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ABSTRACTS OF COMMUNICATIONS

The Eighty-second Meeting of The Nutrition Society was held at the National Institute for Research in Dairying, Shinfield, nr Reading, on Tuesday, 28 July 1953, at 11 a.m., when the following papers were read :

Ascorbic Acid in Public and Preparatory School Meals. By S. S. BROWN and JOAN F. CHURCH, *Food Research Laboratory, Messrs Peter Merchant Ltd., 16 Bell Street, Henley-on-Thames, Oxon.*

In view of the conflicting opinions regarding the daily requirements for vitamin C, data accumulated during the last 2 or 3 years from nutritional surveys on public and preparatory school feeding is of interest.

From the earliest results it was evident that the National Research Council of America figure of 100 mg/day could not be provided from natural foodstuffs at an economic cost but that the B.M.A. Committee recommendation of 20 mg/day would be exceeded given reasonable variety and efficient kitchen practice. No seasonal variation in total vitamin C content of the diet can be established, nor can any relationship be found between vitamin A and vitamin C content of the diet. It is evident, however, that in order to attain a good level of vitamin C intake some provision of vitamin C must be made at either breakfast or the main evening meal or, preferably, at both.

Term	Public schools		Preparatory schools	
	Vitamin C (mg)	Vitamin A (i.u.)	Vitamin C (mg)	Vitamin A (i.u.)
Spring	33	3729	24	2492
	19	3906	24	2438
	57	4696	49	2683
	36	2468	53	1232
	36	3552		
	42	3138		
Summer	22	6189	39	3783
	56	2121	28	4844
			61	2520
			25	2403
Autumn	38	3146	58	4184
	28	2448	48	3183
	48	2879	38	5775
	55	7356	33	3108

Each survey was carried out over a period of 4 consecutive days. Vitamin C content of vegetables and fruits was determined at the time of serving by the titrimetric method of Harris & Olliver (1942). The figures reported in the table are the mean daily intake over the 4-day period from food supplied by the school kitchen. Additional amounts might be obtained from other sources but these are

unlikely to be very large. One instance of a daily dose of 50 mg by capsule has been observed, but this does not afford any evidence in favour of such supplementation.

REFERENCE

Harris, L. J. & Olliver, M. (1942). *Biochem. J.* **36**, 155.

The Biological Value of Fish Meals. By A. E. BENDER, D. S. MILLER and ELIZABETH J. TUNNAH, *The Crookes Laboratories Ltd, Park Royal, London, N.W.10*

The value of white fish meal as an animal-protein concentrate for the feeding of livestock is well recognized and suggestions have been made that it should be used to supplement human diets. The method of Bender & Miller (1953) showed the net protein utilization (biological value \times digestibility) of four British fish meals to be 36 (fertilizer grade), 50 (dogfish), 62 and 58 (white fish), and three South African 32, 36 (oily type) and 73 (white fish). In view of the wide range, samples were prepared in the laboratory from whole fish (cod) under controlled drying conditions, and these materials were assayed in the same manner. The results showed them to be considerably superior and indicate that damage can be effected by heat, namely (a) vacuum dried 84, (b) dried at 50° 82, (c) dried at 100° 70. Extracted proteins which had been acetone-dried showed similar results, namely (d) herring 76, (e) cod 87, (f) cod heated to 45° for 48 h 80, (g) cod heated to 100° for 48 h 49. It seems likely, therefore, that all the commercial meals tested, with the possible exception of one from South Africa, had been damaged by heat during manufacture, and that for the best results the drying temperature should be below 100°. A brief study of commercial methods of drying fish makes this result unsurprising (e.g. Lovern, 1951).

The low values were apparently not due to impaired digestibility as the poorest was 86% digestible. From the colour of the commercial meals it would appear that the damage was of the Maillard type, but supplementation with lysine, the amino-acid most likely to be affected, did not show a deficiency of this amino-acid (Table 1) which was confirmed by lysine determinations.

Table 1. *Supplementation of fish meals with lysine*

	Lysine % CP	Net utilization	
		Unsupplemented	5% L-Lysine
British fish meals 1	5.4	36	35
4	5.4	62	60
5	5.5	55	57
South African 2	—	29	28

We are indebted to Dr K. J. Carpenter of the Rowett Research Institute for the lysine estimations.

REFERENCES

Bender, A. E. & Miller, D. S. (1953). *Biochem. J.* **53**, vii.
 Lovern, J. A. (1951). *Fishing News*, no. 1987.

The Function of the Intra-glandular Branches of the Salivary Ducts.

By C. RUTH HILL and G. H. BOURNE, *Department of Histology, London Hospital Medical College, London, E.1*

The intra-glandular branches of the salivary ducts have long been regarded as simple passive carriers of the acinar secretion yet it was suggested as long ago as 1866 that they had a secretory function.

Simple esterase preparations of the salivary glands show greater activity in the ducts than in the acini. The substrate used for this reaction (naphthoic acid anilide acetate) is also hydrolysed by chymotrypsin and possibly by other proteolytic enzymes as well. There is evidence elsewhere that the tubular parts of the salivary glands may secrete a protease into the saliva. Alkaline phosphatase was found only in the capillary vessels underlying the ducts and acini, but the duct cells contained much more acid phosphatase than the acinar cells. The former were also unusually rich in mitochondria which are now linked with aerobic cellular metabolism. This implied high metabolic activity of the duct cells was confirmed by the localization in them of considerable succinic-dehydrogenase activity. This suggested that they may be secreting into, or absorbing from, the duct lumen.

Secretion. Although the duct cells contained abundant esterase there was no cytological evidence of its secretion into the saliva. They also contained mucopolysaccharides (periodic-Schiff reaction), and there was cytological evidence of secretion of this material into the lumen. However, the amount secreted appeared very small compared with the metabolic state of the cells as judged by the mitochondrial and succinic-dehydrogenase preparations.

Absorption. All cells which are highly active in absorption of solids have alkaline phosphatase activity at their free borders—the duct cells have no alkaline phosphatase activity at all. In the kidney, tubule cells absorbing solids have a high succinic-dehydrogenase activity and alkaline phosphatase activity in their free borders. Those absorbing water (e.g. Henle's loop) have a high succinic-dehydrogenase activity but no alkaline phosphatase. By analogy it is suggested that the salivary duct cells are transferring water. About 1 l. of saliva is produced a day and to enable this quantity to be produced the duct cells must be passing water into the lumen, not removing it. It is suggested therefore, that the salivary duct cells are engaged in diluting the salivary solids produced by the acini and they may also be adding small quantities of polysaccharides and proteolytic enzymes to the saliva. It is of interest that pancreatic duct cells contain a high esterase and succinic-dehydrogenase activity and they may therefore have a similar function to that of the salivary duct cells.

Autoradiographic Studies of Calcium Metabolism in Bones and Teeth.

By D. H. TOMLIN, *Physics Research Department, University of Reading*, and KATHLEEN M. HENRY and S. K. KON, *National Institute for Research in Dairying, University of Reading*

The effects of pregnancy and lactation on the bones and teeth of rats have been studied by the autoradiographic method, using ^{45}Ca , previously described (Tomlin, Henry & Kon, 1953).

Three groups of six female rats were given, when 3 months old, the following treatments: (1) mated, (2) daily subcutaneous injection of $0.5\ \mu\text{g}$ hexoestrol dissolved in $0.1\ \text{ml}$. arachis oil, (3) daily subcutaneous injection of $0.1\ \text{ml}$. arachis oil. As the animals in group (1) were mated they, and corresponding animals in groups (2) and (3), were given a diet consisting of wheat and whole dried milk supplemented with CaCO_3 . The diet contained 0.2% Ca, 0.4% P and about $5\ \mu\text{c}$ $^{45}\text{Ca}/100\ \text{g}$. At the end of both pregnancy and lactation three rats were killed in each group. Longitudinal sections of femora, humeri, tibiae and incisors were prepared and autoradiographs were made.

The bones of animals killed at the end of pregnancy showed surface deposits characteristic of the normal growth mechanism (Tomlin *et al.* 1953). In the femora and tibiae in particular, endosteal deposits extended from the epiphyseal plate at the growing end of the shaft well into the diaphyseal region. Compared with the controls, animals given hexoestrol had more radioactive cancellous bone, whereas the mated animals had less of this spongy bone and a much less intense radioactive region in the growth zone immediately below the epiphyseal plate.

The bones of the mated animals killed at the end of lactation showed a distinct loss of cortical bone from the endosteal surfaces. The radioactive deposits, which were a prominent feature in these regions in the control group, had largely disappeared, and the thickness of the cortex was noticeably reduced. Periosteal accretion of new bone appeared unaffected.

Autoradiographs prepared from the incisor teeth of animals that had undergone pregnancy and lactation showed variations of activity in different growth zones which did not appear in the controls.

It is concluded that mineral reserves held in the cancellous bone of the mothers were sufficient to provide for the young *in utero* but that withdrawal of cortical bone was necessary during lactation. This withdrawal came entirely from the medullary surface of the cortex.

REFERENCE

Tomlin, D. H., Henry, K. M. & Kon, S. K. (1953). *Brit. J. Nutr.* **7**, 235.

The Partial Vitamin E Activity of Malachite Green. By T. MOORE, I. M. SHARMAN and R. J. WARD, *Dunn Nutritional Laboratory, University of Cambridge and Medical Research Council*

In a previous communication (Moore, Sharman & Ward, 1953*a*) we reported that methylene blue can replace vitamin E in preventing brown discoloration of the uterus and degeneration of the kidney tubules in rats deprived of the vitamin. Under the experimental conditions which were chosen, however, we failed to repeat the finding of Dam & Granados (1952) that the dye can prevent resorption gestations in deficient rats. In further tests we found that thiodiphenylamine and Bindschedler's green, which does not contain sulphur, could also prevent brown uterus (Moore, Sharman & Ward, 1953*b*). Several other redox dyestuffs, and a series of anti-oxidants of the hydroquinone type, were ineffective at the levels used.

A further series of experiments has now shown that the triphenylmethane dyestuff, malachite green, and the corresponding leuco derivative, also prevent brown uterus when included as 0.1% of the diet. New methylene blue, which differs from the ordinary form in having an additional methyl group and ethyl amino-groups instead of the two dimethyl amino-groups, was also effective. Ascorbic acid (0.5%) and cystine (0.5%) were ineffective. No protection was given by a series of purine compounds, including xanthine, hypoxanthine, theophylline, adenine and theobromine given at the level of 0.1% of the diet. These substances were tested in view of the finding of Hove & Harris (1947) that xanthine resembles α -tocopherol in increasing the utilization of casein by rats.

The partial vitamin E activity of certain redox dyestuffs is probably related to their well-known ability to take part in enzyme systems. The protection given by malachite green confirms the previous conclusion that the presence of sulphur is not essential for activity. The relationship between chemical structure and physiological activity, however, requires further study.

REFERENCES

- Dam, H. & Granados, H. (1952). *Acta pharm. tox., Kbh.*, **8**, 47.
Hove, E. L. & Harris, P. L. (1947). *J. Nutr.* **34**, 571.
Moore, T., Sharman, I. M. & Ward, R. J. (1953*a*). *Biochem. J.* **53**, xxxi.
Moore, T., Sharman, I. M. & Ward, R. J. (1953*b*). *Biochem. J.* **54**, xvi.

Film Strips for Nutrition Teaching. By VERONICA S. CARMICHAEL, DOROTHY F. HOLLINGSWORTH and JANE O'H. MAY, *Scientific Adviser's Division, Ministry of Food, Great Westminster House, Horseferry Road, London, S.W.1*

These film strips are complementary to the three films of the same titles prepared in 1952 by Central Office of Information for the Ministry of Food with the co-operation and advice of the Ministries of Health and Education. They are intended to present simple nutrition, based on the Three Food Group method, to the older children in Secondary Modern Schools, who are pursuing a non-academic course.

Nutrition teaching is normally given as part of the course in housecraft, and the aim in these strips has been to give a practical positive approach to the need for selecting, preparing and cooking good food. The teaching must necessarily be gradual and spread over the course. The strips can be used, a few frames at a time, as the basis of teaching, and all the nutritional knowledge gained from them and from instruction in practical cookery can be animated and recapitulated by use of the films.

Background teaching notes have been prepared for teachers. The strips can be used for audiences of different ages and standards of education and the fundamental teaching adapted according to their interests. The successful use of the strips depends upon the ability of the teacher to interpret and expand the concepts illustrated by the pictures.

An appreciation of the knowledge of nutrition can only be within the reach of a few. The part played by food in the maintenance of health is the intimate concern of all. An attempt is made to bring within the reach of the future generation of parents, and of those practically concerned with the care of children in schools, in homes and hospitals, knowledge of the benefits of food based upon scientific principles.

The film strips may be obtained from Information Finance Branch, Ministry of Food, Great Westminster House, Horseferry Road, London, S.W.1. Price 10/- each.

Dietary Liver Necrosis and Testicular Degeneration. By J. A. LAING and D. E. TRIBE, *University of Bristol Veterinary School*

When investigating the causes of sterility in bulls one of us (J.A.L.) was impressed with the frequency with which, on post-mortem examination, such animals showed liver as well as testicular damage. In the majority of cases the liver lesion was that of telangiectasis, the aetiology of which in bovines is obscure. Although concurrent liver and testicular damage has previously been reported (e.g. Naftalin, 1951) the interrelationship of the two lesions has not, in fact, been previously studied. It was with this intention that the present investigation was begun.

In order to produce concurrent liver and testicular damage an experiment was devised which was a slight modification of that of Naftalin who consistently produced lesions of this nature. The response of our hooded 'Lister' rats, however, differed from that of the albino rats used by Naftalin. It was demonstrated by the dialuric-acid haemolysis test that our rats were deficient in vitamin E but on low-protein diets we could only sporadically produce the massive liver necrosis that we had expected and only one rat died as a result of this. One possible reason for this appears to be the different caseins used by ourselves and Naftalin, for Naftalin (1953) has recently indicated the critical aetiological importance of casein type in this connexion. However, other interesting macroscopical and microscopical changes were observed. The livers were consistently larger and the spleens smaller in the experimental animals than in the controls. As judged by the weight of the

testicles damage to these organs did not occur until the rats had reached an age of about 100 days. In addition, unilateral testicular atrophy occurred in some rats. Transient alopecia was seen in some animals and it is of particular interest to note that reddening of the white hairs of the coat was a consistent feature of rats after they had been on the low-protein and vitamin E-deficient diet for 6–8 weeks.

REFERENCES

- Naftalin, J. M., (1951). *J. Path. Bact.* **63**, 649.
Naftalin, J. M., (1953). Personal communication.

The Production of Tetany in Calves Fed on Milk Diets. By W. H. PARR and RUTH ALLCROFT, *Ministry of Agriculture, Veterinary Laboratory, Weybridge, Surrey*

Huffman & Robinson (1926) observed that calves cannot be reared to maturity on whole milk alone due to the onset of tetany during growth. This was found to be often associated with hypocalcaemia. Later, evidence was produced which showed it to be due to hypomagnesaemia, with normal serum-calcium levels (Duncan, Huffman & Robinson, 1935; Huffman & Duncan, 1936).

The present studies were made on five groups of four calves (Ayrshire and Friesian), 1 week old at the beginning of the experiment. A group fed whole milk at approximately 10% of their body-weight per day (nominal magnesium intake 10 mg/kg body-weight) showed hypomagnesaemia after 3 weeks and tetany after 10 weeks. An energy supplement, flaked maize, did not prevent these symptoms. A supplement of acid-extracted hay appeared to prevent the occurrence of tetany, but not of hypomagnesaemia. Calves receiving whole milk at approximately 20% of their body-weight showed hypomagnesaemia after 3 weeks but did not develop tetany until after 22 weeks, when they were no longer able to drink the quantity of milk necessary to maintain their intake at the 20% level. Another group given artificial milk at 10% of their body-weight per day (nominal magnesium intakes 5–10 mg/kg body-weight) also developed tetany. All animals received additional iron, copper and vitamins A and D (Radiostoleum).

In all cases where symptoms of tetany occurred the animals showed both low serum-calcium and low serum-magnesium values. In some animals serum-magnesium levels fell to 0.4 mg/100 ml., but clinical symptoms did not appear until the serum-calcium levels also fell. The mean values for eight calves which died or were killed in tetany were 6.2 mg/100 ml. for calcium and 0.6 mg/100 ml. for magnesium. The clinical symptoms were those usually associated with hypomagnesaemia, namely hyperaesthesia followed by convulsions, during which salivation, exophthalmos and opisthotonos were evident. In blood samples sent by veterinary surgeons to this Laboratory from field cases of suspected calf tetany, the usual finding is a hypocalcaemia together with a hypomagnesaemia.

In the above experiment, a daily dose of 1 oz. $MgCO_3$ restored magnesium levels to the normal physiological range, but they fell again when dosing was stopped. The same amounts of $CaCO_3$ and Na_2CO_3 had no apparent effect.

REFERENCES

- Huffman, C. F. & Robinson, C. S. (1926). *J. biol. Chem.* **69**, 101.
 Duncan, C. W., Huffman, C. F. & Robinson, C. S. (1935). *J. biol. Chem.* **108**, 35.
 Huffman, C. F. & Duncan, C. W. (1936). *J. Dairy Sci.* **19**, 440.

The Effect of a High Plane of Nutrition on the Composition of Milk.

By J. A. F. ROOK, *Hannah Dairy Research Institute, Kirkhill, Ayr*

This investigation is part of a more extensive study of the metabolism of the cow, as affected by the plane of nutrition, throughout the entire lactational period. Four cows were placed on experiment at the end of their 1st month of lactation and were subjected to four feed treatments over successive 32-day periods, according to a Latin square design. The levels of treatment were as follows:

(1) A normal ration, calculated from the performance of each cow prior to the first experimental period, on the basis of Woodman's (1948) tables. To allow for the decline in milk yield throughout lactation, the ration was decreased by 1.25 lb. S.E. and 0.25 lb. P.E. at the end of each period. (2) A normal ration + 0.8 lb. P.E. (containing 0.6 lb. S.E.). (3) A normal ration + 3.5 lb. S.E. (4) A normal ration + 0.8 lb. P.E. (containing 0.6 lb. S.E.) + 3.5 lb. S.E.

The mean effects of the energy and protein supplements on the major milk constituents, together with their significance, are recorded in the table. The effects of the energy and protein supplements were additive and there was no significant interaction.

Constituent	Mean value for treatment 1	Protein supplement		Energy supplement	
		Mean effect	<i>P</i> *	Mean effect	<i>P</i> *
Fat (%)†	4.14	+0.04	Not significant	-0.08	Not significant
S.n.F. (%)†	8.89	+0.04	Not significant	+0.18	0.001
Lactose (%)‡	4.71	-0.02	Not significant	±0.00	—
N (%) × 6.38†	3.21	+0.06	0.08	+0.16	0.0004
Total N (mg.%)†	503	+10	0.08	+25	0.0004
Casein N (mg.%)‡	399	+4	Not significant	+18	0.001
Sol. protein N (mg.%)‡	89	+1	Not significant	+6	0.007
Non-protein N (mg.%)‡	21	+4	0.00	±0	—

* *P* = probability that a difference as large as the observed difference would have arisen by chance.

† From data for the last 16 days of each period.

‡ From data for the last 8 days of each period.

In contrast to the findings of Rowland (1946), the change in S.n.F. content was due entirely to an increase in the casein and soluble protein. The marked effect of the protein supplement on the non-protein nitrogen content indicates that the secretion of milk provides an alternative route for the excretion of nitrogenous metabolites.

These results show that the addition of an energy supplement to the rations recommended by Woodman (1948) for dairy cattle gives an increase in the S.n.F. content of milk: the small effect of the protein supplement may be accounted for by its energy content.

REFERENCES

- Rowland, S. J. (1946). *Dairy Ind.* **11**, 656.
Woodman, H. E. (1948). *Bull. Minist. Agric., Lond.*, no. 48.

The Effect of Diet and Molybdenum on Liver Copper Storage in Stalled Sheep. By GWYNETH LEWIS and RUTH ALLCROFT, *Ministry of Agriculture, Veterinary Laboratory, Weybridge, Surrey*

It has been shown (Dick, 1952, 1953) that the nature of the diet can modify the effect of molybdenum on liver copper storage in the ewe and also affect the level of molybdenum in the blood.

Previous unpublished work at Weybridge indicated that the nature of the diet affected liver copper storage in both cattle and sheep. One of various trials showed that lambs from sheep fed kale during pregnancy had low liver copper reserves. In the present experiments the effects of diets of cocksfoot straw, of kale and a mixture of these two on the liver copper content of Swaledale ewes during pregnancy and of their lambs were studied.

Each diet supplied an average of 5 mg copper/day. At this rate of copper intake each group showed a decrease of at least 50% in liver copper values after 5 months, final liver copper levels ranging from 12 to 5.4 p.p.m./D.M. No ataxic lambs were born. Group mean copper levels for lambs killed within 4 days of birth were 70 p.p.m./D.M. for the cocksfoot group, 12.8 p.p.m. for cocksfoot plus kale and 6.4 p.p.m./D.M. for the kale group.

In other groups the diets were adjusted by oral administration of copper sulphate to give a daily intake of 10 mg copper and each ewe received 300 mg molybdenum/week in three doses of 100 mg. This intake of molybdenum did not prevent rise in liver copper on any of these diets but although the mean blood-molybdenum level for the ewes fed cocksfoot straw was approximately four times that for the sheep receiving kale there was no difference in liver copper storage. Lambs (killed as above) from cocksfoot, cocksfoot and kale and kale fed groups had mean liver-copper values of 74.5, 12.5 and 7.3 p.p.m./D.M. respectively.

These results indicate (1) that liver copper storage in the ewe and foetus is controlled by the nature of the diet rather than by the total copper and/or molybdenum intake. (2) That kale fed to ewes during pregnancy has an inhibiting effect on foetal liver copper storage. (3) That the occurrence of ataxia in lambs is not solely dependent on depletion of liver copper in ewes and lambs.

REFERENCES

- Dick, A. T. (1952). *Aust. vet. J.* **28**, 30.
Dick, A. T. (1953). *Aust. vet. J.* **29**, 18.

The Effect of Aureomycin and Penicillin on the Growth of Calves. By S. K. KON, J. OLIVER, J. W. G. PORTER and B. RIDLER, *National Institute for Research in Dairying, University of Reading*

The effect of antibiotics on the growth of calves has been studied in the United States (*cf.* Braude, Kon & Porter, 1953), but so far no observations have been reported in this country. We have carried out trials with procaine penicillin (80 mg penicillin/calf/day) and with aureomycin (6 g Auofac 2A, Lederle Laboratories Inc., supplying 80 mg aureomycin/calf/day) added to the milk feed of young calves reared under different practical conditions of management on the two farms of this Institute. It was impossible to compare the two antibiotics on the same farm at the same time. Accordingly, on each farm alternate calves were allotted to antibiotic treatment or served as controls; the calves were weighed weekly and each experiment lasted for 12 weeks.

At Church Farm a group of twelve calves (initial mean weight 82 lb.) born between September and December received penicillin and gained 1.25 lb./calf/day, while the control group (initial mean weight 83 lb.) gained 1.04 lb./calf/day.

At Arborfield Hall Farm a group of ten calves (initial mean weight 75 lb.) born during November and December received aureomycin and gained 1.63 lb./calf/day, while the control group (initial mean weight 88 lb.) gained 1.16 lb./calf/day. A further group of ten calves (initial mean weight 74 lb.) born between March and May received penicillin and gained 1.40 lb./calf/day, while the control group (initial mean weight 86 lb.) gained 1.31 lb./calf/day.

The difference in the growth rate of the calves on the aureomycin treatment was highly significant ($P = 0.01$), but the differences on the penicillin treatments were not significant.

Besides these increases in the growth of the treated calves there was a marked reduction in the incidence of scouring.

These results show that aureomycin and possibly penicillin improved the growth rate of calves. The effect was greatest with the calves born during the late autumn period and may, therefore, be related to the seasonal incidence of 'infection' commonly experienced in calf rearing. The use of an antibiotic supplement may thus be of value in rearing calves under practical conditions.

We acknowledge with thanks the gift of procaine penicillin from Glaxo Laboratories Ltd. and of Auofac 2A from Lederle Laboratories Inc.

REFERENCE

- Braude, R., Kon, S. K. & Porter, J. W. G. (1953). *Nutr. Abstr. Rev.* (In the Press.)

The Occurrence of Cyanocobalamin and Related Compounds in Natural Materials. By J. E. FORD, E. S. HOLDSWORTH and J. W. G. PORTER, *National Institute for Research in Dairying, University of Reading*

We have shown previously (Ford, Holdsworth, Kon & Porter, 1953) that the vitamin B₁₂ activity of extracts of gut contents and faeces is contributed by cyanocobalamin itself and by four related compounds, factors A, B and C and pseudovitamin B₁₂.

Results of a survey of a wide variety of natural materials show that, in general, extracts of animal tissues contained preponderantly cyanocobalamin, sometimes accompanied by relatively small amounts of the other compounds, and in particular of factor A. On the other hand, in natural materials subjected to bacterial fermentation, and in various bacterial species, the other compounds were generally present in larger amounts.

Extracts of each of the materials examined were assayed microbiologically with *Ochromonas malhamensis*, known to be specific in its response to cyanocobalamin (Ford, 1953), and with *Bacterium coli*, for which organism all five compounds are active. Portions of each test extract were subjected to ionophoresis (Holdsworth, 1953) and to chromatography. The component activities thereby resolved were made visible by a bioautographic technique (Ford & Holdsworth, 1953).

Material	Microbiological assay* ($\mu\text{g/g}$ fresh material)		Ionophoresis and chromatography showed presence of:
	<i>Ochromonas</i>	<i>Bact. coli</i> tube assay	
Cow liver	1.3	1.3	Cyanocobalamin, factor A (trace)
Cow rumen contents	0.4	1.4	Cyanocobalamin, factors A, B, C, pseudovitamin B ₁₂
Pig liver	0.27	0.25	Cyanocobalamin, factor A (trace)
Pig caecal contents	0.005	0.25	Cyanocobalamin, factors A, B, C, pseudovitamin B ₁₂
Silage	0.016	0.046	Cyanocobalamin, factors A, B, C, pseudovitamin B ₁₂

* Vitamin B₁₂ activity expressed as cyanocobalamin.

Some typical findings are shown in the table. When the microbiological results with the two test organisms were in agreement ionophoresis and chromatography confirmed that the material examined contained mainly cyanocobalamin. When the test organisms gave widely differing results the difference was attributable to the presence of relatively large amounts of the other compounds.

The finding that the tissues of ruminants contain preponderantly cyanocobalamin suggests either that the other factors must be poorly absorbed or that after absorption they are rapidly converted into cyanocobalamin, or preferentially excreted. These possibilities are being investigated in studies with the compounds 'labelled' with ⁶⁰Co.

REFERENCES

- Ford, J. E. (1953). *Brit. J. Nutr.* (In the Press.)
 Ford, J. E. & Holdsworth, E. S. (1953). *Biochem. J.* **53**, xxii.
 Ford, J. E., Holdsworth, E. S., Kon, S. K. & Porter, J. W. G. (1953). *Nature, Lond.*, **171**, 150.
 Holdsworth, E. S. (1953). *Nature, Lond.*, **171**, 148.

The B Vitamin Content of the Rumen of Steers Given Various Diets.

By S. K. KON and J. W. G. PORTER, *National Institute for Research in Dairying, University of Reading*

Thiamine, riboflavin, nicotinic acid, pantothenic acid, vitamin B₆, biotin, folic acid, and vitamin B₁₂ activity were determined microbiologically in representative samples of rumen contents from fistulated yearling steers given various diets. The diets included normal ones, such as grass, hay, hay and a concentrate mixture, silage, hay and mangolds, and semi-synthetic low-vitamin diets based on straw treated with caustic soda or straw pulp, supplemented with salts and vitamins A and D, and with gelatin and urea or casein as a source of nitrogen. The effect of penicillin on vitamin synthesis on certain diets was studied by including 200 mg of procaine penicillin in the daily ration of each calf.

	Hay		Hay and concentrates		Straw (NaOH treated), casein		Hay and concentrates with penicillin	
	Diet	Rumen contents	Diet	Rumen contents	Diet	Rumen contents	Diet	Rumen contents
Thiamine	0.8	2.1	5.0	3.0	0	1.8	5.0	—
Riboflavin	13	11	9.0	13	1.0	12	9.0	20
Nicotinic acid	27	50	32	60	2.1	52	32	63
Pantothenic acid	11	10	19	28	1.2	18	19	—
Vitamin B ₆	2.7	2.8	2.5	2.5	0.25	2.4	2.5	3.0
Biotin	0.14	0.16	0.12	0.22	0.004	0.17	0.12	0.16
Folic acid	0.40	1.7	0.25	2.3	0.08	1.0	0.25	—
Vitamin B ₁₂ activity*	0	5.0	0	6.5	0	8.3	0	5.0

All values expressed as $\mu\text{g/g}$ of dry material.

* Measured with *Bacterium coli* plate assay.

Some typical findings are shown in the table, each value being the mean result of the assay of forty to fifty samples. The most striking feature of the results is the close similarity of the vitamin concentration of the rumen material with all the diets. Comparison of the vitamin concentrations in the rumen contents with those of the diets shows that with the normal diets there was relatively little change, except for the synthesis of vitamin B₁₂ and related compounds, whereas with the low-vitamin diets there was evidence of very substantial synthesis of all the vitamins measured.

Penicillin was without marked effect on the synthesis of those vitamins so far measured.

We acknowledge with thanks the gift of procaine penicillin from Glaxo Laboratories Ltd.

Indoor v. Outdoor Rearing of Piglets. By R. S. BARBER, R. BRAUDE and K. G. MITCHELL, *National Institute for Research in Dairying, University of Reading*

In previous experiments with suckling piglets in which the effects of various dietary supplements were tested (Braude, 1949; Braude & Mitchell, 1952), it was shown that piglets reared outdoors on pasture grew better than those reared indoors.

An experiment involving seventy-four litters and extending over 2 years was carried out to determine some of the reasons for this difference in rate of growth between indoor and outdoor reared piglets. The litters were reared either (a) indoors, (b) outdoors on pasture, (c) outdoors on concrete or (d) indoors with soil in the pen. Litters on treatments (a) and (c) were dosed with iron pyrophosphate against anaemia. From the 2nd week onwards all litters were given unrestricted in a creep a suckling-pig meal mixture. The amount of meal consumed by each litter was determined daily. The piglets were weighed weekly throughout the 8-week experimental suckling period.

The results at weaning indicated that the piglets reared outdoors on pasture consumed more creep meal and grew better than piglets on any of the other three treatments. In addition the mean growth and meal consumption of litters reared indoors with soil was higher than that of litters reared either indoors or outdoors on concrete.

The data are being subjected to a detailed statistical examination which will include the effect of different seasons of the year.

These results suggest that access to pasture rather than environmental conditions such as temperature, fresh air and sunlight, was the primary factor responsible for the increased consumption of creep meal and superior growth of litters on treatment (b). The provision of soil to the piglets reared indoors resulted in a similar, but not so marked, effect to that observed for the piglets reared on pasture. This indicates that both actively growing vegetation and soil are beneficial for the young piglet, a conclusion which is supported by the fact that in the winter season the difference between the piglets reared on pasture and those kept indoors with access to soil was small.

REFERENCES

- Braude, R. (1949). *Brit. J. Nutr.* **3**, 293.
Braude, R. & Mitchell, K. G. (1952). *Brit. J. Nutr.* **6**, 398.