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PROCEEDINGS OF THE NUTRITION SOCIETY

ABSTRACTS OF COMMUNICATIONS

The Three Hundred and Fourteenth Meeting of the Nutrition Society was held in the Faculty of Letters, University of Reading, Whiteknights Park, Reading RG6 2AJ on Friday, 7 April, 1978, *when the following papers were read:*

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Dietary fibre and glucose tolerance importance of viscosity. By T. M. S. WOLEVER[1], D. J. A. JENKINS,[1,2,3] A. R. LEEDS[3], M. A. GASSULL[3], J. B. DILAWARI[3], D. V. GOFF[1], G. L. METZ[3] and K. G. M. M. ALBERTI[4], [1]*University Laboratory of Physiology,* [2]*Regius Professor's Department of Medicine, Oxford;* [3]*Gastroenterology Department, Central Middlesex Hospital, London NW10; and* [4]*University Clinical Pathology and Human Metabolism Unit, Southampton, Hants*

To define the type of dietary fibre (Trowell *et al.* 1976) with the greatest potential use in diabetic therapy, groups of 4–6 volunteers took 50 g glucose tolerance tests (GTT) with and without addition of either guar, pectin, gum tragacanth, methyl cellulose, wheat bran or cholestyramine equivalent to 12 g fibre. Addition of each substance significantly decreased blood glucose concentration at one or more points during the GTT (Table) and in general reduced serum insulin levels. The greatest flattening of the glucose was seen with guar, but this effect was abolished when hydrolysed non-viscous guar was used. The reduction in mean peak rise in blood glucose for each substance correlated positively with viscosity (r 0·926, $P<0·01$) as did delay in mouth-to-caecum transit time (r 0·885, $P<0·02$). Viscous types of dietary fibre are therefore most likely to be therapeutically useful in modifying glucose tolerance.

Mean blood glucose levels (mg/100 ml) after taking 50 g glucose with or without the equivalent of 12 g of fibre of various types

Treatment		No. of subjects	Time (min)						
			0	15	30	45	60	90	120
Guar gum	C	6	87	112	140	141	118	93	74
	T		86	91††	113†	113•	110	98	98††
Gum tragacanth	C	6	84	115	138	136	122	95	80
	T		81	91•••	119••	123	110	97	91
Pectin	C	6	84	115	138	136	122	95	80
	T		84	99•	131	135	121	98	88
Methyl cellulose	C	5	82	114	145	142	132	103	81
	T		85	100††	138	124	112•	94	82
Wheat bran	C	6	88	116	143	144	128	91	85
	T		86	100	128•	128	118	96	81
Cholestyramine	C	4	86	109	155	159	146	102	90
	T		84	90•	133••	144	135	90	81

C, control; T, test (=12 g fibre).
Significance of difference between test and control: •$P<0·05$; ••$P<0·02$; •••$P<0·01$; †$P<0·002$; ††$P<0·001$.

Trowell, H., Southgate, D. A. T., Wolever, T. M. S., Leeds, A. R., Gassul, A. R. & Jenkins, D. J. A. (1976). *Lancet* i, 967.

Rumen proteolysis of fraction I leaf protein, casein and bovine serum albumin. By J. H. A. NUGENT* and J. L. MANGAN, *ARC Institute of Animal Physiology, Babraham, Cambridge CB2 4AT*

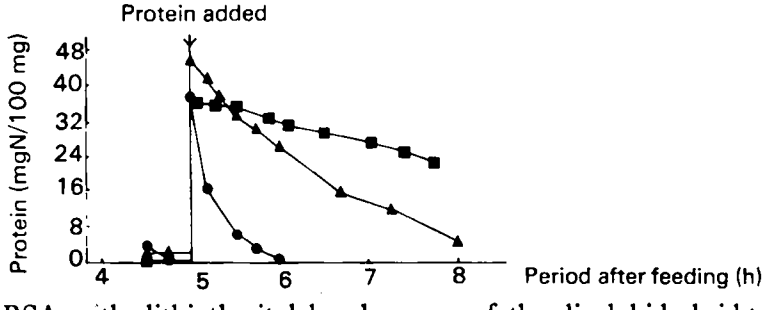
Rates of rumen proteolysis were investigated using a natural substrate, Fraction I protein (*EC* 4.1.1.39) isolated from lucerne leaves, as well as casein and bovine serum albumin (BSA). Previous workers have concluded that the solubility of a protein in rumen fluid governs its rate of rumen hydrolysis (Henderickx & Martin, 1963).

Rates of proteolysis were measured using two diets with both sheep and cattle: (1) hay and oats, (2) fresh lucerne. An in vitro incubation was developed which gave rates of proteolysis close to those obtained in vivo. Experiments were run concurrently in vivo and in vitro to obtain direct comparisons. ^{14}C-labelled Fraction I isolated from lucerne grown in an atmosphere of $^{14}CO_2$ was used in vitro and gave greatly increased sensitivity of assay.

The lucerne diet produced higher but more variable rates of proteolysis than did the hay diet. Rates of proteolysis were reproducible on the hay diet and this allowed values for $V_{max} = 2{\cdot}6 \times 10^{-1}$ mg N/100 ml per min and $K_m = 6$–9 mg N/100 ml to be obtained for Fraction I. Other results confirmed that normally proteolysis is the rate limiting step in rumen protein degradation and that the rumen proteolytic enzymes are mainly bacterial cell wall associated.

Differences were found between proteolysis rates of the three soluble proteins on either diet. Fig. 1 shows that the rates decreased in the order casein >Fraction I >BSA. The use of ^{14}C-labelled Fraction I allowed protein mixtures to be studied and showed that proteins compete for proteolytic enzyme sites.

Fig. 1. Degradation of soluble proteins in vitro using sheep rumen fluid; diet 1000 g hay chaff + 200 g crushed oats once daily, (●), casein; (▲) Fraction I leaf protein; (■) Bovine serum albumin.



Treatment of BSA with dithiothreitol breaks some of the disulphide bridges crosslinking the protein and causes a several fold increase in the rate of rumen proteolysis of this protein. Therefore differences in the rates of rumen hydrolysis of these three proteins are caused by structural differences and not by solubility differences.

We wish to thank Dr F. A. Harrison for rumen fistulating the animals.

Henderickx, H. & Martin, J. (1963). *Compt. Rendus* **31**, 7.

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Biochemical evaluation of the erythrocyte glutathione reductase test for riboflavin deficiency. By A. M. PRENTICE and C. J. BATES, *Dunn Nutritional Laboratory, Milton Road, Cambridge*

The activation coefficient (AC) of erythrocyte glutathione reductase (EGR) is widely used as an index of riboflavin status in human epidemiological studies. To test the assumption that this index is closely related to over-all riboflavin status, correlations between EGR AC values and other biochemical and physiological indices of riboflavin status were investigated in liver, kidney, heart, brain, skin and intestine of rats suffering from acute and chronic riboflavin deficiency.

Acutely deficient rats received a low-riboflavin diet (<0·5 μg/g) from weaning and were studied longitudinally for 7 weeks. Tailcups prevented coprophagy, refection and consequent reversal of deficiency. Chronically deficient animals received diets containing graded, sub-optimal amounts of riboflavin (1·0–2·5 μg/g diet). EGR AC values were measured serially in individual animals until they reached a steady state after 12 weeks.

On sacrificing, the following variables were measured: tissue levels of the flavin coenzymes, FMN and FAD; the activities of the flavin-metabolizing enzymes, flavokinase (*EC* 2.7.1.26), FAD pyrophosphorylase (*EC* 2.7.7.2.) and FMN phosphatase (McCormick & Russell, 1962); the activity of two electron-transport flavoproteins, succinate dehydrogenase (*EC* 1.3.99.1) and NADH dehydrogenase (*EC* 1.6.99.3) and the activity and AC of glutathione reductase (*EC* 1.6.4.2.).

Except for FMN phosphatase activity all the variables responded to riboflavin deficiency in at least some of the tissues investigated. In certain instances age-related or inanition-related changes obscured the specific effects of riboflavin deficiency.

The following showed strong and specific correlations with EGR AC: (a) growth and riboflavin intake in chronic experiments; (b) liver weight : body-weight ratio; (c) flavin coenzyme levels in liver and kidney; (d) flavokinase activity in liver and kidney and FAD pyrophosphorylase activity in liver; (e) basal glutathione reductase activity in all six tissues and GR AC values in liver and skin; (f) succinate dehydrogenase activity in liver, brain and skin; (g) NADH dehydrogenase activity in liver and intestine.

Differences in the rate of change of some of the variables reduced the correlation with the EGR AC in the acute experiments. However, this was avoided in the chronic experiments and correlation coefficients were uniformly higher. It appears that chronic, marginal deficiency more closely matches naturally-occurring human deficiency, and that under these conditions the EGR test accurately reflects over-all riboflavin status.

McCormick, D. B. & Russell, M. (1962). *Comp. Biochem. Physiol.* 5, 113.

Effect of folate deficiency on crypt and villus cells isolated from gut epithelium. By JACQUI BADCOCK and A. M. TOMKINS, *Department of Human Nutrition, London School of Hygiene and Tropical Medicine, London*

Dietary folate deficiency is associated with minimal malabsorption despite atrophy of the gut (Goetsch & Klipstein, 1977) but the nature of the compositional changes in individual cells of the gut in folate deficiency have not been described.

Weanling rats born to mothers taking a folate-free diet developed mucosal folate deficiency after a further 3 weeks on the same diet (Tomkins *et al.* 1976) at which time total mucosal DNA was 44% of control animals. Fractions containing individual epithelial cells from different sites on the villi of the jejunal mucosa were prepared according to a modification of the method of Weiser (1973). In folate deficiency there were lower levels of DNA, RNA, and protein in each fraction but the ratios, RNA:DNA and protein:DNA, were higher at all levels of the villus. Sucrase activity was also much greater in deficient animals at all levels, but especially in villus tip cells (see table).

(Mean values with their standard errors)

Cell Fraction	DNA (mg/cell fraction)		RNA (mg/cell fraction)		Protein (mg/cell fraction)	
	Control	Deficient	Control	Deficient	Control	Deficient
Villus tip cells	0·132±0·005	0·051±0·004	0·498±0·024	0·273±0·015	34·19±4·55	20·05±1·60
Mid villus cells	0·311±0·029	0·096±0·002	0·911±0·036	0·450±0·029	62·52±7·72	36·97±2·82
Crypt cells	0·408±0·020	0·135±0·010	0·843±0·041	0·613±0·039	67·16±6·76	46·38±3·70

Cell Fraction	Protein: DNA		RNA: DNA		Sucrase Activity/mg DNA*	
	Control	Deficient	Control	Deficient	Control	Deficient
Villus tip cells	259±13	395±34	3·77±0·23	5·35±0·32	16·56±0·36	38·39±0·43
Mid villus cells	201± 8	385±19	2.92±0·15	4·68±0·28	4·38±0·10	10·46±0·12
Crypt cells	165± 6	344±16	2.06±0·18	4·54±0·32	2·69±0·09	11·74±0·14

*Units of sucrase activity: μg glucose/μg enzyme protein per h.

This suggests that in dietary folate deficiency the larger size of the individual enterocyte together with the greater specific activity of the absorptive enzyme limits the malabsorption which might be expected due to the gut atrophy.

Goetsch, C. A. & Klipstein, F. A. (1977). *J. Lab. Clin. Med.* **89**, 1002.
Tomkins, A. M., Badcock, J. & James, W. P. T. (1976). *Proc. Nutr. Soc.* **32**, 145.
Weiser, M. (1973). *J. biol. Chem.* **248**, 2536.

Carbohydrases in the small intestine mucosa of sow-reared and 3-week weaned piglets. By D. E. KIDDER and M. J. MANNERS, *Departments of Veterinary Medicine and Animal Husbandry, University of Bristol, Langford House, Bristol BS18 7DU*

Samples of mucosa were taken at 5% intervals along the length of the small intestines of sow-reared piglets 3, 5 and 8 weeks old and 3-week weaned piglets of 5 and 8 weeks old, making 5 groups of 7 piglets each. The levels of lactase (β-galactosidase, *EC* 3.2.1.23), trehalase (*EC* 3.2.1.28), and the maltases, namely sucrase (glucosidosucrase, *EC* 3.2.1.48), isomaltase (*EC* 3.2.1.10), maltase II and maltase III (both α-glucosidases, *EC* 3.2.1.20) were then measured in mucosal homogenates made from these samples.

With a few exceptions the distribution of enzymes along the small intestines resembled that reported for older pigs (Kidder & Manners, 1976, 1978) with low levels at the proximal and distal end and a peak either in the proximal half (trehalase and lactase) or approximately midway (the maltases). Lactase was present at fairly high levels along the proximal 85% of the small intestine in the 3-week-old pigs, falling to very low levels terminally. With increasing age, the distal region with very low lactase formed an increasing part of the length and by 8 weeks, comprised almost one-half, as reported by Manners & Stevens (1972). In several of the 3-week-old pigs, the sucrase was present at relatively high levels in the distal 10–20% of the small intestine, as was found by Manners & Stevens (1972).

Although the lactase levels in the distal half diminished with age, the peak lactase level, which was in the proximal half, showed no noticeable alteration with age. All the other enzymes increased with age from low levels (with trehalase, very low levels) at 3 weeks, to levels at 8 weeks which were a little below those reported for 11-week-old pigs (Kidder & Manners 1976, 1978).

In the 5-week-old piglets, the levels of the maltases were higher in the weaned piglets than in those on the sow, but at 8 weeks no differences were found between the results on the two treatments.

Kidder, D. E. & Manners, M. J. (1976). *Proc. Nutr. Soc.* **35**, 26A.
Kidder, D. E. & Manners, M. J. (1978). *Digestion in the pig.* pp. 98–111. Bristol: Wright-Scientechnica.
Manners, M. J. & Stevens, J. A. (1972). *Br. J. Nutr.* **28**, 113.

How important is dietary linoleate for the young ruminant? By W. M. F. LEAT and CHRISTINE A. NORTHROP, *Biochemistry Department, Agricultural Research Council Institute of Animal Physiology, Babraham, Cambridge*

Very small amounts of linoleic acid (C18:2) pass the placenta of sheep into the developing foetus, and the major source of linoleic acid is found postnatally via milk secretion (Leat, 1970; Leat *et al.* 1978). Dietary fat is important for the growth and well being of the young lamb (Cunningham & Loosli, 1954), but whether essential fatty acids *per se* have any function is unclear. To investigate the importance of dietary linoleic acid during the neonatal period of development lambs have been reared until 9 weeks of age on a liquid diet containing 0·15% of the dietary energy as linoleic acid.

Two sets of twin Clun Forest lambs (3♀ 1♂) were used. Immediately after birth and before suckling one lamb from each pair (1♀ 1♂) were removed from the mother and given 100 ml sheep colostral whey. These lambs were subsequently bottle fed to appetite on a liquid diet consisting of 100 g dried skim milk + 60 g hydrogenated coconut oil suspended in 1000 ml warm water and supplemented with vitamin A (500 μg) vitamin D (5 μg) and vitamin E (50 mg). Iron supplements were given weekly by intramuscular injection. The male lamb increased its weight from 3·4 kg at birth to 16·5 kg at 9 weeks of age, and the female lamb from 3·0 kg at birth to 14·5 kg. The control lambs maintained on the ewe and weaned at 14 weeks increased their weights from 4·3 to 20·0 kg and from 2·9 to 18·0 kg respectively during the same time period. Seven orphan female lambs having a mean birth weight of 3·6 kg and reared on milk and concentrates had a mean body-weight of 15·1 kg at 9 weeks of age. After 8 weeks on the diet low in linoleic acid the plasma fatty acids of the lambs contained low concentrations of C18:2 and a high ratio C20:3/C20:4 compared with the lamb maintained on the ewe (Table).

Plasma fatty acids (% by wt) of lambs given diets low in linoleic acid for 9 weeks

Fatty acid	16:0	16:1	18:0	18:1	18:2	20:3 (ω9)	20:4 (ω6)	$\frac{20:3}{20:4}$
Lamb 1	15·5	10·2	16·3	29·6	3·6	2·5	1·4	1·78
Lamb 2	15·2	12·3	16·6	29·4	4·0	2·5	1·1	2·27
Control lamb	16·4	1·4	15·4	24·8	25·4	0·1	4·9	0·02

It is concluded that the requirement of the lamb for linoleic acid during the first 8 weeks of life is very low (<0·15% of dietary energy) and intake is unlikely to be a limiting factor for growth under normal husbandry conditions.

Cunningham, H. M. & Loosli, J. K. (1954). *J. anim. Sci.* **13**, 265.
Leat, W. M. F. (1970). In *Physiology of Digestion and Metabolism in the Ruminant.* p. 21. [A. T. Phillipson, editor]. Newcastle upon Tyne: Oriel Press.
Leat, W. M. F., Harrison, R. A. & Judge, S. R. (1978). *Proc. Nutr. Soc.* **37**, 5A.

Sodium, potassium and chloride imbalance in broiler diets. By C. J. TALBOT, *Imperial Chemical Industries Ltd, Jealotts Hill Research Station, Bracknell, Berks*

The use of new sources of protein for animal feeds calls for some caution when such materials are included in experimental feeds at levels that are much higher than are appropriate in commercial circumstances. Some such materials are sufficiently different from traditional protein sources in Na, K and Cl content for special consideration to be given to mineral balance. This has been demonstrated using a soya isolate and Pruteen, a fermentation protein of dried microbial cells from ICI.

The Na, K and Cl contents of the soya-bean isolate are 11·9, 2·8 and 2·1 g/kg respectively, and of Pruteen 15·0, 1·4 and 0·3 g/kg respectively.

In view of the finding by Beilharz & McDonald (1960) that an 'excessive dietary NaCl/KCl ratio' induced 'uremia' (avian monocytosis) and mortality, it was important to establish whether such a mineral imbalance effect could be significant if diets containing very large amounts of these novel proteins were fed to broilers.

Two experiments were carried out in which groups of 30 caged male broilers, previously given commercial starter crumbs to 9 d of age, were given cereal-based test diets in which either 250 g Pruteen/kg or 200 g soya-bean isolate/kg were included to replace the soya bean meal and fish meal in the control diet. All diets were formulated to be isonitrogenous, isoenergetic and to contain approximately the same levels of methionine plus cystine, lysine, calcium and phosphorus.

In both experiments it was found that in those groups of birds given diets which contained a high level of Na ($\geqslant$4 g/kg) and relatively low levels of K (3·5 g/kg) and Cl (1·4 g/kg), there was a rapid onset of mortality, within two days after introduction of the test diet, associated with *post mortem* symptoms described as 'visceral gout'.

These effects were observed on diets containing either soya-bean isolate or Pruteen. Mortality could be prevented by either lowering the Na level in the diet to 1·8 g/kg, or by addition of either K to 6 g/kg with Cl at 1·6 g/kg, or Cl to 3·2 g/kg with K at 4 g/kg.

The ratio of K+Cl:Na (on an equivalent weight basis) in such high Na diets should be greater than 1:1. When this ratio is allowed to fall below unity there is severe mortality preceded by accumulation of uric acid in the body ('visceral gout'). The findings of Beilharz & McDonald (1960) are thus supported and this demonstrates the importance of including Na, K and Cl levels as separate nutrients when unusually high levels of novel ingredients are incorporated in poultry diets.

Beilharz, R. G. & McDonald, M. W. (1960). *Aust. vet. J.* **36**, 89.

Foetal accumulation of iron and copper in maternal protein deprivation.

By D. J. NAISMITH and NICOLA M. BINNS, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

It is well established that restriction of dietary protein during pregnancy in the rat retards foetal growth. The effect on the transfer of specific nutrients has, however, received little attention. Rats were given diets containing either 180 g (HP) or 60 g (LP) protein/kg throughout pregnancy, and were killed in groups of six on days 15, 17, 19, 21 and 22 of gestation. Whole foetuses and maternal plasma were analysed for Fe and Cu, and the plasma concentrations of transferrin and ceruloplasmin were measured. The results are given in the table.

	Foetus				Maternal plasma							
	Total Fe (μg)		Total Cu (μg)		Fe (μmol/l)		Cu (μmol/l)		Transferrin (TIBC) (μmol Fe/l)		Cerulo-plasmin (units)	
Day	HP	LP	HP	LP	HP	LP	HP	LP	HP	LP	HP	LP
15	2·5	2·6	0·12	0·10	45·7	26·1	27·7	30·9	91·7	61·2	363	408
17	15·7	16·6	0·61	0·54	18·3	15·0	29·9	27·2	86·3	58·9	366	323
19	37·3	50·5	2·30	1·78	7·9	7·9	29·1	19·5	92·0	49·2	333	200
21	80·3	86·7	6·53	4·75	6·4	5·4	25·8	18·1	101·0	42·1	284	194
22	124·2	125·5	9·15	5·88	5·2	6·9	17·1	13·7	84·7	33·7	177	116

Mean values for individual weights of the foetuses at day 22 were 5·16 g (HP) and 4·16 g (LP).

The rate of foetal accretion of Fe and Cu was rapid after day 17. Feeding the low-protein diet reduced the rate of Cu transfer, and consequently the total Cu content and the concentration of Cu in the foetuses ($P<0.001$). With Fe, the pattern was reversed; the concentration of Fe in the foetuses was raised when the low-protein diet was fed ($P<0.001$). In the well-nourished mothers (HP), plasma Fe, Cu and ceruloplasmin fell markedly during the last week of pregnancy. Transferrin did not change. In comparison with the HP group, rats fed the protein-deficient diet had lower concentrations of Cu, cerulopasmin and transferrin in their plasma ($P<0.001$). These results support the in vitro observation that the placental uptake of Fe is dependent on the concentration of Fe, not of transferrin, in the medium (Laurell & Morgan, 1964). The reduced transfer of Cu resulting from maternal protein deprivation could be due to the low plasma Cu and ceruloplasmin, or to impaired function of the placenta.

Laurell, C–B. & Morgan, E. (1964). *Acta. physiol. scand.* **62**, 271.

An explanation for the elevated efficiency of the genetically obese (obob) **mouse.** By P. L. THURLBY, P. TRAYHURN and W. P. T. JAMES, *MRC Dunn Nutrition Unit, Milton Road, Cambridge*

Pair-feeding studies have shown that the obese (obob) mouse is metabolically more efficient than its lean littermates (Woodward *et al.* 1977). The increased efficiency is not due to a reduced energy cost of growth but to a reduction in the maintenance requirement (Woodward *et al.* 1977). For mice kept at normal temperatures (18–25°) the major components of the maintenance requirement are the basal metabolic rate and the energy cost of maintaining homeothermy (thermoregulatory thermogenesis). In a recent study we found that with adult animals the resting metabolic rate at thermoneutrality was little different for lean and obese mice (Trayhurn & James, 1978). However, at temperatures below thermoneutrality the resting metabolic rate of the obese mice was some 20% less than that of the lean. These results suggest that the lower maintenance requirement of the obese mouse at 'normal' temperatures is due to reduced thermoregulatory thermogenesis.

We have now undertaken two dietary studies to assess the quantitative significance of thermogenesis. First, the energy cost of weight maintenance of young lean and obese mice was determined at thermoneutrality (33°) and at 23°. Animals, aged about 28 d, were fed to weight maintenance for 6 d. At 23° maintenance was achieved with a mean intake of $50{\cdot}3\pm2{\cdot}8$ and $58{\cdot}1\pm2{\cdot}2$ kJ/d ($\pm$ SEM, $P<0{\cdot}02$) for obese and lean animals respectively. However, at 33° the maintenance cost of the obese at $33{\cdot}7\pm0{\cdot}7$ kJ/d was not significantly different ($P>0{\cdot}05$) from the lean at $35{\cdot}7\pm2{\cdot}0$ kJ/d. In the second study 25-d- old obese mice were pair-fed for 10 d to the *ad-lib.* intake of their lean littermates at the same two temperatures. Carcass analysis showed an excess energy gain for the obese of 41 ± 10 kJ at 33° and 116 ± 11 kJ at 23°. Thus two-thirds of the excess gain of the obese mice at 23° was eliminated by conducting the experiment at thermoneutrality.

The results for both the pair-feeding and the weight maintenance study are consistent with the view that the major cause of the high efficiency of the obob mouse is the reduced energy expenditure on thermoregulatory thermogenesis. We suggest that the primary reason for this is that the obob mouse has a lower hypothalamic 'set point' for body temperature (Trayhurn & James, 1978).

Trayhurn, P. & James, W. P. T. (1978). *Pflügers. Arch. Eur. J. Physiol.* **373**, 189.
Woodward, C. J. H., Trayhurn, P. & James, W. P. T. (1977). *Proc. Nutr. Soc.* **36**, 115A.

The time course of the response of rat protein metabolism to trenbolone acetate. By B. G. VERNON and P. J. BUTTERY, *Department of Applied Biochemistry & Nutrition, University of Nottingham School of Agriculture, Sutton Bonington, Loughborough, Leics. LE12 5RD*

The mode of action of the anabolic agents used in farm animals is not fully understood. We have been studying the effect of trenbolone acetate (TBA) (3-oxo-17-β-hydroxy-4, 9, 11-oestratriene acetate) on protein metabolism in the female rat and have previously reported that trenbolone acetate increased the liveweight gain, nitrogen retention and, more interesting, after 21 d, reduced both protein synthesis and degradation in muscle (Vernon & Buttery, 1977; 1978).

Rats were injected daily via the neck skinfold with either a placebo or trenbolone acetate (80 μg/100 g body-weight). Trenbolone acetate injected rats grew significantly faster than the placebo controls during the experimental period. On days 1, 7 and 14 following commencement of trenbolone acetate treatment rats were infused with L-[U-^{14}C]tyrosine and the fractional synthetic rates of various tissues measured (Waterlow & Stephen, 1968). Results are recorded in Table 1. The 'activity of skeletal muscle RNA' was calculated by dividing the fractional synthetic rate by the RNA:protein ratio (Millward *et al.* 1975) (see table).

(Mean values with standard errors of 6 determinations)

(I) Fractional synthetic rates (/d)

	Muscle		Heart		Uterus		Liver	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
(a) Day 1								
TBA	0·103	0·0022	0·144	0·01414	0·44	0·037	0·80	0·045
Control	0·110	0·0026	0·169	0·0122	0·48	0·026	0·82	0·046
(b) Day 7								
TBA	0·073	0·0027**	0·126	0·0093	0·33	0·049	0·67	0·043
Control	0·097	0·0025	0·140	0·010	0·40	0·033	0·73	0·044
(c) Day 14								
TBA	0·074	0·0034*	0·125	0·0081	0·26	0·027**	0·81	0·054
Control	0·091	0·0042	0·130	0·011	0·51	0·019	0·73	0·077

(II) Activity of skeletal muscle RNA (g protein synthesized /g RNA per d)

Day	1	7	14
TBA	18·7±0·76	10·3±0·72**	10·0±0·55**
Control	19·0±1·21	16·0±1·13	14·7±0·61

Statistical significance of differences between the means: *$P<0.05$; **$P<0.001$, n 6.

We acknowledge the support of the SRC, the ARC and the gift of trenbolone acetate from Roussel-Uclaf, France.

Millward, D. J., Garlick, P. J., Stewart, R. J. C., Nnanyelugo, D. O. & Waterlow, J. C. (1975). *Biochem. J.* 150, 235.
Vernon, B. G. & Buttery, P. J. (1976). *Br. J. Nutr.* 36, 575.
Vernon, B. G. & Buttery, P. J. (1978). *Amin. Prod.* (In the Press).
Waterlow, J. C. & Stephen, J. M. L. (1968). *Clin. Sci.* 33, 489.

The effect of dry matter concentration on milk substitute intake by the calf. By J. H. TERNOUTH*, I. J. F. STOBO and J. H. B. ROY, *National Institute for Research in Dairying, Shinfield, Reading RG2 9AT*

Two milk substitute diets, based on either a 'mildly' preheated spray-dried or liquid skim milk, containing 200 g fat/kg dry matter (DM), were reconstituted at five concentrations (see table). Each of the 10 milks was offered to four Friesian male calves, twice daily, from 2 d of age. *Ad lib.* conditions were achieved before 2 weeks of age.

The source of skim milk had no effect on the intake of milk substitute, liveweight gain, food conversion ratio or the incidence of diarrhoea.

Liquid milk intake was negatively related and DM intake was positively related to the DM concentration of the milk so that calves given milks containing 170 and 200 g DM/kg were heaviest at 12 weeks of age. Three calves given diets containing 200 g DM/kg had daily intakes exceeding 80 g DM/$W^{0.75}$ during the third week of life. Intakes/$W^{0.75}$ were higher at 3–4 than at 7–8 or 11–12 weeks of age. Milk concentration had no effect on the food conversion ratio or the incidence of diarrhoea.

	Milk substitute concentration (g DM/kg)						Source of skim milk		
	80	110	140	170	200	Pooled SE	Spray-dried	Liquid	Pooled SE
Live wt (kg)									
at birth	38·9	40·2	39·3	39·2	38·9	1·75	39·2	39·4	1·10
at 12 weeks	122·1	135·3	137·3	146·5	145·7	3·87	136·3	138·3	2·45
Daily liquid milk intake (ml/$W^{0.75}$)									
3–4 weeks	766	586	504	426	382	16·2	542	523	10·3
7–8 weeks	698	532	444	397	338	13·4	487	478	8·5
11–12 weeks	680	536	437	381	355	18·8	474	482	11·9
Daily dry matter intake (g/$W^{0.75}$)									
3–4 weeks	59·7	63·5	70·5	71·6	74·9	2·14	67·7	68·4	1·36
7–8 weeks	54·4	57·7	62·2	66·9	66·2	1·63	61·0	62·0	1·03
11–12 weeks	55·3	57·9	60·8	63·6	69·2	3·05	60·2	61·7	1·98

The results indicate that the calf can drink very large quantities of liquid milk, especially when the DM concentration is low and that abomasal distension does not appear to impose an absolute limit on milk intake even at a concentration of 80 g DM/kg. Rapid passage of hypo-osmolar whey fluids to the small intestine may be modifying the effect of abomasal distension. When the concentration of the milk is high, there is no evidence that the intake of DM is being controlled by a different physiological mechanism than when the concentration is low.

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Mineral absorption from the digestive tract of calves before and after weaning. By R. N. B. KAY, P. THIVEND, E. D. GOODALL and A. C. DALGARNO, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

Certain divalent metals are absorbed from the digestive tract of calves and lambs more readily before weaning than after. It is not clear whether this effect is related to age, or to the change of diet and onset of fermentative digestion in the rumen.

Four calves were cannulated in the terminal ileum when 1–2 weeks old. At weekly intervals thereafter samples of ileal contents and faeces were taken every 2 h during a 24-h period. Polyethylene glycol (PEG) was included in the diet, and the ileal flow and faecal excretion of minerals were estimated from their concentrations relative to PEG in the pooled 24-h samples.

The calves were given a fluid milk substitute (Volac Ltd, Royston, Herts.) by bucket, twice daily, before weaning and a dry semisynthetic diet (Mills *et al.* 1976) subsequently. Both diets contained a balanced mineral content. Two calves were weaned when about 8 weeks old, the other two when about 12 weeks old.

The minerals fell into three groups. Calcium and phosphorus: net absorption occurred only before the ileum, and was greatly reduced after weaning. Magnesium and manganese: net absorption occurred mainly in the large intestine; absorption of Mg before the ileum declined after weaning while that of Mn increased. Copper, iron and zinc: before weaning only Cu was readily absorbed before the ileum though all three elements were absorbed in substantial amounts from the large intestine. After weaning net secretion occurred before the ileum, only partially compensated by absorption from the large intestine, and for Cu and Fe a negative balance was recorded. The changes in absorption occurred abruptly at weaning; no trend related to age was evident.

These results parallel the observation by Grace (1975) that in sheep given diets of fresh herbage net secretion of some trace elements occurs in the stomach and net absorption in the large intestine.

Grace, N. D. (1975). *Br. J. Nutr.* **34**, 73.
Mills, C. F., Dalgarno, A. C. & Wenham, G. (1976). *Br. J. Nutr.* **35**, 309.

A quantitative light microscopical study of muscle from adult rats previously undernourished in early life. By K. S. BEDI, M. MAHON* and J. L. SMART, *Departments of Child Health and Anatomy*, The Medical School, Oxford Road, Manchester M13 9PT*

Growth in weight of male rats undernourished during gestation and the suckling period is permanently stunted, despite adequate nutrition post-weaning (Smart *et al.* 1973). Muscle, which is the major component of body-weight, has been little studied in such animals and only then in terms of weight, length and DNA content.

Rats were undernourished during gestation and the suckling period by restricting maternal food intake to about half that of control mothers fed *ad lib.* All offspring were fed *ad lib.* from weaning at 25 d. At 11 months of age six previously undernourished (PU) and six control (C) males were killed; the extensor digitorum longus (EDL) muscle from the right hind limb of each animal was removed, weighed and frozen in melting Arcton for 3 min. Transverse sections (10 μm thick) of the mid-belly region of each muscle were cut on a Bright cryostat and stained for succinate dehydrogenase in order to differentiate muscle fibre types.

The total number of fibres in a section from each muscle was counted on low magnification photomicrographs. On higher magnification photomontages of tracks extending medio-laterally across each muscle the cross-sectional area of individual fibres was measured by planimetry. These fibres were subsequently classified as red, intermediate or white (R, I or W) according to their staining pattern. PU rats had similar deficits in body-weight and EDL weight (Table). Their lower EDL weight was probably, at least partly, a function both of a lower total number of fibres (though this was not significantly so) and of the smaller cross-sectional area of their W and I fibres. The mean cross-sectional area of the R fibres did not differ between groups; nor were there differences in the proportions of the three types of fibres.

		C		PU		% deficit	*P*
		Mean	SE	Mean	SE		
Body wt (g)		572·5	21·5	432·7	11·8	24	<0·01
EDL wt (mg)		221·7	7·3	173·0	5·7	22	<0·01
Total fibre no.		2730	225	2235	199	18	NS
Fibre cross-sectional area (μm²):	W	4663	286	3746	130	20	<0·02
	I	2811	199	2378	112	15	<0·10
	R	1540	87	1427	69	7	NS

NS, not significant

These observations indicate that undernutrition during the early part of an animal's life may lead to irrecoverable changes in the histological architecture of muscle tissue.

Smart, J. L., Dobbing, J., Adlard, B. P. F., Lynch, A. & Sands, J. (1973). *J. Nutr.* **103**, 1327.

Absorption of magnesium and phosphate in the omasum of the young steer. By R. H. SMITH and B. M. EDRISE, *National Institute for Research in Dairying, Shinfield, Reading RG2 9AT*

The part played by the omasum in magnesium absorption by the ruminant is uncertain (Horn & Smith, 1978; Tomas & Potter, 1976) and little is known of the role of this organ in exchanges of inorganic PO_4.

Steers with rumen and abomasal cannulas and a flexible sleeve sutured at the omasal-abomasal orifice (Edrise *et al.* 1977) were fed twice/d either with 0·42 kg flaked maize (FM); 0·60 kg dried grass (DG) (diet 1) or 0·70 kg FM; 0·34 kg DG (diet 2). Feeds contained polyethylene glycol (PEG) and ^{103}Ruthenium-phenanthroline complex as markers and some were supplemented with 5·9 g Mg (as sulphate). All contained suitable supplements of other minerals. After diet adaptation digesta samples were taken from the reticulum and omasal outflow just before a morning feed and at 2 and 5 h after it. Ratios of ^{103}Ru, Mg and total P (mainly inorganic PO_4) to PEG were determined in omasal outflow samples. This could not be done directly for digesta entering the omasum so reticulum contents were fractionated into solid and liquid phases and relationships between ^{103}Ru and Mg and P contents determined. By assuming that, with the essentially steady state conditions obtaining, ^{103}Ru:PEG ratios were the same in material entering as in that leaving the omasum, ratios of Mg and P to PEG for material entering the omasum could be calculated. Thus proportions of constituents absorbed during passage of digesta across the omasum could be estimated.

Results for Mg were as follows:

Steer no.	Time after morning feed (h)	Diet 1		Diet 2	
		High Mg	Low Mg	High Mg	Low Mg
1	Before feeding	0·17	0·29	0·15	0·28
	2	0·06	0·20	0·17	0·20
	5	0·19	0·24	0·26	0·27
2	Before feeding	0·11	0·14	0·19	0·23
	2	0·07	0·14	0·21	0·21
	5	0·06	0·07	0·10	0·28
	Mean	0·11	0·18	0·18	0·24
	SE	0·023	0·032	0·021	0·015

Apparent absorption of Mg from the omasum occurred consistently to approximately the extent previously observed between mouth and duodenum in steers given FM and hay (Horn & Smith, 1978). Considerable proportions of phosphorus also were apparently absorbed with over-all mean values (±SE; 11 df) of 0·16±0·029 and 0·26±0·015 for diets 1 and 2 respectively.

Edrise, B. M., Smith, R. H. & Buttle, H. L. (1977). *Proc. Nutr. Soc.* **36**, 8A.
Horn, J. P. & Smith, R. H. (1978). *Br. J. Nutr.*
Tomas, F. M. & Potter, B. J. (1976). *Br. J. Nutr.* **36**, 37.

'Chemically available' iron in foods. By SUSAN J. FOX and A. E. BENDER, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

The amounts of iron absorbed from a particular food by different subjects vary over an extensive range. For example, in 14 non-anaemic subjects values for Fe absorption from soya beans ranged between 1·5 and 42·2% (Layrisse *et al.* 1969). Since one specific food is involved the chemical form of the Fe is constant and the variability appears to be entirely due to physiological conditions. Hence the term 'available iron' which is commonly used to mean the proportion absorbed is a misnomer and should instead refer to the chemical form alone.

An attempt was made to measure the chemical availability of Fe in foods *in vitro* so avoiding variability due to physiological conditions. Samples of gastric juice were obtained from volunteers by stomach tube 15–20 min after drinking 500 ml of water. The foods were finely ground, incubated for 1·5 h with 25 volumes of gastric juice, centrifuged, and the total Fe content of the supernatant determined by atomic absorption spectrometry.

Table 1 shows the fractions of the total Fe solubilized by the gastric juice from subject A on ten occasions and on 16 other subjects each on one occasion.

Table 1. *Fractions of iron solubilized by gastric juice*

Group No.	Foods	Subject A (10 separate occasions) Mean	SD	16 subjects Mean	SD
1	Curry powder	0·020	0·007	0·015	0·007
	Egg (hardboiled)	0·0		0·013	0·018
	Oats (cooked)	0·038	0·015	0·028	0·019
	Watercress (raw)	0·023	0·015	0·019	0·011
2	Red wine	0·971	0·037	0·985	0·038
3	Bran (uncooked)	0·237	0·030	0·199	0·051
	Cocoa	0·067	0·019	0·044	0·021
4	Soya-bean flour (cooked)	0·132	0·026	0·167	0·040
	Peas (canned)	0·061	0·041	0·099	0·060
	Lentils (cooked)	0·013	0·009	0·025	0·031
5	Spinach (canned)	0·308	0·013	0·321	0·051
	Almonds	0·20	0·034	0·166	0·062

Almost no Fe is 'chemically available' from foods in group 1. Foods in groups 3 and 4 show a variation between subjects which appears to be pH dependent. Higher gastric juice acidity releases more Fe in group 3 foods and less in group 4 foods. The differences between subjects for group 5 foods are not pH dependent.

Nine foods similar to those used by Layrisse and co-workers show 'chemically available' Fe values in good agreement with the total Fe absorbed by human subjects (Layrisse *et al.* 1969): r 0·82 ($P<0·01$).

Layrisse, M., Cook, J. D., Martinez, C., Roche, M., Kuhn, I. N., Walker, R. B. & Finch, C. A. (1969). *Blood* 33, 430.