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## **PROCEEDINGS OF THE NUTRITION SOCIETY**

### **ABSTRACTS OF COMMUNICATIONS**

*The Three Hundred and Fifty-third Meeting of the Nutrition Society was held at the University of Reading, Whiteknights Park, Reading, on Thursday, 18 December, 1980 when the following papers were read:*

**Source and level of supplemental N for cows early in lactation.** By J. D. OLDHAM, R. J. FULFORD and D. J. NAPPER, *National Institute for Research in Dairying, Shinfield, Reading, Berks RG2 9AT*

Attention has been focussed on the relatively high protein needs of dairy cows early in lactation, and in particular on the potential advantages of giving dietary supplements of proteins which are substantially resistant to degradation within the rumen (Roy *et al.* 1977). There is little clear evidence of the relative benefits of different supplemental N sources in cows. We therefore designed these experiments to compare milk production of cows given isonitrogenous rations containing urea (U), soya-bean meal (S), formaldehyde-treated ('protected') soya-bean meal (PS) or fishmeal (F) as the major source of supplemental N. The N sources were compared at four levels of inclusion to produce rations containing 103, 123, 143 and 163 g crude protein (CP)/kg dry matter (DM).

Sixteen mature Friesian cows were used. N sources were compared within 4×4 Latin Squares, one square at each level of N inclusion. Treatment periods lasted 3 weeks beginning 2 weeks post-partum. Level of feeding was constant at (kg/d) 20 maize silage, 2 hay and 10.5 concentrates. Digestibility of rations containing 123 and 163 g CP/kg DM was measured in a repeat experiment in mid-lactation.

Ration CP (g/kg DM)	Milk yield (kg/d)	FCM (kg/d)	Milk protein (g/d)	N source	Milk yield (kg/d)	FCM (kg/d)	Milk protein (g/d)
103	21.5 <sup>c</sup>	21.9 <sup>b</sup>	553 <sup>c</sup>	U	23.1 <sup>c</sup>	25.0 <sup>b</sup>	627 <sup>c</sup>
123	25.7 <sup>ab</sup>	28.4 <sup>a</sup>	728 <sup>ab</sup>	PS	25.1 <sup>b</sup>	26.4 <sup>ab</sup>	671 <sup>b</sup>
143	24.5 <sup>b</sup>	26.5 <sup>a</sup>	684 <sup>b</sup>	S	25.2 <sup>ab</sup>	27.1 <sup>a</sup>	692 <sup>b</sup>
163	27.8 <sup>a</sup>	29.2 <sup>a</sup>	767 <sup>a</sup>	F	26.1 <sup>a</sup>	27.5 <sup>a</sup>	742 <sup>a</sup>
SE of difference	1.32	1.60	22.8		0.44	0.82	16.5

*a, b, c, values in same column which do not share a common superscript differ significantly ( $P < 0.05$ ).*

Milk yield, fat-corrected milk yield (FCM) and milk protein output all increased significantly ( $P < 0.05$ ) as ration CP content increased. Most of the increase was achieved with the first increment in CP intake. Milk yield, FCM yield and milk protein output were least with urea rations and greatest with fishmeal. Milk protein yield was significantly greater with fishmeal than with the other protein supplements ( $P < 0.05$ ). DM digestibility was lower ( $P < 0.01$ ) with 123 g CP/kg DM than 163 g CP/kg DM but N source had no effect.

These results show a small nutritional advantage in using fishmeal rather than soya as an N supplement for dairy cows. This is likely to be related to the relative resistance of fishmeal to degradation in the rumen. It is not clear why PS, which is designed to be resistant to degradation in the rumen maintained lower production than F.

Roy, J. H. B., Balch, C. C., Miller, E. L., Ørskov, E. R. & Smith, R. H. (1977). In *Protein metabolism and Nutrition*, p. 126. Pudoc: Wageningen.

**The role of the stomach in the digestion of protein and carbohydrate in growing pigs.** By TERESA ZEBROWSKA and HANNA ZEBROWSKA, *Institute of Animal Physiology and Nutrition, Polish Academy of Sciences, 05-1100 Jabłonna, Warsaw, Poland* and A. G. LOW, *Nutrition Department, National Institute for Research in Dairying, Shinfield, Reading, Berks RG2 9AT*

The objectives of this study were to measure the amount of protein and carbohydrate digestion in the stomach of growing pigs and to examine whether any of the digestion products were absorbed there.

Eight pigs of approximately 35 kg live weight were prepared with a small pouch of duodenum into which the pancreatic duct opened. The parts of the duodenum proximal and distal to the pouch were linked by a re-entrant cannula. The pouch contained a T-piece cannula which was connected externally to the entry part of the re-entrant cannula. Thus it was possible to collect digesta before it was hydrolysed by pancreatic enzymes. The pigs received diets based on barley and soya (A) and starch, sucrose and casein (B), daily in equal amounts at 08.00 hours and 20.00 hours. Digesta were collected during three 12 h periods for each diet. The mean ( $\pm$ SD) dry matter, nitrogen and glucose contents of the diets and digesta are shown in the Table.

	Dry matter (g/24 h)	Nitrogen (g/24 h)		Glucose (g/24 h)	
		Total	TCA soluble	Total	Free
<b>Diet A</b>					
Diet	1324.0	38.3	2.1	767.4	—
Duodenum	1365.8	40.9	19.0	679.4	20.0
SD	85.80	1.59	1.71	88.05	14.03
<b>Diet B</b>					
Diet	1350.0	49.6	1.6	817.0	—
Duodenum	1408.0	52.1	12.1	768.4	50.6
SD	58.01	2.70	1.82	60.75	18.40

We conclude that although extensive digestion of protein occurred, there was little or no absorption of N in the stomach. However, there was evidence of digestion and absorption of carbohydrate; we assume that this was because of the action of salivary amylase and hydrochloric acid only, since maltase is not present in the gastric mucosa. In view of the N content of the bile collected from pigs given similar diets (Sambrook, 1978) it appeared that the amount of N secreted by the salivary glands, oesophagus and stomach was about 2 g/24 h.

We thank the Polish Academy of Sciences and the Agricultural Research Council for supporting this study.

Sambrook, I. E. (1978). *Proc. Nutr. Soc.* **37**, 84A.

**The influence of feed intake, protein:energy value and environmental temperature on the efficiency of protein utilization in the pig.** By F. BERSCHAUER\*, W. H. CLOSE and D. B. STEPHENS, *ARC Institute of Animal Physiology, Babraham, Cambridge CB2 4AT*

The utilization of digestible protein for protein retention is dependent on endogenous (e.g. body-weight, sex and breed) and exogenous (e.g. composition of feed, feed intake and environment) factors. The extent to which some of these factors influence protein utilization in the pig was investigated in the present experiments. The design of the experiment was similar to that described by Close & Berschauer (1981). Immediately following the calorimetric investigations the pigs on the low- and medium-feed intakes were catheterized. Blood samples were taken on the second and third post-operative days and blood urea concentration (BU), which is an indicator of protein utilization (Berschauer, 1977), was determined. The temperature was then reduced from 22° to 10° and, following appropriate habituation, the N-balance and BU were again measured. N retention, NR, ( $y$ ; g/kg<sup>0.75</sup> per d) at 22° was related to the intake of digestible N, IDN, ( $x$ ; g/kg<sup>0.75</sup> per d) in a curvilinear manner, while that at 10° was linear. These relationships are presented in the Table which also shows values for NR:IDN and BU.

Protein content of ration (g/kg DM)	Equations for the prediction of N-retention	NR:IDN		BU (mg/100 ml)	
		Mean	SE	Mean	SE
	22°				
150	$y = -0.20 + 0.836x - 0.027x^2$ ( $r 0.99$ )	0.64	±0.02	16.5	±0.8
200	$y = -0.37 + 0.965x - 0.073x^2$ ( $r 0.98$ )	0.60	±0.02	24.8	±1.5
250	$y = -0.21 + 0.725x - 0.040x^2$ ( $r 0.97$ )	0.52	±0.02	30.9	±1.9
	10°				
150	$y = -0.38 + 0.909x$ ( $r 0.90$ )	0.61	±0.05	21.8	±4.2
200	$y = -0.53 + 0.701x$ ( $r 0.94$ )	0.39	±0.05	32.2	±2.8
250	$y = -0.75 + 0.678x$ ( $r 0.90$ )	0.31	±0.05	41.8	±2.0

At each temperature, an increase in the protein content of the diet was associated with a reduction in protein utilization and an increase in BU. From the combined results at 10° and 22° the following relationship between BU ( $x$ ; mg/100 ml) and NR:IDN ( $y$ ) was obtained:

$$y = 0.825 - 0.013x \quad (n 50; r 0.718; P < 0.001).$$

On the medium- and high-protein rations the efficiency of N utilization (NR:IDN) was lower at 10° than at 22° and these differences were reflected in the higher values of BU at 10°.

Berschauer, F. (1977). Thesis, Universität Hohenheim. Verlag Eugen Ulmer, Stuttgart.  
Close, W. H. & Berschauer, F. (1981). *Proc. Nutr. Soc.* **40**, 33A.

\*On leave from Institut für Tierernährung der Universität Hohenheim, Federal Republic of Germany.

**The influence of the protein:energy value of the ration on the efficiency of energy utilization in the pig.** By W. H. CLOSE and F. BERSCHAUER\*,  
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The maintenance energy requirement ( $ME_m$ ) and the efficiency with which energy is utilized for production ( $k_g$ ), protein ( $k_p$ ) and fat ( $k_f$ ) deposition have been investigated in relation to various animal and environmental factors (Fowler *et al.* 1980; Mount, 1980). There is, however, little information to show the extent to which dietary composition may influence these factors. This was investigated in the present experiments.

Three rations of different crude protein (CP; nitrogen  $\times 6.25$ ) concentration (150, 200 and 250 g/kg DM) but of similar metabolizable energy content (16 MJ ME/kg DM) were fed at three levels, 20 (low), 35 (medium) and 50 (high) g/kg body-weight per d. Four Large White entire male pigs were allocated to the nine different treatments at a constant environmental temperature of 22°. From measurements of heat loss and energy and nitrogen balance, it was possible to partition ME intake into heat loss (H), energy retention (ER) and protein (P) and fat (F) deposition. From the linear regression equation,  $ER = a_1 ME + c_1$ , estimates of  $ME_m$  and  $k_g$  were obtained. Similarly, from the multivariate regression equation  $ME = a_2 P + b_2 F + c_2$ , estimates of  $ME_m$ ,  $k_p$  and  $k_f$  were calculated. The results of these equations (intercept and regression coefficients ( $\pm SE$ )) are given in the Table.

CP content of ration (g/kg DM)	Linear		Multivariate		
	$a_1$	$c_1$	$a_2$	$b_2$	$c_2$
150	0.74 (0.04)	-394 (17)	2.37 (1.21)	1.08 (0.27)	475 (24)
200	0.76 (0.02)	-432 (10)	1.82 (0.59)	1.15 (0.17)	525 (14)
250	0.70 (0.04)	-346 (15)	1.52 (0.67)	1.35 (0.24)	503 (22)

Values of  $ME_m$ , calculated from the linear equation, were 532, 568 and 494 kJ/kg<sup>0.75</sup> per d, on the low-, medium- and high-protein rations, respectively. The corresponding values of  $k_g$  were 0.74, 0.76 and 0.70. The intercepts in the multivariate analysis represent estimates of  $ME_m$ , while the reciprocals of the coefficients  $a_2$  and  $b_2$  represent  $k_p$  and  $k_f$ , respectively. The higher the protein content of the ration, the higher was the value of  $k_p$  (0.42, 0.55 and 0.66, respectively) and the lower was the value of  $k_f$  (0.92, 0.86 and 0.74, respectively). These results can be explained on the basis of the contribution made by the carbohydrate, fat and protein fractions of the ration to its total energy content.

Fowler, V. R., Close, W. H., Fuller, M. F. & Whittemore, C. T. (1980). In *Energy Metabolism* [L. E. Mount, editor]. London: Butterworths.

Mount, L. E. (1980). *Energy Metabolism*. London: Butterworths.

\*On leave from Institut für Tierernährung der Universität Hohenheim, Federal Republic of Germany.

**The ingestion of sows' faeces by piglets.** By B. F. SANSOM and P. T. GLEED, *Agricultural Research Council, Institute for Research on Animal Diseases, Compton, Newbury, Berkshire RG16 0NN*

Sow's milk provides approximately one-tenth of a piglet's iron requirements (approximately 8 mg Fe/d), but under natural conditions piglets do not become anaemic because they ingest soil (Venn *et al.* 1947). Under artificial conditions it has been shown that enriching the sow's diet with iron does not increase the iron concentrations in her milk but that nevertheless the piglets are protected to some extent from anaemia. It was suggested that this effect was due to ingestion by the piglets of either the sow's diet or her faeces (Spruill *et al.* 1971). Other experiments have supported this (Veum *et al.* 1965) but no quantitative results have been obtained. The following experiments were designed to measure the amount of faeces ingested by suckling piglets born to sows housed in solid-floored farrowing pens.

Four sows and their litters were used. Two (A and B) were dosed daily from 2 d before farrowing until 10 d after farrowing with 200  $\mu\text{Ci}$   $^{198}\text{Au}$  absorbed onto the concentrate ration; the others (C and D) received the same treatment from 10 d after farrowing until the piglets were 20 d old. The piglets had no access to the sows' feed. The sows' faeces became highly radioactive but their milk remained free of radioactivity. Sawdust (2 kg) was used daily as bedding and the pens were cleaned daily. The total radioactivity in the faeces and bedding was measured in a whole-body counter (Sansom *et al.* 1971). The radioactivity in the piglets was measured daily in the same way after they had been thoroughly washed to remove external contamination. The results are shown in the Table.

Litter (no. of piglets)	Mean radioactivity in piglets (counts/sec ( $\pm$ SEM))	Mean radioactivity in faeces and bedding (counts/sec per g)	Mean retention of faeces and bedding (g)
A (9)	513 (56)	20	26
B (9)	316 (23)	16	20
C (7)	1207 (138)	20	60
D (10)	220 (20)	22	10

At slaughter at 10 d of age (litters A and B) and 20 d of age (litters C and D) it was shown that all the radioactivity was present in the gastrointestinal tract of the piglets.

These results suggest that piglets could be supplied with 8 mg Fe/d by supplementing the sow during lactation with a diet enriched with Fe so that her faeces contained approximately 800 mg Fe/kg (8 mg Fe/10 g faeces). If the Fe were less than 100% available higher concentrations would be necessary.

Sansom, B. F., Taylor, P. J., Wheelock, D. & Vagg, M. J. (1971). In *Mineral Studies with Isotopes in Domestic Animals*, p. 125. Vienna: I.A.E.A.

Spruill, D. G., Hays, V. W. & Cromwell, G. L. (1971). *J. Anim. Sci.* **33**, 376.

Venn, J. A., McCance, R. A. & Widdowson, E. M. (1947). *J. comp. Path.* **57**, 314.

Veum, T. L., Gallo, J. T., Pond, W. G., Van Vleck, L. D. & Loosli, J. K. (1965). *J. Anim. Sci.* **24**, 1169.

**Prevention of piglet anaemia by feeding sows during lactation with diets enriched with iron.** By B. F. SANSOM and P. T. GLEED, *Agricultural Research Council, Institute for Research on Animal Diseases, Compton, Newbury, Berkshire RG16 0NN*

In modern husbandry systems piglets become anaemic within a few days of birth unless they receive prophylactic doses of up to 200 mg iron as Fe dextran injected intramuscularly. In the previous paper it was shown that piglets eat sufficient faeces to make it likely that supplementation of the sow's diet during lactation should help to prevent anaemia in her piglets. This experiment was designed to measure the size of the effect.

Approximately 2 d before farrowing sows were housed in solid-floored farrowing pens and allocated at random to one of two diets. Diet A was the conventional sow diet (containing approximately 200 mg Fe/kg DM) and diet B was the same diet into which had been incorporated 10 kg/tonne  $\text{FeSO}_{47}\text{H}_2\text{O}$  (approximately 2000 mg Fe/kg). Equal numbers of piglets from eight sows receiving diet A were injected intramuscularly a few days after birth with either 100 mg or 200 mg Fe dextran. Piglets from five sows receiving diet B were given no Fe supplement. The piglets were weighed and blood samples were taken from the anterior vena cava for haematological measurements when they were 7, 14 and 21 d old, when they were weaned. There were no significant differences between the mean body-weights of the groups at any stage. The mean haemoglobin concentrations (g/100 ml) are shown in the Table.

Age (d)	Piglets from sows fed diet A		Piglets from sows fed diet B
	100 mg Fe dextran	200 mg Fe dextran	
7	9.6	9.6	8.8
14	10.1	11.1	9.5
21	9.7	11.8	10.5

The piglets of sows fed the diet enriched with 2000 mg Fe/kg maintained adequate and increasing haemoglobin concentrations until weaning at 21 d when their mean haemoglobin concentration lay between those of the groups supplemented with either 100 or 200 mg Fe dextran. These piglets may therefore be presumed to have absorbed at least 5 mg Fe/d from the sow's faeces, which contained approximately 0.67 mg Fe/g fresh matter. Thus, assuming that the availability of the faecal iron was 100%, the piglets ingested at least 5/0.67, i.e. 7.5 g faeces daily. This estimate is in good agreement with the direct measurements of the ingestion of faeces and bedding reported in the previous paper (Sansom & Gleed, 1981).

The sows accepted the Fe-enriched diet readily. Supplementing the sow's diet with Fe is cheap and can protect piglets from anaemia without the work, trauma and other possible hazards associated with Fe dextran injections.

Sansom, B. F. & Gleed, P. T. (1981). *Proc. Nutr. Soc.* **40**, 34A.

**Tyrosine oxidation during pregnancy in normal and protein deficient rats.**

By S. MAYEL-AFSHAR, R. F. GRIMBLE and T. G. TAYLOR, *Department of Nutrition, School of Biochemical and Physiological Sciences, University of Southampton, Southampton SO9 5NH*

Amino acid oxidation is reduced in protein deficiency (Reeds, 1974) and urea cycle enzyme and blood urea measurements suggest that a similar response occurs in pregnancy (Naismith & Fears, 1972). Naismith has suggested two phases for protein metabolism during rat pregnancy. Up to day 14 protein accumulates in muscle in an anabolic phase. Subsequently, it is broken down to provide amino acids for foetal use during a catabolic phase.

Amino acid oxidation during both phases of pregnancy in rats on normal (198 g protein/kg *ad lib.*) and protein deficient (50 g protein/kg *ad lib.*) diets was examined by measuring L-[<sup>14</sup>C]tyrosine oxidation at 8, 12, 17 and 21 d. Non pregnant rats given the low-protein diet were also studied.

Wistar rats (250 g) started their diets 3 d before mating. On the appropriate day, rats, five/group, were put into a gas collection apparatus and received a constant infusion of [<sup>14</sup>C]tyrosine (3.3 µCi/h) via a tail vein. <sup>14</sup>CO<sub>2</sub> was collected in Dreschel bottles of 2M-KOH. <sup>14</sup>CO<sub>2</sub> production was monitored at regular intervals for 6 h by liquid scintillation counting of aliquots. The rate of <sup>14</sup>CO<sub>2</sub> production rose to a plateau after 120 min.

Days pregnant or equivalent . . .	Anabolic phase			Catabolic phase		SEM	F test	
	0	8	12	17	21			
Diet and status	Percentage of dose as <sup>14</sup> CO <sub>2</sub>							
Control pregnant (A)	17.50	16.69	16.32	14.36	10.2	0.64	d <sub>17-21</sub> < 8-12	
Low-protein pregnant (B)		8.90	8.06	7.68	6.23		d <sub>21</sub> < 8	
Low-protein non-pregnant (C)			8.75		8.23			
		SEM	0.65		0.65			
		F test	CB < A		B < CA			

Normal pregnancy resulted in a fall in tyrosine oxidation during the catabolic phase while in protein deficient pregnancy a dramatic fall in oxidation occurred during both phases. Although this reduction was largely due to protein deficiency *per se*, pregnancy enhanced the effect particularly at the end of the catabolic phase.

This study suggests that reduced amino acid oxidation plays a minor part in over-all amino acid economy during the anabolic phase of normal rat pregnancy but is more important during the catabolic phase. Furthermore, reduced amino acid oxidation in protein deficiency is further enhanced by pregnancy, particularly during the catabolic phase.

This study was supported by the Medical Research Council.

Naismith, D. J. & Fears, R. B. (1972). *Proc. Nutr. Soc.* **31**, 8A.

Reeds, P. J. (1974). *Br. J. Nutr.* **31**, 259.



**Dietary restriction and the kinetics of glucose absorption across the rat small intestine.** By E. S. DEBNAM (*Introduced by P. P. SCOTT*), *Department of Physiology, Royal Free Hospital School of Medicine, London NW3 2QG*

The presence of high concentrations of glucose in the rat jejunum during feeding is known to influence directly the jejunal absorption of this sugar (Debnam & Levin, 1976). Because the distal small intestine is not usually exposed to such high and variable concentrations of glucose direct influences are questionable. In an attempt to show whether the presence of intraluminal glucose is able to indirectly modify sugar absorption, the effects of a 72 h fast on proximal and distal glucose absorption were compared. Following pentobarbitone anaesthesia, sections of small intestine 200 mm long either beginning at the Ligament of Treitz or ending at the ileal-caecal junction were cannulated. Gut contents were collected for the determination of glucose concentration. Glucose absorption was measured in vivo and kinetic variables were determined after correction for the phlorrhizin-insensitive component of absorption (Debnam & Levin, 1975).

Following starvation,  $K_t$  was reduced by 32 and 38%, respectively in the proximal and distal regions and  $V_{max}$  was reduced by 44 and 42%, respectively. However, the distal intraluminal glucose concentration was similar in both fed and fasted conditions ( $P < 0.05$ ), suggesting that in this region influences other than a direct action of glucose on the sugar absorption process are operative.

(Mean values with their standard errors for at least six rats/group)

	$K_t$ (mM)		$V_{max}$ ( $\mu\text{mol}/10 \text{ cm per } 15 \text{ min}$ )		Luminal glucose concentration (mM)	
	Mean	SEM	Mean	SEM	Mean	SEM
Proximal intestine						
<i>Ad lib.</i> fed	19.6	0.9	67.8	4.8	104.4	7.8
72 h fasted	13.3	0.8**	38.1	3.7**	2.86	0.5*
Distal intestine						
<i>Ad lib.</i> fed	11.6	0.6	23.0	3.1	1.59	0.6
72 h fasted	7.2	0.4*	13.3	1.1***	1.23	0.4

\* $P < 0.001$ , \*\* $P > 0.001 < 0.005$ , \*\*\* $P < 0.02$ .

Financial support from the MRC and Peter Samuel Royal Free Fund is acknowledged.

Debnam, E. S. & Levin, R. J. (1975). *J. Physiol., Lond.* **246**, 181.

Debnam, E. S. & Levin, R. J. (1976). *Gut* **17**, 92.

**The replacement of sucrose by xylitol in the diet, and its influence on the level of dental caries in the rat.** By T. H. GRENBY, *Guy's Hospital, London SE1 9RT*

Within the last few years xylitol has been put forward as a food ingredient which can replace sucrose and improve dental health, but the question of whether it can actively combat dental caries or is merely less cariogenic than sucrose has not been answered conclusively.

A series of trials with more than 500 laboratory rats was set up to investigate this and to study the toleration of the rats to various levels of xylitol fed continuously or intermittently, in comparison with sucrose, wheat starch and other polyols. Weanlings of a caries-active Osborne-Mendel strain were divided into matched groups and kept on the experimental regimes for the next 8 weeks. Their condition and weight were monitored regularly, and food and water intake were recorded. At the end of the experiments the teeth were examined for dental plaque and caries, and in some cases tissue lipid levels were determined.

Xylitol was tolerated better than the hexitols, sorbitol and mannitol. Toleration problems with xylitol began at a dietary level of 100 g/kg, but could be avoided by allowing the rats time to adapt to gradually increasing levels. The rats tended to deposit less body-fat on the xylitol regimes than on sucrose or starch.

The dental results showed that xylitol was non-cariogenic but gave no indication of any positive caries-inhibitory action. Compared with the high level of caries produced by sucrose, xylitol behaved in the main like wheat starch, neither advancing nor actively retarding the dental caries process.

*Summary of caries scores on xylitol regimens relative to basic 46% sucrose regimen*

(Results are from five expts. Level of caries on basic 46% sucrose regimen set at 100%)

Xylitol in diet (%)	0	2	2	10	20	20
No. of rats/regimen		15	10	16	15	16
Percentage change in mean caries score	—	+58	+54	+3	-55	-61
Comparison with sucrose diet	—	$P < 0.01$	$P < 0.02$	NS	$P < 0.001$	$P < 0.01$

NS, not significant.

**The importance of free T<sub>3</sub> measurements in assessing thyroid status in protein deficient rats.** By M. D. COX, SHREEDEVI S. DALAL and C. R. C. HEARD, *Clinical Nutrition and Metabolism Unit, London School of Hygiene and Tropical Medicine, 4 St. Pancras Way, London NW1 2PE*

Weanling rats fed on very low-protein diets reduce their food intake. If litter mates are fed on a high-protein diet at a similar low level of intake ('energy deficient') they and the 'protein-deficient' animals both show significantly depressed resting metabolic rate (RMR). Paradoxically the protein-deficient rats have high levels of plasma tri-iodothyronine (T<sub>3</sub>) (Millward *et al.* 1979). To complicate the picture further, Tulp *et al.* (1979) reported that their low-protein rats had high T<sub>3</sub> levels with high RMR. Their rats were fed on a diet containing 80 g casein/kg.

In the present experiments groups of weanling rats were fed *ad lib.* on diets containing 50, 100 or 200 g casein/kg in each case supplemented with methionine (Heard *et al.* 1977). A fourth group was given 8 g/d of the 200 g casein/kg diet. Measurements at the end of 14 d on the diets are shown in the Table. The values are given as a percentage of those in the 200 g casein/kg group. Absolute values for the 200 g casein/kg group are given in parenthesis.

Diet	Body-wt (g)	Food intake (g/d per kg <sup>0.56</sup> )	VO <sub>2</sub> (l/d per kg <sup>0.56</sup> )	Free T <sub>3</sub> (pg/ml)	Total T <sub>3</sub> (ng/ml)
	(165)	(58.4)	(22.3)	(5.7)	(1.32)
200 g casein/kg <i>ad lib.</i>	100	100	100	100	100
100 g casein/kg <i>ad lib.</i>	71	90	98	101	158*
50 g casein/kg <i>ad lib.</i>	47	61	76*	75*	170*
200 g casein/kg 8 g/d	55	52	75*	71*	97

\*Significantly different from 200 g casein/kg diet.

Body-weights and food intakes differed significantly between all groups. The results for VO<sub>2</sub> and total T<sub>3</sub> confirmed our earlier observations and those of Tulp *et al.* (1979). The paradox of low RMR and high T<sub>3</sub> in our protein deficient rats (50 g casein/kg) was explained by the fact that it was free T<sub>3</sub> not total T<sub>3</sub> which correlated with RMR. The elevations in total T<sub>3</sub> suggest increased levels of thyroid binding globulin (TBG). Earlier observations with the 'Thyopac-3' kit (Radiochemical Centre) had already revealed greatly increased unsaturated T<sub>3</sub> binding capacity on rats fed on 50 g casein/kg diet. We are investigating the possibility that changes in TBG levels could be mediated through altered androgen/oestrogen status in response to altered nutrition.

Heard, C. R. C., Frangi, S. M., Wright, P. M. & McCartney, P. R. (1977). *Br. J. Nutr.* **37**, 1.

Millward, D. J., Holliday, M. A., Bates, P. C., Dalal, S., Cox, M. & Heard, C. R. C. (1979). *Proc. Nutr. Soc.* **38**, 33A.

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**The effect of zinc supplementation in aged persons on serum albumin and binding capacity.** By W. H. PARRY, *Bristol Polytechnic, Coldharbour Lane* and D. M. FLINT, M. L. WAHLQVIST and P. A. DRYDEN, *Human Nutrition Section, Deakin University, Australia* and D. M. PRINSLEY, *University of Melbourne, Australia* and F. AL-MUKHTAR, *Bristol Polytechnic*

Certain groups within Western Society may be at risk from sub optimal zinc and protein (Sandstead, 1973). We have identified a group (fourteen) of institutionalized aged persons in whom there are combined apparent deficiencies of Zn and protein as evidenced by hypozinaemia ( $<11.0 \mu\text{mol zinc/l}$  plasma and hypoalbuminaemia ( $<35.0 \text{ g/l}$  plasma) (Flint *et al.* 1979).

Zn supplementation for 21 d caused a significant increase in serum albumin concentration from a baseline level of  $33.5 \text{ g/l}$  to a peak of  $36.3 \text{ g/l}$  at 21 d (Wahlqvist *et al.* 1980).

The results of albumin bound Zn suggested that the newly synthesized albumin measured after 21 d of supplementation might have generated more binding sites for zinc thus increasing the capacity of albumin for zinc transport. After zinc supplementation ( $50 \text{ mg Zn/d}$ ) ceased the albumin level showed a significant decrease ( $P < 0.01$ ) after 42 d; the albumin bound zinc, however, did not show a corresponding significant decrease. This suggested that the additional albumin at 21 d did not have the capacity to bind additional zinc.

	Baseline		Zn supplementation						Post supplementation	
			7 d		21 d		42 d		21 d	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Plasma zinc ( $\mu\text{mol/l}$ )	10.7	0.4	11.1	0.5	12.9	0.7**	12.3	0.5**	11.6	0.4
Serum albumin (g/l)	33.5	0.5	34.4	0.4	36.6	0.6**	35.2	0.5**	32.6	0.5
Albumin bound zinc ( $\mu\text{g/ml}$ )	0.012	0.002	0.013	0.003	0.025	0.003***	0.024	0.003***	0.023	0.003***
Folic acid ( $\mu\text{g/l}$ )	4.3	0.6	5.2	0.7	4.6	0.7	4.5	0.6	5.0	0.8

Significance of differences from baseline \* $P < 0.05$ , \*\* $P < 0.01$ , \*\*\* $P < 0.001$ .

As zinc supplementation caused an increase in serum albumin levels this suggested that similar treatment may be worthwhile in this community. This treatment may also have some value for other hypoalbuminaemic groups such as renal patients who are sometimes found to be hypoalbuminaemic and hypozinaemic.

These results will also be discussed in relation to the copper levels in the albumin bound zinc fractions.

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**A double blind dietary challenge of food allergic subjects.** By MARY BROWN and M. J. GIBNEY, *Department of Nutrition, University of Southampton* and M. J. RADCLIFFE and P. R. HUSBANDS, *Hythe Medical Centre, Hythe, Hampshire*

It is now well established that food allergens can play a major role in certain diseases e.g. gluten and coeliac disease. However, the relationship of dietary allergens to such vague symptoms as depression, insomnia, irritability and backache is much less certain. We examined the possibility of such a relationship in a double blind food challenge. Ten patients were chosen in whom the response to an exclusion diet followed by sequential challenge with individual foods had defined an allergen free diet of considerable benefit. The foods commonly implicated were potatoes, cereals, milk, eggs and corn and the most common symptoms were depression, incoherence, irritability, constipation and insomnia.

A basal soup was prepared; tomatoes 500 g, onions 30 g, carrots 20 g, water 550 ml, salt, pepper and mixed herbs to taste. The basal soup was supplemented separately with homogenates of corn, egg, potatoes (25 g/275 ml soup) and whole wheat flour, dried milk and white bread (12.5 g/275 ml soup). These soups, which served as allergen or placebo depending on individual patients were frozen and dispatched double blind in 275 ml duplicate batches. The patients recorded symptoms during the 3 or 7 d intervals between consuming soups, the latter interval if symptoms occurred.

Twenty-four out of twenty-eight allergen soups produced the correct response, i.e. symptoms, while eighteen out of twenty-eight placebo soups produced the correct response, i.e. no symptoms. The results showed a significant heterogeneity of response ( $P < 0.05$ ) emphasizing the value of double blind food challenges. The pooled results were analysed as a set of ten  $2 \times 2$  contingency tables. A highly significant association ( $P < 0.001$ ) between the occurrence of symptoms and the consumption of allergen soups was observed.

**Dietary fibre, sodium, and blood pressure.** By P. M. DODSON, *Lipid Laboratory, St. Bartholomew's Hospital, London* and D. M. HUMPHREYS, O. PATRICK and E. V. COX (Introduced by A. B. McALLAN), *The Royal Berkshire Hospital, Reading*

During the past 30 years, an increasing number of epidemiological studies of primitive populations have shown that blood pressure did not rise with age and essential hypertension did not occur if adult sodium intakes averaged less than 64 mmol/d. Diets of these primitive groups were characterized by low fat intake 10–25% energy, high unrefined carbohydrate intake 50–70% energy, high dietary fibre intake 30–50 g/d, and a  $K^+ : Na^+$  value of greater than unity.

A diet was therefore formulated from English foods matching the primitive diet and provided 6.5–7.5 MJ/d, Na intake of 40–50 mmol/d and K intake 80–90 mmol/d (sodium–potassium, 1:2).

Thirty-two outpatients with essential hypertension were treated with this regime at the Royal Berkshire Hospital for a minimum of 3 months. On the initial and each subsequent visit, blood pressure (lying and standing) and weight were recorded. As the Table shows, the mean diastolic blood pressure fell on the dietary regime. Twelve patients ceased anti-hypertensive therapy as supine diastolic pressure was below 90 mm Hg on many occasions. Thirteen patients reduced therapy but supine diastolic blood pressure remained similar to that on entry to the trial. Seven patients showed no hypotensive response to the regime. A fasting lipid profile was taken on the initial and 3 monthly outpatient attendance from twenty-seven of the hypertensive patients on the dietary regime. There was a significant increase in HDL-cholesterol levels (mean  $\pm$  SE;  $0.91 \pm 0.07$  to  $1.17 \pm 0.07$ ;  $P < 0.01$ ) and a significant decrease in two cardiovascular risk factors; total cholesterol:HDL-cholesterol value ( $7.3 \pm 0.6$  to  $5.0 \pm 0.3$ ;  $P < 0.001$ ) and LDL-cholesterol:HDL-cholesterol value ( $5.03 \pm 0.6$  to  $3.28 \pm 0.27$ ;  $P < 0.01$ ).

We conclude that there is a lowering of cardiovascular risk factors, which may be accompanied by a hypotensive effect, in patients with essential hypertension treated with a dietary regime based on primitive populations.

#### *Diastolic blood pressure (mm Hg) during dietary treatment*

(Values are means with their standard deviations. No. of subjects in parentheses)

	At entry* (32)		3 months (32)		6 months (23)		9 months (13)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Supine	98.3	8.2	86.1	9.0	83.5	8.3	85.0	8.2
Standing	104.7	8.7	90.6	9.0	92.0	8.2	91.5	6.6

\*Mean of all recording during preceding 6–12 months while receiving anti-hypertensive drugs.

**Effects, on serum tri-iodothyronine and reverse tri-iodothyronine concentrations, of an acute intake of sucrose or glucose in man.**

By N. SHARIEF, *Department of Physiology, Guy's Hospital Medical School, London SE1* and P. MARSDEN, *Department of Medicine, Greenwich District Hospital, London SE10*

In six clinically and biochemically euthyroid male subjects changes in serum total tri-iodothyronine ( $T_3$ ) and reverse tri-iodothyronine ( $rT_3$ ) concentrations were measured serially after a single oral dose of 5 g sucrose and glucose/kg ideal body-weight. Control experiments were conducted using water. Serial measurements of oxygen consumption, carbon dioxide production and blood glucose concentration were made. Statistical analysis demonstrated a significant reduction in mean serum  $rT_3$  concentration in subjects taking sucrose compared to those taking water and this decrease was also significantly correlated with time. In subjects taking glucose,  $rT_3$  concentrations changed in the same direction as in those taking sucrose but the values were not significantly different from control nor were they correlated with time. No significant differences in serum  $T_3$  concentrations were observed after the three test meals. There were no significant differences between serum glucose concentrations following ingestion of glucose or sucrose.

The significant decrease in mean serum  $rT_3$  concentration after sucrose was accompanied by a significant increase in metabolic rate (MR) compared with glucose and water, though the ingestion of glucose itself did produce a significant increase in metabolic rate. These results show that iodothyronine metabolism in man is altered following acute ingestion of a dose of carbohydrate. The significant fall after oral sucrose but not after oral glucose in serum  $rT_3$  concentration was independent of serum glucose concentrations which suggests that the difference is not a peripheral glucose load effect.

*Serum tri-iodothyronine concentration (nmol/l)*

Oral dose . . .	Sucrose		Glucose		Control	
	Mean	SE	Mean	SE	Mean	SE
Period after dose (min):						
0	0.37	0.026	0.37	0.040	0.38	0.041
30	0.36	0.024	0.37	0.049	0.38	0.038
60	0.30	0.026	0.35	0.033	0.38	0.044
120	0.29	0.026	0.34	0.034	0.37	0.029
180	0.28	0.028	0.34	0.030	0.40	0.043
Slope	-0.00049	0.000101	-0.00022	0.000170	0.00010	0.000097
Total change in MR (kJ)	158.7	10.37	88.5	6.70	-41.4	9.97

**Additivity of effects of ruminal acetate and either portal propionate or rumen distension on food intake in sheep.** By G. B. ADAMS and J. M. FORBES, *Department of Animal Physiology & Nutrition, University of Leeds LS2 9JT*

It is well established that distension of the rumen or infusion of products of digestion into several sites can depress food intake, but in order to inhibit feeding completely, with any one treatment, supraphysiological levels are required. Feeding activity could be controlled by a series of negative feedback loops, each gaining information from 'receptor sites' signalling the presence of individual metabolites with the total of numerous small negative signals being sufficient to cause satiety (Baile & Forbes, 1974; Martin & Baile, 1971).

Two 3 × 3 factorial experiments were carried out, with a total of nine mature sheep.

The first combined 0, 2 or 4 mmol Na acetate/min infused into the rumen for 3 h, with 0, 0.6 or 1.2 mmol Na propionate/min infused into the hepatic portal vein (Anil & Forbes, 1980).

The second combined the acetate treatments with 0, 1 or 2 l of water pumped into a balloon placed in the rumen. After 3 h the balloon was emptied. In each case the highest level of treatment was supraphysiological and the middle level approximately physiological. A complete pelleted diet, water and salt were available at all times; the Table shows the mean weights (g) eaten during the 3 h treatment period.

Acetate infusion into rumen (mmol/min)	Propionate infusion into hepatic portal vein (mmol/min)			Volume of balloon in rumen (l)		
	0	0.6	1.2	0	1	2
	0	250.4	201.8	143.7	269.7	220.7
2	140.4	100.2	110.2	237.4	140.5	44.5
4	88.9	99.7	96.2	118.8	57.2	90.7

All three factors individually depressed intake, and when given in combination at physiological levels their effects were additive, whereas at higher levels there was considerable interaction. At the termination of the treatments, the sheep always compensated for any depression in feeding caused by the treatments, and the 24 h intakes were unaffected.

These results support the hypothesis of multifactorial satiety in the sheep.

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**Comparative evaluation of apparent digestibility in dogs and cats.** By  
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Little published information is available on food digestibility in dogs and cats. Workers in the area of dog and cat nutrition (NRC, 1974, 1978) have relied mainly on modified Atwater factors to predict energy value. In this system, dogs and cats are assumed to digest and absorb the same food with equal efficiency to man. To examine this assumption, apparent digestibility of commercial canned dog food (CD), canned cat food (CC) and fresh mince (FM) was measured with either six adult dogs (Beagles) or cats. Foods were offered in 14 (dogs) or 21 d (cats) periods in duplicated  $3 \times 3$  Latin square arrangement. Faecal collection excluded the first 7 d. Mean daily food intakes were 1000, 1000, and 410 g/dog for CC, CD and FM. Respective daily food intakes were 227, 271, and 101 g/cat for CC, CD and FM. Mean apparent digestibility coefficients and digestible energy (DE) levels are shown in the Table.

	CD		CC		FM		SED
	Dogs	Cats	Dogs	Cats	Dogs	Cats	
Nitrogen	0.84	0.79	0.87	0.81	0.98	0.96	0.015
Gross energy	0.86	0.62	0.86	0.78	0.97	0.95	0.028
Fat	0.90	0.52	0.91	0.78	0.98	0.96	0.051
DE (MJ/kg DM)	18.5	14.0	19.6	17.8	27.4	26.8	0.84

SED, standard error of the difference between two means.

Apparent digestibility of the commercial diets tested was significantly higher in dogs than cats ( $P < 0.05$  for each nutrient). Though the same tendency was evident for mince, the differences were not significant. The interaction between foods and species was significant ( $P < 0.01$ ) for apparent fat and energy digestibility, but not nitrogen.

The lower digestibility of commercial foods relative to FM was expected. The commercial diets tested were both complementary foods and are based on raw materials surplus to human food requirements which may be of variable food quality. FM is mainly muscle and fat.

In general, the lower the apparent digestibility of the food, the wider the mean difference between dogs and cats. Some of the interspecific digestibility difference could be explained by variations in food intake ( $W^{0.75}$ ), since there was a clear tendency for fat apparent digestibility to increase asymptotically with intake. But whatever the explanation for the species differences, the results do show that equal apparent digestibility cannot be assumed in calculating energy value of dog and cat diets.

National Research Council (1974). *Nutrient Requirements of Dogs*, No. 8. Washington, D.C.: National Academy of Sciences.

National Research Council (1978). *Nutrient Requirements of Cats*, No. 13. Washington, D.C.: National Academy of Sciences.

**The effect of maternal protein source on the induction of immune tolerance to dietary protein in weanling rabbits.** By CHITRA PATHIRANA, N. J. GOULDING, M. J. GIBNEY, P. J. GALLAGHER, JENNIFER PITTS and T. G. TAYLOR, *Departments of Nutrition and Pathology, Faculty of Medicine, University of Southampton, Southampton SO9 3TU*

Macromolecular uptake of dietary protein in the gut is a frequent occurrence which in the short term leads to the production of circulating antibodies to the particular protein. In a previous experiment (Pathirana *et al.* 1979) we failed to record anti-soya antibodies in rabbits fed purified soya-based diets and we speculated that this tolerance might be due to the inadvertent replacement of fish protein by soya protein in the commercial stock diet of the breeding colony. The present experiments set out to test this hypothesis.

Six female New Zealand white rabbits were raised from weaning on a soya-free stock diet. At 3 months of age the stock diets were supplemented with soya protein or cow's milk protein (50 g protein supplement/kg). At 4 months of age the rabbits were mated and the resultant litters weaned onto purified iso-nitrogenous (3 g N/MJ) and iso-energetic diets (17 MJ/kg). Half of each litter received the same protein source as the dam while the other half received the alternative protein. Serum antibodies of weaned rabbits were determined at weekly intervals for 4 weeks after weaning using an enzyme-linked immunosorbent assay (ELISA).

The mean initial values for serum anti-soya protein antibodies (ELISA units/ml $\pm$ SEM) were 146 $\pm$ 76 and 28 $\pm$ 6 for litters from soya and milk fed dams, respectively. By 4 weeks post-weaning the corresponding values for anti-soya antibodies were 265 $\pm$ 78 and 971 $\pm$ 218.

Similarly, mean initial values for anti-milk protein serum antibodies were 50 $\pm$ 3 and 25 $\pm$ 8 units/ml for litters from soya and milk protein fed dams respectively. By 4 weeks post-weaning the corresponding values for anti-milk protein antibodies were 630 $\pm$ 196 and 202 $\pm$ 91. Clearly the maternal dietary protein produced in the offspring a highly significant suppression ( $P<0.001$ ) of immune response to that protein. A second experiment confirmed these observations and demonstrated its persistence for 4 months after weaning.

Pathirana, C., Gibney, M. J., Gallagher, P. J. & Taylor, T. G. (1979). *Proc. Nutr. Soc.* **38**, 26A.

**Excretion of iron in the bile of calves infected with *Fasciola hepatica*.** By H. W. SYMONDS, C. B. MALLINSON, DENISE L. MATHER and D. L. HUGHES, ARC Institute for Research on Animal Diseases, Compton, Newbury, Berkshire RG16 0NN

Six 3-month-old Friesian or Angus bull calves had their duodenum modified surgically to enable the collection of bile and the measurement of its flow rate. Four weeks later four of the calves were each given an oral dose of 1000 metacercariae of *Fasciola hepatica* and two calves were kept as uninfected controls. The rate of flow of bile (ml/min) was determined weekly for up to 28 weeks by measuring the volumes excreted during five consecutive 30 min periods between 09.00 and 11.30 hours. Samples of bile were taken for analysis of total iron content. The excretion of Fe in bile increased after 10 weeks post-infection reaching thirteen to thirty-two times the pre-infection excretion rate before declining as the calves began to lose their fluke burden (see Table).

*Iron excreted in bile ( $\mu\text{g}/\text{min}$ )*

Animal no.	Pre-infection	Weeks post-infection					
		1	5	10	15	17	20
Infected	Mean						
1	1.04	1.28	1.66	2.42	9.00	13.80	12.69
2	1.02	1.20	1.48	3.30	12.28	21.80	16.93
3	1.31	1.06	1.11	5.98	30.50	31.67	32.11
4	1.37	0.86	1.29	6.11	42.23	43.77	29.58
Control							
5	1.30	1.43	1.17	1.34	1.80	1.70	0.88
6	2.36	1.83	1.24	5.22	1.47	1.43	1.44

There was a threefold increase in bile flow rate in the infected animals during the experimental period. When the excretion of Fe in bile was maximal the daily losses would have been approximately 20, 32, 46 and 63 mg Fe/d for calves nos. 1 to 4, respectively and only 2 mg Fe/d in the uninfected controls. Radioactive  $^{59}\text{Fe}$  studies indicated that the biliary Fe originated predominantly from the erythrocytes, presumably as a result of the flukes feeding.

Fe compounds can enhance bacterial virulence in vivo (Rogers, 1973) and several organisms including *Salmonella* spp. produce Fe chelating compounds to enable survival under conditions of low-Fe availability. Aitken *et al.* (1978) have shown that *Salmonella dublin* persists in the bile of fluke infected but not in healthy cattle after exposure to the organism. Part of the reason for this persistence could be the increased biliary Fe concentration arising from the fluke infection.

Aitken, M., Jones, P. W., Hall, G. A., Hughes, D. L. & Collis, K. A. (1978). *J. comp. Path.* **88**, 75.

Rogers, H. J. (1973). *Infect. Immun.* **7**, 445.

**The maximum capacity of the bovine liver to remove manganese from plasma.** By EVELINE D. HALL, H. W. SYMONDS and C. B. MALLINSON, *ARC Institute for Research on Animal Diseases, Compton, Newbury, Berkshire RG16 0NN*

Dietary manganese may vary over a wide range of concentrations often exceeding 100–200 mg/kg, but the concentration of Mn in the systemic plasma of the cow is maintained at approximately 0.02 µg/ml, and the tissue concentration at 0.5 mg/kg. Almost all absorbed Mn present in portal blood is removed during the first passage of blood through the liver (Gibbons *et al.* 1976). This Mn is then excreted in bile. However, Mn toxicity with elevated systemic Mn concentrations does occur, therefore the liver has a limited ability to remove all the Mn reaching it.

To measure the maximum capacity of the liver to remove Mn, three adult Friesian cows with cannulas in the hepatic, portal and mesenteric veins and in a carotid artery were used (Symonds & Baird, 1973). A MnCl<sub>2</sub> solution was infused into the mesenteric vein, and the infusion rate increased at hourly or 2 hourly intervals until systemic plasma Mn concentrations increased. During each infusion rate blood samples were drawn simultaneously from the hepatic and portal veins and carotid artery, and plasma Mn concentrations determined.

Cows . . .	Infusion rate (mg/min)		Plasma Mn concentration (µg/ml)					
	A & B		Cow A		Cow B		Cow C	
	Cow C		Portal	Hepatic	Portal	Hepatic	Portal	Hepatic
Time (h)								
1	1.7	0.8	0.156	0.036	0.100	0.010	0.055	0.025
2	2.4	1.6	—	—	—	—	0.100	0.050
3	2.4	2.4	0.174	0.072	0.150	0.063	0.160	0.030
4	4.8	3.3	—	—	—	—	0.225	0.055
5	4.8	4.0	0.770	0.545	0.225	0.010	0.360	0.115
6	12.0	4.8	4.370	3.975	2.335	1.740	0.665	0.325

The Table shows plasma Mn concentrations (µg/ml) in the portal and hepatic vein at the end of each infusion rate of Mn into the three cows, A, B and C. The mean ( $\pm$ SE) maximum uptake of Mn by the liver was  $0.78 \pm 0.13$  mg/min per kg liver weight, calculated from portal and hepatic venous, and arterial plasma Mn concentrations, at the time when the Mn concentration in the systemic circulation had increased, the rate of plasma flow through the liver and liver weights at slaughter. Normal liver uptake of Mn calculated from plasma Mn concentrations during a control period was 0.01 mg/min per kg liver weight. Liver damage was indicated as a result of the infusion by increased activity of the enzymes sorbitol dehydrogenase (*EC* 1.1.1.4), glutamate dehydrogenase (*EC* 1.4.1.3) and  $\gamma$ -glutamyl transpeptidase (*EC* 2.3.2.2) in plasma.

Gibbons, R. A., Dixon, S. N., Hallis, K., Russell, A. M., Sansom, B. F. & Symonds, H. W. (1976). *Biochim. Biophys. Acta.* **444**, 1.

Symonds, H. W. & Baird, G. D. (1973). *Res. vet. Sci.* **14**, 267.

**Quantitative effects of defaunation on rumen fermentation and digestion in the sheep.** By J. B. ROWE, A. DAVIES and A. W. J. BROOME (*Introduced by J. D. SUTTON*), *ICI Pharmaceuticals Division, Mereside, Alderley Park, Macclesfield, Cheshire SK10 4TG*

The quantitative aspects of the role of protozoa in ruminant digestive function are not well understood. In this study six mature wethers prepared with permanent cannulas in the rumen and duodenum, were defaunated using 'Manoxal-OT' (BDH Chemicals Ltd, Poole). Three weeks later, the animals were divided into two groups of three, housed separately and one group was re-inoculated with protozoa. Each animal received a ration of 500 g concentrate/d (g/kg; oats 450, sugar 230, maize 140, fishmeal 140, urea, minerals and vitamins 40), and 500 g moderate quality chopped hay/d, fed continuously, and which supplied 9.8 MJ/d, calculated metabolizable energy and 25.0 gN/d.

The rates of irreversible loss (IL) from, and interconversion of C between rumen acetate, propionate, butyrate,  $\text{HCO}_3^-$ , and blood  $\text{HCO}_3^-$  were estimated using isotope dilution techniques (Nolan *et al.* 1976). Isotopes were uniformly labelled with [ $^{14}\text{C}$ ] except in the case of [ $2\text{-}^{14}\text{C}$ ]propionate and [ $1\text{-}^{14}\text{C}$ ]butyrate. The apparent digestibility of the diet was measured and the amount of OM and N entering the small intestine estimated with reference to the inert marker, Ru-phenanthroline. Samples of rumen fluid were examined microscopically to estimate the population densities of bacteria and protozoa. The main aspects of the results are summarized in the Table.

	Faunated	Defaunated	Significance of difference <i>P</i>
Protozoal population density ( $\times 10^5/\text{ml}$ rumen fluid)	4.7	0	—
Bacterial population density ( $\times 10^9/\text{ml}$ rumen fluid)	8.2	28.3	<0.001
IL in the rumen (g C/d) of: acetate	59	61	NS
propionate	59	60	NS
butyrate	32	49	0.01
$\text{HCO}_3^-$	43	45	NS
C transfer between rumen acetate, propionate and butyrate (g C/d)	30	124	0.03
IL in venous blood of $\text{HCO}_3^-$ (g C/d)	138	106	0.03
Apparent OM digestibility (%): forestomachs	34	32	NS
whole tract	72	67	NS
N entering the small intestine (g/d)	19	22	0.01
Duodenal N of microbial origin (%)	62	68	NS
Faecal N (g/d)	3.6	5.3	0.002

NS, not significant.

It was calculated that although there was more N entering the small intestine in defaunated animals, as was suggested by Bird & Leng (1978), this was associated with a decreased amount of energy available to the animal.

Bird, S. H. & Leng, R. A. (1978). *Br. J. Nutr.* **40**, 163.

Nolan, J. V., Norton, B. W. & Leng, R. A. (1976). *Br. J. Nutr.* **35**, 127.

**Whole body protein synthesis in cattle sustained by infusion of volatile fatty acids and casein.** By P. J. REEDS, E. R. ØRSKOV and N. A. MACLEOD, *The Rowett Research Institute, Aberdeen AB2 9SB*

Oldham *et al.* (1980) have reported experiments in which protein synthesis was measured in lactating cows which received a conventional diet. When the diet was supplemented with an abomasal infusion of sodium caseinate they noted an increase in the secretion of milk protein but body protein synthesis (corrected for milk protein) was little altered. It is possible to sustain ruminants completely by infusions of volatile fatty acids (VFA) into the rumen and of protein into the abomasum (Ørskov *et al.* 1979). Under such circumstances the true availability of protein can be controlled reliably.

Two dry cows (575 and 565 kg live weight) which had been sustained in this way for over a year received infusions of VFA (acetate-propionate-butyrate 65:25:10) and casein at rates (52 MJ and 80 g N per d) which maintained them in N equilibrium. After 5 d [ $^{14}\text{C}$ ]leucine was infused, via a jugular vein, at a constant rate (10  $\mu\text{Ci}$  in 2.3 ml/h). After 4 h of infusion, four samples (20 ml) of venous blood were removed at 20 min intervals and the specific radioactivity of free leucine was measured (Lobley *et al.* 1980). Protein synthesis was calculated from the difference between the flux of leucine and the loss of N in urine, assuming that leucine contributed 6.7% of body protein. The rate of casein infusion was then increased by 50% and after 5 d a second measurement of body protein synthesis was made.

Period	Intake		Amino N flux (g/d)	Nitrogen excretion (g/d)	Body protein synthesis (g N/d)
	Gross energy (MJ/d)	Nitrogen (g/d)			
1	64.8	80.5	358	78.9	279
2	70.4	117.6	414	120.6	293

The values for protein synthesis in period 1 (1740 g protein/d) were very similar to those found by Lobley *et al.* (1980) in a dry cow of similar digesta-free body-weight receiving a conventional diet. In contrast to growing pigs, in which an increase in dietary protein improved N retention and markedly stimulated protein synthesis (Reeds *et al.* 1980), in the present experiment a 50% increase in protein input stimulated body protein synthesis by less than 10%. As the retention of N was not increased there also appears to have been an increase in protein breakdown. The results suggest that marked changes in body protein synthesis in response to changes in dietary protein are seen only when accompanied by a change in N retention.

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**Carbohydrate conservation in the obese: a theory to explain the ease of weight gain.** By D. J. NAISMITH, M. D. HOLDSWORTH, J. L. BAILEY and A. P. FLEWITT, *Department of Nutrition, Queen Elizabeth College, London W8 7AH*

It is postulated that obese people tend to store ingested carbohydrate as glycogen in the liver, whereas lean people tend to convert carbohydrate to fat. Since the conversion of glucose to fat dissipates some 20% of the energy that could be obtained from its direct oxidation carbohydrate conservation thus provides a very substantial 'bonus' on the energy ultimately derived from its indirect oxidation.

Evidence to test this hypothesis was sought by measuring the concentrations of the major metabolic fuels in circulation during the longest between-meals period of a normal day. It was reasoned that the more abundant hepatic glycogen reserve of obese subjects would maintain the blood glucose for longer than in lean subjects, and so delay the induction of lipolysis.

Fifteen obese and fifteen lean healthy volunteers, paired with regard to age and sex, consumed a standard breakfast (2.3 MJ; 550 kcal) at 9.30 hours to provide a common nutritional baseline, followed by a standard lunch (4.6 MJ; 1100 kcal) at 13.00 hours. 10 ml venous blood samples were drawn for analysis immediately before lunch, and 1.25, 3, 5 and 7 h thereafter. Throughout the day normal activities were followed (reading, light exercise, watching television); no additional food or drink except water was allowed.

The mean serum insulin concentration was higher in the obese subjects 1.25 h after the start of the meal, but at no other time ( $69.6 \pm 15.2$  compared with  $40.6 \pm 3.3$   $\mu\text{U/l}$ ;  $P < 0.05$ ). No significant differences were found in the blood glucose concentrations although insulin appeared to exert a greater influence on blood glucose in the obese. The mean value for plasma non-esterified fatty acids (NEFA) was higher in the lean subjects immediately before the main meal ( $377 \pm 44$  compared with  $251 \pm 56$   $\mu$  equiv/l;  $P < 0.05$ ). 3 h after the meal, mean concentrations had fallen to a similar low value indicating suppression of lipolysis, but during the following 4 h striking differences emerged; the mean total increase for the obese was  $167 \pm 39$   $\mu$  equiv/l compared with  $345 \pm 74$   $\mu$  equiv/l for the lean ( $P < 0.01$ ). The difference was most marked during the final 2 h of the study when the lean subjects, but not the obese, experienced an acute sensation of hunger. Surprisingly, four of the obese subjects showed a modest fall in plasma NEFA, and the mean rise for the group as a whole was  $42 \pm 44$   $\mu$  equiv/l. In contrast all the lean subjects showed the greatest increase during this period; their mean rise was  $285 \pm 47$   $\mu$  equiv/l ( $P < 0.001$ ).

Our results indicate that, in the obese subjects, the energy needs of the tissues during the long periods that separate the meals are largely met by a fuel other than fatty acids, presumably glucose.