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PROCEEDINGS OF THE NUTRITION SOCIETY

ABSTRACTS OF COMMUNICATIONS

The Four Hundred and Forty-fifth Meeting of the Nutrition Society (One Hundred and Seventy-fifth of the Scottish Group) was held in the Assembly Hall, School of Agriculture, Aberdeen on Tuesday and Wednesday, 29/30 September 1987, when the following papers were read:

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Effect of short-term nutritional supplementation on protein turnover during pulmonary disease in cystic fibrosis. By R. W. SHEPHERD, T. L. HOLT, G. CLEGHORN, L. C. WARD, A. ISLES and P. FRANCIS, *Departments of Child Health and Biochemistry, University of Queensland, Brisbane, Australia*

Impetus for aggressive nutritional support for undernourished children with cystic fibrosis (CF) has come from studies indicating that malnutrition is prevalent, that relative underweight for age, height and sex is a major factor affecting survival, and that the clinical state and course of pulmonary disease can be improved with nutritional rehabilitation.

The effects of short-term nutritional supplements on minimizing cumulative weight loss and adverse effects on body protein turnover during pulmonary exacerbations in CF were studied by randomly assigning patients with an acute episode to receive pulmonary therapy and standard hospital-ward diet (n 10) or adjunctive enteral nutrient supplements (n 12; Criticare HM, Mead & Johnson, at 140% recommended daily energy allowance) for 2–3 weeks. Protein turnover was determined as described previously (Holt *et al.* 1985) and the results are shown in the Table.

			Protein metabolism (g/kg per 10 h)					
	Body-wt (kg)		Synthesis		Catabolism		Deposition	
Treatment	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Unsupplemented								
Initial	30·40	2·90	1·86	0·24	1·58	0·27	0·31	0·18
Final	31·60	2·90	2·10	0·31	2·15	0·34	–0·05	0·15
Change	1·20***	0·30	0·25	0·28	0·57	0·38	–0·33	0·22
Supplemented								
Initial	31·50	2·80	2·10	0·32	2·02	0·37	0·09	0·13
Final	33·00	2·70	3·96	0·50	3·18	0·42	0·78	0·20
Change	1·50***	0·30	1·85***	0·31	1·15**	0·25	0·69***	0·10
Significance of difference between groups: $P<$	NS		0·01		0·05		0·001	

NS, not significant.
Within a group the mean change was statistically significant: $^{**}P<0·01$, $^{***}P<0·001$.

Body-weight increased and pulmonary function (33% increase in FEV_1) improved to a similar extent with treatment in both groups. Furthermore, significant increases in mean protein synthesis and protein deposition were observed in the supplemented group.

These results confirm that pulmonary exacerbations have important adverse effects on body protein metabolism; effects which may, in part, be ameliorated by nutritional support.

This work was supported by NHMRC (Australia) and CF Association (Queensland).

Holt, T. L., Ward, L. C., Francis, P. J., Isles, A., Cooksley, W. G. E. & Shepherd, R. W. (1985). *American Journal of Clinical Nutrition* **41**, 1061–1066.

Effect of oestradiol-17β and Cimaterol alone or combined on corticosteroid status and other endocrine and body characteristics of male castrate lambs. By H. GALBRAITH, J. R. SCAIFE, ELIZABETH M. ALDERSON, P. R. HATENDI, D. WATT and E. C. STEWART, *University of Aberdeen, School of Agriculture, 581 King Street, Aberdeen AB9 1UD*

The mechanism of action of oestradiol-17β and the β-adrenergic agonist Cimaterol (Boehringer Ingelheim Vetmedica GmbH) in altering the partition of dietary nutrients in sheep is inadequately known. Measurements of hormones and metabolites in blood, endocrine glands and hormone-sensitive tissues were made in twenty-four male castrate lambs. The lambs weighed 18 kg on average initially and were slaughtered after 62 or 63 d (Hatendi *et al.* 1988). The treatments were: untreated controls (U); 15 mg oestradiol-17β implanted subcutaneously (Compudose, Elanco Products Ltd) (O); Cimaterol (2 mg/kg dry matter (DM) in the diet) (C); 15 mg oestradiol-17β implant + Cimaterol (2 mg/kg DM in the diet) (OC). The results were evaluated by analysis of variance and the significance of main effects are shown in the Table.

	Treatment groups					Statistical significance: $P<$		
	U	O	C	OC	SED	X	Y	Z
Pituitary gland (mg/kg empty body-weight (EBW))	25·3	30·3	18·7	28·7	2·39	0·001	0·05	—
Thyroid glands (mg/kg EBW)	127	87·0	77·0	78·0	16·0	—	0·05	—
Adrenal glands (mg/kg EBW)	45·5	36·1	29·7	40·2	5·96	—	—	0·05
Teat length (mm)	8·52	21·3	7·54	17·6	1·56	0·001	—	—
Blood samples*:								
Urea (mg/l plasma)	540	514	438	382	35·5	—	0·001	—
Free fatty acids (μeq/l plasma)	589	525	446	429	100	—	—	—
Oestradiol-17β (μg/l serum)	6·10	35·0	6·50	41·3	13·1	0·001	—	—
Insulin (μg/l plasma)	0·27	0·41	0·24	0·63	0·16	0·05	—	—
Triiodothyronine (μg/l serum)	0·81	0·65	0·95	0·73	0·11	0·05	—	—
Cortisol (μg/l serum)	19·5	16·5	16·6	16·7	2·6	—	—	—
Corticosteroid receptors: maximum binding capacity (fmol/mg protein)	30·8	28·0	28·3	32·4	2·0	—	—	—

SED, standard error of the difference; X, oestradiol-17β; Y, Cimaterol; Z, interaction between X and Y.
*Samples taken at 09.00–10.00 hours at weekly intervals; only values at day 21 are presented.

Oestradiol-17β treatment on average increased the weight (mg/kg EBW) of the pituitary gland. Cimaterol reduced the concentrations of plasma urea and the weight of the thyroid glands and also adrenal and pituitary glands; effects were effectively reversed by oestradiol-17β. Cimaterol, unlike oestradiol-17β, had no effect on teat length (as an indicator of sex hormone activity) and on concentrations of plasma insulin and triiodothyronine at day 21. Neither oestradiol-17β nor Cimaterol significantly altered total cortisol concentrations nor the maximum binding capacity of skeletal muscle cytosol (gluteus prepared post mortem) for ^{3}H-dexamethasone. These results do not suggest a mediating role for the corticosteroid variables studied in the action of oestradiol-17β and Cimaterol.

Hatendi, P. R., Galbraith, H. & Scaife, J. R. (1988). *Proceedings of the Nutrition Society* **47**, 108A.

Effect of Cimaterol and oestradiol-17β alone or combined on growth and body composition of male castrate lambs. By P. R. HATENDI, H. GALBRAITH and J. R. SCAIFE, *University of Aberdeen, School of Agriculture, 581 King Street, Aberdeen AB9 1UD*

Certain oestrogens such as oestradiol-17β and β-adrenergic agonists such as Cimaterol (Boehringer Ingelheim Vetmedica GmbH), are known to alter the deposition of lean and fat tissue in sheep (Singh *et al.* 1985; Beermann *et al.* 1986). The degree of commonality in mechanism of action is not known. We investigated the response of twenty-four Finn × Dorset male castrate lambs of 18 kg initial mean live weight (LW) to the following treatments: untreated controls (U); 15 mg oestradiol-17β implanted subcutaneously (Compudose, Elanco Products Ltd) (O); Cimaterol (2 mg/kg dry matter (DM) in the diet) (C); 15 mg oestradiol-17β implant + Cimaterol (2 mg/kg DM in the diet) (OC). The lambs were offered a good quality diet restricted to 38 g/kg LW for the experimental period of 62 or 63 d before slaughter. The results were evaluated by analysis of variance and the significance of main effects shown in the Table.

	Treatment groups					Statistical significance: $P<$		
	U	O	C	OC	SED	X	Y	Z
LW gain (kg)	13·0	16·2	15·8	17·0	0·56	0·001	0·001	0·05
DM intake (kg)	49·4	53·2	51·0	51·5	1·24	0·05	—	—
Empty body-weight (EBW) (kg)	26·0	28·3	28·9	29·8	0·576	0·01	0·001	—
Carcass:								
Wt (CW) (kg)	13·9	15·7	16·8	16·6	0·52	—	0·001	0·05
Crude protein (kg)	2·22	2·68	2·58	2·86	0·10	0·05	0·001	0·001
Crude protein (g/kg CW)	159	171	175	171	6·3	—	—	—
Lipid (kg)	2·38	2·48	1·76	1·82	0·28	—	0·01	—
Lipid (g/kg CW)	173	157	102	108	16·5	—	0·001	—
Omental fat (g/kg EBW)	0·031	0·022	0·022	0·024	0·003	—	—	0·05
Perirenal and retro-peritoneal fat (g/kg EBW)	0·015	0·012	0·010	0·010	0·002	—	0·05	—

SED, standard error of the difference; X, oestradiol-17β; Y, Cimaterol; Z, interaction between X and Y.

Oestradiol-17β treatment on average resulted in greater LW gain, food intake, EBW, and weight of carcass crude protein (nitrogen × 6·25), but had no overall effect on fat deposition in carcass or depots. Cimaterol treatment increased LW gain, EBW, carcass weight and its weight of crude protein and, unlike oestradiol-17β, reduced the weight and proportion of fat in the carcass and in the perirenal and retro-peritoneal depots. The presence of significant interactions indicate that there was incomplete or no additivity between the effect of Cimaterol and oestradiol-17β for LW gain, CW, carcass crude protein and omental fat.

Beermann, D. H., Hogue, D. E., Fishell, V. K., Dalrymple, R. H. & Ricks, C. A. (1986). *Journal of Animal Science* **62**, 370–380.

Singh, S. B., Galbraith, H. & Scaife, J. R. (1985). *Proceedings of the Nutrition Society* **44**, 93A.

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The effect of oestradiol-17β and Cimaterol given alone or in combination on lipid metabolism and cAMP concentrations in tissues of male castrate lambs. By E. M. ALDERSON, J. R. SCAIFE and H. GALBRAITH, *University of Aberdeen, School of Agriculture, 581 King Street, Aberdeen AB9 1UD*

Oestradiol-17β and β-adrenergic agonists such as Cimaterol alter carcass lean tissue deposition when administered to ruminants (Singh *et al.* 1985; Beermann *et al.* 1986). However, unlike oestradiol-17β which has inconsistent effects on carcass lipid deposition, β-adrenergic agonists stimulate lipolysis and reduce carcass lipid concentration (Thornton *et al.* 1985).

In the present study, twenty-four male castrate lambs, initial live weight 18 kg, were allocated to the following treatments: untreated controls (U); 15 mg oestradiol-17β implanted subcutaneously (Compudose, Elanco Products Ltd) (O); Cimaterol (2 mg/kg dry matter (DM) in the diet) (Boehringer Ingelheim Vetmedica GmbH) (C); 15 mg oestradiol-17β implant + Cimaterol (2 mg/kg DM) (OC). Animals were fed on a good quality diet and were slaughtered after 62 or 63 d. Samples of subcutaneous adipose tissue (SCAT) were taken for the measurement of lipoprotein lipase (LPL: *EC* 3.1.1.34) activity and the rates of lipogenesis and lipolysis in the absence or presence of Cimaterol added to the incubation medium (33 and 330 ng/ml). cAMP concentrations were measured in samples of liver, muscle (m. gluteus) and SCAT. The results were evaluated by analysis of variance. The significance of the main effects is shown in the Table. In addition, data for lipogenesis and lipolysis were analysed by split plot ANOVA.

	Cimaterol concentration (ng/ml)	Treatment groups U	C	O	OC	SED	Overall mean	Statistical significance: $P<$ X	Y	Z
Lipogenesis*	0	10·6	13·3	11·0	9·5	3·4	11·2[a]	—	—	—
	33	8·2	9·0	10·6	13·8	2·4	10·5[a]	—	—	—
	330	8·4	7·9	10·3	8·4	2·6	8·6[b]	—	—	—
Lipolysis†	0	330	412	363	295	103	365[a]	—	—	—
(basal)	33	1738	1267	1735	968	376	1478[b]	—	0·05	—
	330	1822	2007	1842	1494	309	1857[c]	—	—	—
LPL‡		12·4	13·6	6·1	7·9	2·4		0·01	—	—
cAMP (nmol/g)										
SCAT		0·155	0·202	0·226	0·205	0·026		—	—	—
Liver		2·24	2·04	2·08	1·85	0·326				
Muscle		1·00	0·74	1·04	0·78	0·131		—	0·05	—

SED, standard error of the difference; X, oestradiol-17β; Y, Cimaterol; Z, interaction between X and Y.

[a,b,c] Overall mean values with different superscript letters were significantly different: lipogenesis ($P<0·05$), lipolysis ($P<0·001$).

*μmol acetate incorporated/g tissue per 2 h.

†μmol glycerol released/g tissue per h.

‡μmol fatty acids released/g tissue per h.

These results indicate that administration of oestradiol-17β and Cimaterol in vivo had no significant effect on lipogenesis or lipolysis. Oestradiol-17β treatment significantly reduced LPL activity, while Cimaterol treatment significantly reduced cAMP concentrations in muscle. Overall mean rates of lipogenesis in vitro were significantly reduced by the addition of 330 ng Cimaterol/ml. Lipolysis was greatly increased by the addition of Cimaterol to the incubation medium; however, the effect of Cimaterol (33 ng/ml) was significantly less in Cimaterol-treated animals than in groups U and O.

Beermann, D. H., Hogue, D. E., Fishell, V. K., Dalrymple, R. H. & Ricks, C. A. (1986). *Journal of Animal Science* **62**, 370–380.

Singh, S. B., Galbraith, H. & Scaife, J. R. (1985). *Proceedings of the Nutrition Society* **44**, 93A.

Thornton, R. F., Tume, R. K., Payne, G., Larsen, T. W., Johnson, G. W. & Hohenhaus, M. A. (1985). *Proceedings of the New Zealand Society of Animal Production* **45**, 97–101.

Effect of litter removal and prolactin on lipolysis in parametrial white fat cells of lactating rats. By M. A. RADCLIFFE and C. COLVILLE, *Department of Physiology, University of Aberdeen, Marischal College, Aberdeen AB9 1AS*

Removal of the litter from lactating rats results in a rapid (within 24 h) increase in capacity for lipid synthesis (Flint *et al.* 1981) and a decrease (within 48 h) in maximal rate of catecholamine-stimulated lipolysis (Vernon *et al.* 1987) in white fat tissue. Each of these changes may be opposed by prolactin (PRL) administration. We have investigated the effect of PRL on changes in lipolysis that occur in lactating dams within 24 h of litter removal (Expt 1), and on lipolysis in virgin rats (Expt 2).

Hooded Lister rats were maintained on CRM diet (Labsure). In Expt 1, dams mated at 10–11 weeks of age reared litters of eight pups until killed at 12–15 d of lactation (group A); or such dams were deprived of litters for 24 h before being killed (groups B–D), in which period some (see Table) received two subcutaneous injections of saline vehicle or 20 IU ovine pituitary PRL (Sigma, Poole, Dorset). In Expt 2, virgin rats (in metoestrus or dioestrus) of similar age underwent similar injection regimens before being killed. Basal lipolysis and isoprenaline-stimulated lipolysis (expressed as E_{max} and EC_{50}, see Table) were estimated by glycerol release from cells incubated at 37° in HEPES-buffered Krebs saline containing bovine albumin (40 g/l), pH 7·4. Cell diameters were measured by microscopy (n 100–120).

	Expt 1								Expt 2			
State . . .	Lactating throughout		Lactating; litter removed 24 h						Virgin			
Group . . .	A		B		C		D		V		P	
Treatment . . .	—		—		Vehicle		PRL		Vehicle		PRL	
n . . .	7		6		7		7		4		4	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Live body-wt (g)	260	2·3	280	3·9	276	5·0	275	4·0	211	2·5	212	4·3
Fat cell diameter (μm)	97	1·3	99	0·6	97	2·0	96	1·3	117	1·7	120	3·1
Lipolysis:												
Basal*	31·2	5·16	14·8	3·43	21·6	1·48	29·0	4·53	6·7	2·45	4·4	1·58
Isoprenaline-stimulated:												
E_{max}*	199·6	27·02	136·4	23·53	139·3	17·06	167·3	23·84	212·4	28·17	200·7	28·85
EC_{50} ($\times 10^{-8}$M)	5·9	1·29	14·8	3·73	12·1	3·44	3·9	1·12	5·8	1·19	5·3	1·27

E_{max}, maximal increment above basal: EC_{50}, concentration required for half-maximal stimulation.
*nmol glycerol liberated/10^4 cells in 90 min.

In Expt 1 there were no inter-group differences in fat cell size. Litter removal resulted in reduced E_{max} accompanied by reduced basal lipolysis and raised EC_{50} (data pooled from groups B and C *v.* group A, $P<0·05$ in each case), while none was significantly different in cells from PRL-treated dams (group D *v.* group A). In Expt 2, lipolysis was unaffected by PRL treatment in virgin rats.

We conclude that several changes in lipolysis may arise within 24 h of litter removal in mid-lactating dams and that these changes may be prevented by PRL at a dose which fails to influence lipolysis in virgin rats.

Flint, D. J., Clegg, R. A. & Vernon, R. G. (1981). *Molecular and Cellular Endocrinology* **22**, 265–275.
Vernon, R. G., Finley, E. & Flint, D. J. (1987). *Biochemical Journal* **242**, 931–934.

The effects of the nutritional composition of lunch on psychomotor performance. By A. RALPH[1], A. P. SMITH[2] and G. MCNEILL[1], [1]*Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB* and [2]*MRC Perceptual and Cognitive Performance Unit, Laboratory of Experimental Psychology, University of Sussex, Brighton BN1 9QG*

There is evidence that eating lunch may adversely affect mental performance (Craig, 1986). This study was designed to test mood and performance before and after lunch in which the total energy value of the meal was tailored to provide one-third of each subject's individual daily energy requirement (Food and Agriculture Organization/World Health Organization/United Nations University, 1985) and in which the protein and carbohydrate varied isoenergetically.

Five men, aged 23–37 years, and six women, aged 19–33 years, assessed mood by an eighteen item visual analogue scale, and were given attention tasks measuring ability to resist distraction and speed of searching a visual field for a central or peripheral target. The tests were performed on a Sinclair Spectrum computer, before and after lunch, on three separate days, each 2 weeks apart. On test days subjects had a standard breakfast at 08.30 hours. The prelunch test was performed at 12.00 hours, lunch was eaten at 13.00 hours and the post-lunch test performed at 14.00 hours. On each occasion the lunch varied in nutrient composition. Lunch A was a high-protein meal with 55% energy as protein, 15% as carbohydrate and 30% as fat. Lunches B and C were high-carbohydrate meals comprising 55% carbohydrate, 15% protein and 30% fat; in lunch B the carbohydrate was 40% starch and 15% sugar, and in lunch C there was 40% sugar and 15% starch.

The results indicate that the nutrient composition of lunch had no short-term effect on mood. Subjects were marginally more susceptible to distraction after the protein meal and after carbohydrate meals there was a slower response to peripheral targets ($P<0{\cdot}05$, ANOVA) (see Table).

Lunch . . .	A High protein		B High starch		C High sugar	
Target . . .	Central	Peripheral	Central	Peripheral	Central	Peripheral
Response time (ms)	462	481	462	494	466	492

The results suggest that protein and carbohydrate affect different aspects of mental performance; whether these effects persist after the immediate post-prandial phase remains uncertain.

Craig, A. (1986). *Nutrition Reviews* **44**, Suppl., 163–171.

Food and Agriculture Organization/World Health Organization/United Nations University (1985). *Technical Report* no. 724. Geneva: World Health Organization.

Measurement of body composition in lean athletic women. By R. J. MAUGHAN[1], P. HAGGARTY[3], B. A. MCGAW[3], D. GVOZDANOVIC[2] and S. GVOZDANOVIC[2], *Departments of [1]Environmental and Occupational Medicine, [2]Bio-Medical Physics and Bio-Engineering, University Medical School, Foresterhill, Aberdeen AB9 2ZD* and *[3]Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

A variety of methods are commonly used to assess body-fat content in humans. All methods involve a number of assumptions, and it is not clear whether these are valid for individuals whose body composition is outwith the normal range. We have used four methods to estimate body-fat content in four highly trained female distance runners: their mean age was 28 (SD 9) years, height 1·63 (SD 0·02) m and body-weight 52·3 (SD 4·0) kg. Body fat content was estimated from skinfold thickness as described by Durnin & Rahaman (1967); body density was measured as described by Siri (1956) with correction for residual volume measured by an oxygen dilution method (Wilmore *et al.* 1980); total body potassium was measured by whole-body counting for ^{40}K (Gvozdanovic, 1981); total body water was measured as the distribution space of $H_2\ ^{18}O$ by determination of isotope decay curves over a 21 d period and extrapolation back to zero time (Coward & Prentice, 1985). Results are shown in the Table.

Table. *Body fat content of four subjects expressed as a percentage of body-weight*

Subject no. . . .	1	2	3	4
Method				
Skinfold	14	19	23	21
Hydrostatic weighing	7	14	18	18
Total body K	7	14	20	12
$H_2\ ^{18}O$ distribution space	14	13	20	17

There are clearly large discrepancies in the results obtained by the different methods, and no consistent pattern is apparent. The skinfold thickness method consistently overestimates the body fat content, but the data on which this prediction is based are not applicable to this subject group. It appears that the assumptions inherent in the other three methods used may also cause estimates of body fat content to be inaccurate in subjects such as these.

Coward, W. A. & Prentice, A. M. (1985). *American Journal of Clinical Nutrition* **41**, 659–661.
Durnin, J. V. G. A. & Rahaman, M. M. (1967). *British Journal of Nutrition* **21**, 681–689.
Gvozdanovic, S. (1981). *Physics in Medicine and Biology* **26**, 184.
Siri, W. (1956). Advances in *Biological and Medical Physics* **4**, 239–280.
Wilmore, J. H., Vodak, P. A., Parr, R. B., Girandola, R. N. & Billing, J. E. (1980). *Medicine and Science in Sports and Exercise* **12**, 216–218.

Duration of tracer infusion affects estimates of whole body protein turnover. By S. MELVILLE, M. A. MCNURLAN, K. C. MCHARDY, A. G. CALDER and P. J. GARLICK, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

The effect of food intake on whole body protein turnover is still far from clear, despite the advent of isotopic labelling techniques which should allow the measurement of acute changes. Papers have been advanced showing, by comparison with the post-absorptive state, no change in protein synthesis with a variety of energy and nitrogen intakes (Motil *et al.* 1981), a moderate increase with feeding (Clugston & Garlick, 1982; Hoffer *et al.* 1985), or a large increase with feeding (Rennie *et al.* 1982).

We have performed a series of experiments on six subjects studied repeatedly over several months with infusions of [1–^{13}C]leucine and indirect calorimetry. Each study was carried out after an overnight fast and food was taken hourly to achieve steady-state feeding. The results are summarized in the Table and are expressed as the ratios of the fed:post-absorptive rates of leucine metabolism.

			Leucine flux		Leucine oxidation		Protein synthesis		Protein breakdown	
Protocol	Period of infusion (h)	*n*	Mean	SD	Mean	SD	Mean	SD	Mean	SD
12 h fed + 12 h fast	24	3	1·46	0·14	2·02	0·62	1·33	0·18	0·60	0·10
12 h fed/12 h fast	4/4	5	1·30	0·15	2·43	0·45	1·05	0·10	0·57	0·12
4 h fast + 4 h fed	8	6	1·11	0·12	1·62	0·34	1·00	0·08	0·34	0·07

A 24 h infusion of [^{13}C]leucine during 12 h feeding followed by 12 h fasting gave an apparently higher rate of protein synthesis and lower rate of protein breakdown in the fed state. These results are similar to those obtained with [^{14}C]leucine (Clugston & Garlick, 1982). When the same feeding schedule was used, but measurements made with separate 4 h infusions at the end of each of the fed and fasted periods, there was no change in protein synthesis. Similarly, when an 8 h infusion was performed on post-absorptive subjects, starting with 4 h fasting followed by 4 h feeding, there was no change in protein synthesis on feeding. Furthermore, we have found detectable ^{13}C label in both breath and plasma 12 h after the end of a 12 h infusion. The discrepancies between the various protocols may be due to recycling of label which should be more pronounced during long (24 h) than short (4–8 h) infusions. There are changes in protein turnover occasioned by food, but these seem to be mediated mainly by changes in breakdown rather than synthesis of protein.

Clugston, G. A. & Garlick, P. J. (1982). *Human Nutrition:Clinical Nutrition* **36C**, 57–70.

Hoffer, L. J., Yang, R. D., Matthews, D. E., Bistrian, B. R., Bier, D. M. & Young, V. R. (1985). *British Journal of Nutrition* **53**, 31–38.

Motil, K. J., Matthews, D. E., Bier, D. M., Burke, J. F., Munro, H. N. & Young, V. R. (1981). *American Journal of Physiology* **240**, E712–E721.

Rennie, M. J., Edwards, R. H. T., Halliday, D., Matthews, D. E., Wolman, S. L. & Millward, D. J. (1982). *Clinical Science* **63**, 519–523.

The effect of fasting and insulin infusion on muscle protein synthesis in immature and adult rats. By A. G. S. BAILLIE, CHARLOTTE A. MALTIN and P. J. GARLICK, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

Most studies on skeletal muscle metabolism in rats are conducted in young, fast-growing animals which give a rapid response to acute changes in experimental conditions. Previous work has shown a drop in the synthesis rate of muscle protein in young rats after short periods of food deprivation and recovery with 1 h of refeeding (Garlick *et al.* 1983), but acute responses have not been investigated in older, non-growing animals.

Two sets of female rats were studied, the first at approximately 2 weeks after weaning, weighing about 100 g, and the second at approximately 1-year-old and weighing about 350 g. These were sub-divided into three experimental groups: fed controls, rats fasted for 12 h, rats fasted for 36–42 h. The fractional synthesis rates (k_s) of the muscles were measured by the flooding dose technique of Garlick *et al.* (1980).

After 12 h fasting, body-weights fell to 93% of control values in the young group and 97% of controls in the adult group, while after 36 h fasting the adult group had fallen to 93%, with the young group falling to 72% of control values. Plasma glucose concentrations also responded less rapidly in adults, falling to 89% and 81% of control values after 12 h and 36 h respectively, compared with the values in young rats which fell to 80% and 69% for the same fasting periods. In the adult rats neither soleus (Sol) nor plantaris (Pl) muscles showed any decrease in k_s after 12 h fasting, and even after 36 h fasting there was only a very small and non-significant decrease in k_s values for Pl. The RNA:protein ratios also remained unchanged over both fasting periods. In contrast, in the young animals, a 12 h fast decreased k_s in Sol and Pl to 83% and 60% of control values respectively, with a further fall to 33% and 19% after 36 h. The RNA:protein ratios in the young rats fell over the fasting period, with the fall after 36 h being highly significant in both muscles ($P<0{\cdot}01$).

These results show a remarkably slow response to fasting in adult rats, as measured by muscle k_s. The adult animals were similarly insensitive to the infusion of insulin. After a 12 h fast, infusion of the hormone for 0·5 h into young or old animals depressed the plasma glucose concentration, but increased the rate of muscle protein synthesis only in the young and not in the adult animals.

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Nutrient utilization patterns in lean and obese subjects. By A. C. BRUCE[1], M. A. MCNURLAN[1], K. C. MCHARDY[2], J. BROOM[2], A. G. CALDER[1], E. MILNE[1], W. P. T. JAMES[1] and P. J. GARLICK[1], [1]*Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB* and [2]*Aberdeen University Medical School, Foresterhill, Aberdeen AB9 2ZD*

To investigate the possibility of a different metabolic response to feeding in obesity, a group of grossly obese women (*n* 6, mean age 32 years, mean body mass index 45·1) were recruited along with a group of normal-weight controls (*n* 6, mean age 32 years, mean body mass index 21·7). All subjects were healthy, non-smoking and euthyroid. After an overnight fast and while at rest under a ventilated hood, all subjects underwent 8 h of continuous indirect calorimetry. During this time an intravenous infusion of ^{13}C-labelled leucine was given to measure the rate of protein oxidation (Garlick *et al.* 1987). Subjects continued fasting for the first 4 h of measurement followed by 4 h of feeding with small hourly meals (% energy: fat 35%, protein 15%, carbohydrate (CHO) 55%).

Initially each meal was designed to provide one-twelfth of the subject's estimated sedentary energy requirements, based on ideal body-weight (IBW) for height. For the obese group this proved to be 2·17 × resting energy expenditure (REE) and for the control group, who had a lower energy expenditure, it was 2·78 × REE (control 1). The control group then repeated the study, following the same protocol except that their energy intake was reduced to 2·17 × REE (control 2). Values for nutrient utilization (kJ/h) (see Table) were calculated as described by Garlick *et al.* (1987).

	Fasting						Fed					
	CHO		Fat		Protein		CHO		Fat		Protein	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Obese	84	33	216	32	36	10	192	31	152	40	45	16
Control 1	65	28	174	30	35	7	194	47	69	25	59	7
Control 2							157	40	87	22	60	8

Although the REE in the controls was lower than in the obese, there was no significant difference in the proportions of nutrients utilized in the fasted state. However, in the fed state, when energy was given in relation to IBW (control 1), the obese utilized a smaller proportion of that energy as protein ($P<0·02$) and CHO ($P=0·06$) and a greater proportion as fat ($P<0·005$). By comparison, when fed the same in relation to their metabolic needs, the pattern in the obese was more similar to that in the controls, but the proportion of fat utilized by the obese was still higher ($P<0·05$) and the protein significantly lower ($P<0·01$) than that in the control group. This may be due to an inherent difference in the hormonal response to a meal in the obese, or an acquired difference due to previous diet or habitual food intake.

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Effects of iron deficiency on blood metallothionein-I concentrations in rats. By AILEEN ROBERTSON, J. N. MORRISON, ANNE M. WOOD and I. BREMNER, *Rowett Research Institute, Bucksburn, Aberdeen AB2 9SB*

Problems are experienced in diagnosing zinc deficiency. The assay of metallothionein (MT) in blood or urine may form the basis of a specific test for this condition (Bremner & Morrison, 1987). Information is needed on the effects of other nutritional factors on MT concentrations. MT present in blood is associated mainly with cell fractions rich in reticulocytes and concentrations are increased in rats rendered anaemic by injection of phenylhydrazine (Morrison *et al.* 1987). It was therefore of interest to establish the effects of iron deficiency, which results in increased reticulocyte production, on MT levels in blood cells of rats.

Groups of five male Hooded Lister rats (Fe D) aged 4 weeks were fed on a semi-purified diet containing 5 mg Fe/kg diet. Groups of control rats were fed on the same diet with 50 mg Fe/kg diet *ad lib.* (AL) or were pair-fed (PF) against the Fe D rats. MT-I was measured in plasma and lysed blood cells by radioimmunoassay (see Table). Mean haemoglobin concentrations in the Fe D rats were 80 (SE 4), 62 (SE 3), 64 (SE 5) g/l at 2, 4 and 6 weeks, compared with about 140 g/l in control rats. Plasma MT-I concentrations were slightly reduced in the Fe D rats but were not consistently affected by the restricted food intake in the PF rats. Concentrations of MT-I in blood cells were increased greatly in the Fe D rats whereas they were less in the PF rats than in the AL group.

Table. *MT-I concentrations in plasma and blood cells*

(Mean values with their standard errors for five rats per group)

	Blood cells (ng/ml blood)						Plasma (ng/ml plasma)					
Period of experiment	Fe D		PF		AL		Fe D		PF		AL	
(weeks)	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
2	120	17	34	6	50	4	6·6	1·5	6·8	1·1	6·1	0·7
4	161	17	16	2	50	5	3·8	0·6	4·8	0·9	5·9	0·3
6	111	12	5	1	15	1	2·9	0·4	6·4	2·1	4·2	0·3

The increase in blood cell MT-I in the Fe D rat was not accompanied by similar changes in liver (not shown), indicating that Fe D did not induce general MT synthesis. The increase in blood was probably associated with the increased production of reticulocytes.

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Dietary intake of medical students at the Chinese University of Hong Kong. By N. C. CHAN, H. K. LAW, Y. C. NGAR and K. TADESSE, *Department of Physiology, The Chinese University of Hong Kong, Shatin, N.T., Hong Kong*

In order to evaluate the nutritional intake of our students, we conducted a dietary survey of fifty-one (forty-six male, five female) volunteer Chinese medical students. The survey was conducted during the summer vacation when the students were relatively more active and eating mainly at home. Two standard methods of dietary survey were employed, namely a 24 h dietary recall and a 7 d written dietary record. After estimation of the total quantity of different items of food consumed, nutrient intake was calculated using computer-based data from two food tables (Leung *et al.* 1972; Paul & Southgate, 1978). The age of the students ranged from 21 to 25 years and their mean body-weight was 60 kg. Some of the dietary results are summarized in the Table.

Table. *Daily nutrient intake of Hong Kong medical students*

		Energy (MJ/d)		Carbohydrate (g/d)		Fat (g/d)		Protein (g/d)		Fibre (g/d)	
	n	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
24 h recall	51	14·7	0·7	550·6	21·5	92·0	6·8	109·6	5·0	21·5	1·3
7 d record	30	12·8	0·5	473·4	18·5	79·6	3·9	102·8	4·3	18·1	1·0

The 24-h recall method appears to have overestimated the intake but there was no significant difference between the two methods in the assessment of the proportional contribution of each nutrient to the daily energy consumption. The major source of energy was carbohydrate, contributing over 60% towards the daily energy intake. In contrast the contribution by fat (24%) was relatively low. This may be related to local cooking and eating habits, which are low in dairy products. However, inherent methodological errors in estimating fat intake cannot be ruled out. The contribution of alcohol to the daily energy intake was negligible. The intake of protein was relatively high while that of dietary fibre was on the low side. This may reflect the current eating pattern of the present generation. Overall, the nutritional intake of the students adequately meets the expected energy and protein requirements for their age, weight and occupation (Food and Agriculture Organization/World Health Organization/United Nations University, 1985).

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Estimation of energy expenditure associated with Na^+,K^+-ATPase activity in ovine liver.

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Milligan & McBride (1985) estimated the energy cost of Na^+,K^+-ATPase activity in ovine tissues to be between 30 and 50% of total cellular energy production. Their measurements were made by determining the ouabain-induced reduction in oxygen consumption by tissue preparations incubated in simple buffers providing only one energy substrate. Cells incubated in such buffers may, however, be stressed as indicated by their reduced half-life (Dickson & Pogson, 1977), and Na^+,K^+-ATPase activity is also influenced by insulin and other hormones (Moore, 1983).

To test the influence of incubation medium, lamb hepatocytes were incubated in a more complex buffer containing both glucose and amino acids as energy substrates, with or without insulin (1·5 ng/ml). Cellular O_2 consumption was measured for 10 min in the absence and then in the presence of ouabain (10^{-4} M). Results together with those from McBride & Milligan (1985) for hepatocytes prepared from lambs of the same age and breed as used in the present study but incubated in a simple buffer are given in the Table.

Incubation* medium	Hepatocyte viability (%)		Total O_2 consumption		Na^+,K^+-ATPase dependent respiration†		Na^+,K^+-ATPase independent respiration†		% inhibition by ouabain		
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	*n*
Minimal essential medium	86	3	3·36	0·30	0·32	0·08	3·04	0·26	10·7	1·4	8
Minimal essential medium + insulin	86	3	3·73	0·32	0·81	0·09	2·92	0·24	21·9	1·5	8
Krebs Ringer + glucose‡	93	2	5·62	0·28	2·67	0·21	2·95	0·31	47·8	3·8	5

*All media contained fatty acid-poor bovine serum albumin (20 g/l).
†nmol O_2/mg dry wt per min.
‡Results of McBride & Milligan (1985).

Total O_2 consumption was higher ($P<0·05$) on addition of insulin to the incubation medium. This increase was due entirely to a higher energy cost of Na^+,K^+-ATPase activity ($P<0·01$). O_2 consumption was much higher by hepatocytes incubated in a single energy substrate medium (McBride & Milligan, 1985). The composition of the incubation medium did not alter Na^+,K^+-independent respiration, but the energy cost of Na^+,K^+-ATPase activity varied markedly.

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Factors affecting fluid and carbohydrate replenishment during exercise in man. By C. E. FENN and R. J. MAUGHAN, *School of Nutritional Science, Robert Gordon's Institute of Technology, Queen's Road, Aberdeen AB9 2PG* and *Department of Environmental and Occupational Medicine, University Medical School, Foresterhill, Aberdeen AB9 2ZD*

The depletion of carbohydrate (CHO) stores and disturbances in fluid balance are possible limitations to prolonged exercise. The ingestion of significant quantities of CHO during exercise can delay gastric emptying which may interfere with both fluid and CHO delivery (Foster *et al.* 1980). We have examined the effect of exercise intensity and glucose concentration on CHO supply and fluid balance during exercise.

Six healthy male subjects exercised for 60 min on two occasions, 1 week apart, at workloads corresponding to approximately 50% maximum oxygen uptake ($\dot{V}_{O_2max}$, on two occasions 1 week apart at 60% $\dot{V}_{O_2max}$, and on two occasions 1 week apart at 70% $\dot{V}_{O_2max}$. On one occasion at each exercise intensity glucose (1·5 g/kg) was given orally immediately before exercise as a glucose polymer solution (Lucozade®, 190 g glucose/l, osmolality 630 mosmol/kg). On the other occasion an equivalent volume of a commercially available dilute glucose–electrolyte solution (Dioralyte®, 40 g glucose/l, osmolality 300 mosmol/kg) was given. Blood samples were obtained from a superficial forearm vein immediately before exercise, at 15 min intervals during exercise and for a 60 min rest period following exercise. Heart rate and rectal temperature were measured at 5-min intervals during exercise.

Blood glucose concentration during exercise was higher than resting values following Lucozade ingestion regardless of exercise intensity and was significantly ($P<0·05$) higher than that on the Dioralyte trial after 30, 45 and 60 min of exercise at 60, 50 and 70% $\dot{V}_{O_2max}$ respectively. Plasma insulin was significantly higher ($P<0·05$) after exercise on the Lucozade trial at 70% $\dot{V}_{O_2max}$ compared with 50% $\dot{V}_{O_2max}$. The rate of CHO oxidation during exercise, calculated from the respiratory gas exchange, was not different between the two trials at each exercise intensity. Plasma volume changes were calculated from haemoglobin and packed cell volume measurements as described by Dill & Costill (1974); there was a tendency towards a greater decrease in plasma volume when Lucozade was given but this was statistically significant only at 60% $\dot{V}_{O_2max}$. The rise in rectal temperature was not different between the Lucozade and Dioralyte trials. Heart rate was significantly higher after 30 min of exercise at 60% $\dot{V}_{O_2max}$ and remained higher until 50 min of exercise on the Lucozade trial compared with Dioralyte.

These results suggest that when Lucozade is taken a higher blood glucose concentration is maintained throughout exercise, within the range 50–70% $\dot{V}_{O_2max}$, but this is not reflected in a higher rate of CHO oxidation; moreover, at 60% $\dot{V}_{O_2max}$, CHO supply seems to occur at the expense of fluid replacement. Consequently the usefulness of CHO feeding during prolonged exercise of moderate intensity where thermoregulation is a priority, is questionable.

This study was approved by the local Ethics Committee.

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The acute effects of dietary variation on the distribution of high-density lipoprotein subspecies in active and inactive subjects. By B. A. GRIFFIN and E. R. SKINNER, *Department of Biochemistry, University of Aberdeen, Marischal College, Aberdeen AB9 1AS* and R. J. MAUGHAN, *Department of Environmental and Occupational Medicine, University Medical School, Foresterhill, Aberdeen AB9 2ZD* (Introduced by C. E. FENN)

Epidemiological studies have demonstrated a strong negative relation between the concentration of plasma high-density lipoproteins (HDL) and the incidence of coronary heart disease. Specific subspecies of HDL are now believed to confer coronary protection through the active removal of cholesterol from artery walls by a process of reverse cholesterol transport. Diet and physical activity have a major influence on HDL metabolism; their combined effect on HDL subspecies, however, is poorly understood and, as a consequence, has rarely been considered in studies designed to demonstrate the effects of diet and exercise in isolation. The present study aimed to determine the independent and combined effects produced by dietary variation and exercise on HDL subspecies.

The effects of a high-carbohydrate diet (85% energy) and a high-fat diet (75% energy) were each examined separately in active and inactive groups of six male subjects. The dietary periods lasted 4 d during which group 1 subjects walked 12·3 km/d while group 2 subjects remained inactive. The effects of a normal diet were also examined in both groups.

The high-carbohydrate and high-fat diets produced opposite effects with respect to the concentration of total HDL-cholesterol (HDL-C). The high-fat diet significantly raised HDL_2-C during the dietary period ($P<0·001$ and $P<0·05$ for groups 1 and 2 respectively). Conversely, the high-carbohydrate diet produced significant decreases in HDL_3-C ($P<0·01$) for group 1 and HDL_2-C ($P<0·01$) for group 2. In general the combination of exercise and a high-fat intake produced an enhanced response, less pronounced changes being observed with exercise and a high-fat diet alone, whereas the exercise minimized the reductive influence of the high-carbohydrate diet on HDL_2.

From these observations an independent but additive stimulation of lipoprotein lipase (LPL, *EC* 3.1.1.34) activity by a high-fat diet and exercise could be inferred. Similarly, the decrease in HDL_2 produced by the high-carbohydrate diet suggests a suppression of LPL-mediated lipolysis. Therefore, acute dietary variation can independently cause a redistribution of HDL subspecies, this response being greatly influenced by levels of physical activity.

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The effect of diet and prolonged walking on fluid homeostasis. By J. B. LEIPER, C. E. FENN and R. J. MAUGHAN, *Department of Environmental and Occupational Medicine, University Medical School, Foresterhill, Aberdeen AB9 2ZD*

Prolonged, low-intensity walking produces an increase in plasma volume in healthy, adequately hydrated subjects consuming a mixed diet. We proposed that the primary stimulus for the increase in plasma volume was a postural effect activating the renin–aldosterone system. However, diet may play an important role in plasma expansion. We therefore examine the effect of isoenergetic high-carbohydrate (H-CHO) and low-carbohydrate (L-CHO) diets on fluid homeostasis in the same individuals during prolonged walking.

Six healthy males completed a 3 lap daily walk of 37 km for four consecutive days on two separate occasions. Mean exercise intensity was equivalent to 17 (SE 1)% maximum oxygen uptake ($\dot{V}_{O_2max}$) for these subjects. On one occasion half of the subjects consumed the H-CHO (CHO 85% energy) and the other subjects consumed the L-CHO (CHO 25% energy) diet. This dietary regimen was reversed on the other occasion. Food intake was adjusted to equate with the energy intake of each subject on a previous walking study where an unrestricted mixed diet was consumed. Subjects walked fasted each day and food was consumed after collection of the final blood sample for that day. Water was allowed *ad lib.* between laps. Nude body-weight was measured on each morning of the study. Fluid balance was calculated from total daily fluid intake and 24 h urine output. Before exercise and within 1–2 min of completing each lap, each day venous blood samples were collected while the subject remained standing. Plasma volume changes were calculated from the haematological variables. Serum and urinary electrolytes, total protein and urea were measured. By the first post-exercise day subjects on the L-CHO diet had lost 2·0 (SE 0·3) kg in weight; there was no weight change on the H-CHO diet. Fluid balance was similar on the two diets except for day 3 where total fluid intake on the L-CHO diet was lower (2558 (SE 230) *v.* 3360 (SE 249) ml, $P<0·05$). Plasma volume increased during each day's walk on the H-CHO diet and by day 4 was 19 (SE 4)% greater compared with the day 1 pre-exercise level. There was no comparable haemodilution during exercise on the L-CHO diet, although on the first post-exercise day plasma volume increased by 11 (SE 3)%. On the L-CHO diet serum chloride but not sodium concentration increased during exercise on days 3 and 4 ($P<0·05$). Compared with the day 1 pre-exercise value, serum potassium concentration was higher ($P<0·05$) on days 2 and 3 on the H-CHO diet; no other consistent change in serum electrolytes occurred. Serum total protein ($P<0·05$) and urea ($P<0·001$) concentrations were higher on the L-CHO than on the H-CHO diet. By day 3, urinary Na losses and Na:K ratios were higher on the L-CHO diet.

These results indicate that plasma volume increases which can occur during low-intensity exercise are greatly affected by the diet consumed. The plasma expansion seen on the H-CHO diet may be due to insulin-induced Na retention.

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