29 May 1959, at 4.15 p.m., when the following papers were read :

Pancreatic fibrosis and calcification in Uganda Africans. By A. G. SHAPER, (introduced by Z. A. LEITNER), Makerere College Medical School, Kampala, Uganda

Protein deficiency has been considered to be responsible for the pancreatic calcification seen in young people in Indonesia (Zuidema, 1955). Diffuse pancreatic lithiasis has been demonstrated in eleven adult Africans in Kampala, Uganda, and these cases are described and the possible relationship to protein malnutrition is discussed. Two patients, aged 21 and 24 years, presented with severe malabsorption states; steatorrhoea, hypoalbuminaemia, skin and hair changes, oedema and fatty liver were present in both subjects. Their nutritional background was fairly good although low in protein. A second group of five males aged 23-45 years presented with diabetes mellitus and were shown to have pancreatic calcification. Steatorrhoea was present in two of these patients but with no clinical evidence of malabsorption. The dietary history was poor in four of this group, the remaining subject having a good protein intake. The third group comprised four subjects in whom the pancreatic calcification was a finding at routine autopsy examination.

The histological appearances in five cases suggest a low-grade inflammatory reaction which has given rise to replacement fibrosis of the exocrine elements. All cases show varying degrees of fibrosis, the earliest phase of which appeared to be periductal and perilobular, followed by interacinar fibrosis until only isolated remnants of the exocrine tissue remained. The islets showed mainly hypertrophy, hyperplasia, fibrosis and hydropic degeneration. The presence of dilated ducts, ductular secretion and calcification and acinar dilatation suggests that the cause may be obstructive in nature.

While there is evidence that severe protein deficiency in children and adults can depress pancreatic function and produce fibrosis in the pancreas, the demonstration of pancreatic fibrosis and calcification in an area where protein malnutrition is common is only circumstantial evidence of a relationship between the two conditions. It may be that a variety of agents may produce lesions in a pancreas rendered more susceptible to injury by low protein intakes, but the possibility must be considered of altered protein metabolism leading to abnormally viscid secretions and ultimately to pancreatic lithiasis.

I am grateful to Professor Kenneth Hill, Royal Free Hospital Medical School, for his report on the pancreatic histology.

REFERENCE

Zuidema, P. J. (1955). Docum. Med. geogr. trop. 7, 229.