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Correlation between dietary and urinary phyto-oestrogens (genistein and daidzein) in healthy adults. By M.R. RITCHIE[1], M.S. MORTON[2], C. BOLTON-SMITH[3] and J.H. CUMMINGS[1], [1]*Department of Molecular and Cellular Pathology, Ninewells Hospital and Medical School, Dundee DD1 9SY,* [2]*Bioclinical Services, Cardiff CF3 OEF and* [3]*MRC-HNR, Downhams Lane, Cambridge CB4 1XY*

The intake of phyto-oestrogen (PE) rich foods varies considerably by country and in populations where the incidence of breast and prostate cancers is low, intake of PE tends to be high (Messina *et al.* 1994). The aim of this study was to assess PE intake in healthy adults and to examine the relationship with urinary excretion of PE.

Ten vegetarians and nine omnivores, aged 19–76 years, completed individual 7-day weighed food diaries. Volunteers were weighed at the beginning and end of the recording period and height was measured. The BMR was calculated for each volunteer and the energy intake (EI) to BMR ratio of ≥1.2 was used as a measure of acceptable food recording. Subjects with EI:BMR below this were excluded. Around 600 PE (total genistein + daidzein) values for UK foods were identified from published (Liggins *et al.* 2000) and unpublished sources (J Liggins & S Bingham, unpublished results; H Wiseman, unpublished results), major retail outlets and recipe calculations. These data were incorporated into the *Microdiet* food analysis programme. Mean PE intakes and food sources were determined for the vegetarians and omnivores separately. Subjects also completed a 24-hour urine collection after taking para-amino benzoic acid (PABA) to provide a marker of complete collection (Bingham & Cummings, 1983). Urine collections with a PABA recovery <85 % were rejected. Urine collections from sixteen subjects (ten vegetarians, six omnivores) were analysed by GC-MS for PE (genistein, daidzein, equol and glycitein) and lignan (enterolactone) content.

	Dietary genistein + daidzein (mg/d)		Urinary genistein + daidzein (mg/d)		Urinary equol (ug/d)		Urinary enterolactone (mg/d)	
Dietary group	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Vegetarian (*n* 10)	6.6	10.0	2.9	3.3	30.1	71.0	1.75	2.5
Omnivore (*n* 9, *n* 6)[a]	4.2	6.2	1.4	2.8	11.1	3.9	0.83	0.9
All (*n* 19, *n* 16)[b]	5.5*	8.3	2.4*	3.1	22.9	55.8	1.41	2.1

[a]*n* 9 for dietary values, *n* 6 for urinary values; [b]*n* 19 for dietary values, *n* 16 for urinary values; * P=0.042 for Pearson correlation coefficient between total dietary and urinary genistein+daidzein.

The main food sources of PE for the total group in order of decreasing PE content were tofu, soya mince, soya yoghurts, soya milk, vegetarian sausages made with soya protein and all types of bread. The Pearson correlation coefficient (r) between dietary and urinary genistein + daidzein (*n* 16) was (r 0.514, P=0.042) for both groups together. Whilst intakes and urinary amounts of PE tended to be higher in the vegetarian group, these differences were not significant due to small sample size and large within-group variation.

This study used a newly set-up, and preliminary, database of the PE (genistein and daidzein) content of foods commonly eaten in Britain. Estimating PE intake from food diaries is a major, and ongoing, challenge because soya flour is routinely, but not uniformly, added to commercial products, particularly bakery goods. Obtaining an accurate assessment of total urinary excretion of PE is also hampered by the current lack of knowledge about the variety of PE metabolites. In spite of these problems in assessing PE intake and urinary excretion, a significant correlation has been demonstrated between the two measures used in this study. These results compare favourably with those reported by Arai *et al.* (2000), where correlation coefficients between dietary and urinary levels were (r 0.365) for daidzein and (r 0.346) for genistein (P<0.001), with a sample size of 106 women.

Arai Y, Uehara M, Sato Y, Kimira M, Eboshida A, Adlercreutz H & Watanabe S (2000) *Journal of Epidemiology* **10**, 127–135.
Bingham S & Cummings JH (1983) *Clinical Science* **64**, 629–635.
Liggins J, Bluck LJC, Runswick S, Atkinson C, Coward WA & Bingham SA (2000) *British Journal of Nutrition* **84**, 717–725.
Messina MJ, Persky V, Setchell KDR & Barnes S (1994) *Nutrition and Cancer* **21**, 113–131.

Dietary cravings and aversions in pregnancy. By B. AL-RASASI[1], R. SIEGLER[2], J. NICHOLS[3], J. COAD[1] and J. MORGAN[2], [1]*European Institute of Health and Medical Sciences, University of Surrey, Guildford GU2 7TE,* [2]*School of Biomedical and Life Sciences, University of Surrey, Guildford GU2 5XH and* [3]*Fairlands Medical Centre, Guildford GU3 3NA*

The occurrence of dietary cravings and aversions during pregnancy is well known, yet relatively little is known of the aetiology or epidemiology of this complex of symptoms. The objectives of this study were to ascertain the prevalence of cravings and aversions in a sample of women in Guildford, to find out the food types craved and avoided by pregnant women, and to investigate maternal factors associated with dietary cravings and aversions in pregnancy.

A retrospective study was performed using questionnaires mailed to women who had delivered a baby in the past year. The questionnaire was divided into three sections gathering information about the mother, father and baby, both open and closed questions were used; specifically for cravings and aversions only opened questions were used. The women were recruited from two GP surgeries in Guildford. The sample size was 201, all white women who had given birth to a single baby. Data were analysed using SPSS, both descriptive and comparative statistics were used.

We found that 51% (*n* 102) of the women reported at least one craving and 42% (*n* 82) at least one aversion. The foods most commonly craved were fruit and fruit juices (*n* 37 (36%)), confectionery (*n* 31 (29%)) and dairy products (*n* 31 (29%)). With respect to aversions, the most common foods reported were stimulating drinks such as tea, coffee and cola (*n* 73 (87%)), strong-tasting foods such as curries and other spicy food (*n* 25 (30%)), and fatty/greasy foods such as fried foods (*n* 13 (15%)). There were also two reportings of pica; one for ice cubes, and the other for coal.

We found that women who drank alcohol pre-pregnancy reported a higher frequency of cravings compared to those women who did not drink alcohol pre-pregnancy (*n* 86 (83%) and *n* 16 (15%) respectively, *P*=0.05). We also found that women of social classes I and II were more likely to develop aversions than those women from social classes III, IV and V (*n* 70 (86%) and *n* 9 (12%) respectively, *P*=0.032). Women suffering from nausea and vomiting in pregnancy (NVP) reported significantly more cravings compared to those women who did not suffer from NVP (*n* 66 (80%) and *n* 15 (20%) respectively, *P*=0.014). Women suffering from NVP also reported significantly more aversions compared to those women who did not suffer from NVP (*n* 86 (81%) and *n* 17 (16%) respectively, *P*=0.017). The frequency of aversions was also positively higher in women with a degree or post degree compared to those women without a degree (*n* 57 (67%) and *n* 26 (29%)) respectively, *P*<0.001). A positive relationship was also seen between women reporting aversions and those reporting cravings (*n* 52 (61%) and *n* 30 (38%) respectively, *P*=0.008). We also investigated the aversions and cravings of those women who had NVP and found that they cited slightly different foods, compared with those who did not have NVP.

In this affluent area in the south-east of England, the incidence of cravings seems to be similar to that reported in other studies carried out in the UK. The incidence of aversions was lower, which may be related to geographic and socio-economic differences. Although many authors have suggested that NVP could be a contributory factor to cravings and aversions in pregnancy, few, if any, studies have actually found a significant relationship between them. Our study has shown that NVP and aversions/cravings in pregnancy are strongly related. The relationship between NVP and aversions may provide further evidence that NVP could be a protective mechanism in causing the mother to reduce her intake of foods that may be potentially harmful to the fetus.

B. Al-Rasasi is in receipt of a University of Surrey Scholarship and an Overseas Research Council Scholarship.

Comparison of a semi-quantitative food frequency questionnaire with 4-day weighed diet records in Scottish men and women. By L.F. MASSON[1], G. McNEILL[1], J.O. TOMANY[1], H.S. PEACE[1], C. BOLTON-SMITH[2] and D.A. GRUBB[3], [1]*Department of Medicine & Therapeutics, University of Aberdeen, Foresterhill, Aberdeen AB25 2ZD,* [2]*Nutrition Research Group, CVEU, University of Dundee, Dundee DD1 9SY and* [3]*Rowett Research Institute, Bucksburn, Aberdeen AB21 9SB*

The Scottish Collaborative Group 150-item semi-quantitative food frequency questionnaire (FFQ) has been developed over a number of years for use in a wide range of epidemiological studies. The current version (6.31) was compared with 4-d weighed records (WR) in healthy subjects recruited from different locations in Aberdeen. Forty-one men, mean age 36 (range 21–56) years, and forty women, mean age 33 (range 19–58) years carried out a WR for three weekdays and one weekend day, starting a few (range –9 to +9) days before or after completing the FFQ. Wide ranges of nutrient intakes were obtained by both methods, e.g. by WR total fat varied from 21 to 46% of energy intake for men and from 7 to 44% of energy intake for women. Nutrient intakes were log-transformed and energy adjusted. Pearson correlation coefficients between the FFQ and WR were calculated for all subjects and for those with energy intake/basal metabolic rate (EI:BMR) ratios >1.14 for FFQ and >1.06 for WR (Goldberg *et al.* 1991). Correlation coefficients for log-transformed energy were 0.35 (P=0.03) for all men (n 41) and 0.40 (P=0.01) for all women (n 40). Subjects were grouped into thirds of intake, and the percentages of subjects in the same third and opposite third by the two methods were calculated.

Nutrient	Correlation coefficients (r)				Classification into thirds			
	All subjects		EI/BMR>cut-off		% same third		% opposite third	
	M (n 41)	F (n 40)	M (n 29)	F (n 30)	M (n 41)	F (n 40)	M (n 41)	F (n 40)
Total fat	0.54*	0.83*	0.63*	0.86*	46	50	15	5
SFA	0.55*	0.81*	0.51‡	0.87*	51	60	5	10
PUFA	0.08	0.68*	0.11	0.67*	29	50	20	5
MUFA	0.33†	0.86*	0.50‡	0.83*	39	40	12	5
NSP	0.64*	0.73*	0.66*	0.79*	54	53	2	3
Starch	0.57*	0.55*	0.49‡	0.58*	51	45	7	5
Sugar	0.41‡	0.72*	0.40†	0.71*	46	68	10	3
Alcohol	0.83*	0.70*	0.79*	0.78*	71	50	5	5
Retinol	–0.49*	0.37†	–0.46†	0.40†	24	50	29	15
β-carotene	0.16	0.51*	0.19	0.48‡	37	48	22	8
Folate	0.56*	0.78*	0.42†	0.82*	42	58	5	3
Vitamin C	0.64*	0.68*	0.69*	0.66*	49	60	7	10
Vitamin E	0.21	0.52*	0.35	0.51‡	39	53	12	8
Calcium	0.52*	0.78*	0.37†	0.77*	54	63	7	3
Iron	0.63*	0.64*	0.65*	0.68*	56	53	5	8

SFA, saturated fatty acids; PUFA, polyunsaturated fatty acids; MUFA, monounsaturated fatty acids; NSP, non-starch polysaccharides. *$P\leq0.001$, ‡$P\leq0.01$, †$P\leq0.05$.

In general the FFQ performed well, with correlation coefficients >0.50 ($P\leq0.001$) for all nutrients except retinol in men and women, and PUFA, MUFA, sugar, β-carotene and vitamin E in men. Excluding individuals using EI:BMR cut-offs made little change to the coefficients in women, but more inconsistent changes in men. Women tended to have better correlation coefficients and classification into thirds than men, except for alcohol which was better in men, and starch and iron which were similar in both respects in men and women. The lower results for most nutrients in men may be due to lower awareness of the types and amounts of foods consumed, e.g. fat spreads in relation to PUFA. The poor results for retinol in both men and women are likely to reflect the fact that 4 d is too short a time for making an adequate estimation of habitual retinol intake by WR.

Goldberg GR, Black AE, Jebb SA, Cole TJ, Murgatroyd PR, Coward WA & Prentice AM (1991) *European Journal of Clinical Nutrition* **45**, 569–581.

Infusion of glucose and essential amino acids downregulate the expression of components of the ubiquitin-dependent proteolytic pathway in growing calves. By F. SADIQ[1], M.C. WALLACE[1], J. STRUTHERS[1], J.R. SCAIFE[1], L.M. BIRNIE[1], D. ATTAIX[2] and M.A. LOMAX[1], [1]*Department of Agriculture & Forestry, Aberdeen University, 581 King Street, Aberdeen AB24 5UA and* [2]*INRA, Nutrition and Protein Metabolism Unit, 63122 Ceyrat, France*

The ubiquitin proteasome pathway is the predominant biological pathway for the breakdown of myofibrillar proteins (MF) in skeletal muscle. Anabolic hormones such as insulin and IGF-1 (Fang *et al.* 2000) have been observed to control this pathway through their anti-proteolytic effects. Larbaud *et al.* (1996) have shown that a 6-h hyperinsulinaemic, hyperaminoacidaemic, euglycaemic clamp-down regulates ubiquitin gene expression in ruminants. Amino acids have been proposed to increase the sensitivity of muscle protein turnover to insulin, although our previous studies failed to observe any effect of essential amino acids (EAA) on the decrease in 3-methyl histidine excretion (an index of MF breakdown) during glucose infusion in calves. The present study aimed to establish the effects of a continuous infusion of essential amino acids, either alone or in combination with a low or high dose of glucose (to stimulate insulin secretion) on the expression of components of the ubiquitin proteolytic pathway in growing calves.

Six Holstein-Friesian calves (approximately 4 months of age and mean body weight 90 kg) were housed separately in metabolism units. Calves were kept on a restricted diet to achieve 0.3 kg daily body weight gain. All animals were allocated to six treatments in a randomized 3 × 2 factorial design. Treatments comprised: saline (control); high-dose glucose (20 µmol/kg body weight (BW)/min; HDG); low-dose glucose (9.6 µmol/kg BW/min; LDG); mixtures of EAA (0.8 mg/kg BW/min); HDG & EAA and LDG & EAA. Treatments were infused for 5 d via jugular vein catheters and blood samples and muscle biopsies were collected on day 5 of the infusion period. Plasma was analysed for glucose, insulin and IGF-1. RNA was extracted from muscle biopsy samples and Northern hybridization was performed for components of the ubiquitin-dependent proteolytic pathway (14-kDa ubiquitin-conjugating enzyme E2, polyubiquitin, C2 and C8 20S proteasome subunits). RNA blots were also hybridized against GAPDH, a housekeeping gene. Ratios of components of ubiquitin pathway to GAPDH were analysed statistically using the Minitab general linear model .

Infusion of LDG & HDG significantly increased plasma glucose, insulin and IGF-1 concentrations, compared with control values, in a dose-dependent manner (P<0.05, Sadiq *et al.* 2001). Relative to GAPDH, expression of 14-kDa E2, C2 and C8 was decreased by either LDG or HDG, with no consistent effects of level of glucose infusion. Co-infusion of amino acids did not alter the response to glucose infusion. Decreased ubiquitin gene expression was observed when glucose was infused; however, this decrease was not significantly different from control levels (P>0.05).

	Control		LDG		HDG		EAA		LDG+EAA		HDG+EAA	
	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
14-kDa E2	57.1[a]	11.8	20.7[bc]	3.7	27.7[ac]	6.2	39.9[ac]	5.1	35.7[ac]	4.2	30.5[ac]	4.5
Polyubiquitin	12.2[a]	2.9	9.5[a]	1.4	9.3[a]	1.4	7.6[a]	0.9	8.6[a]	1.2	8.7[a]	1.8
C2	19.9[a]	3.9	6.4[bc]	3.0	11.1[ac]	2.5	12.3[ac]	3.2	9.2[bc]	1.9	7.1[bc]	0.5
C8	3.2[a]	0.5	2.6[ac]	0.3	1.7[bc]	0.5	2.8[ac]	0.4	2.3[ac]	0.3	2.5[ac]	0.4

[a,b,c] Values (arbitrary units) within the same row with different superscripts are significantly different (P<0.05).

The present results suggest that long-term infusion of insulinogenic compounds may suppress the expression of the C2 proteasome subunit (which is part of the proteolytic core of the 26S proteasome that degrades ubiquitin conjugates) *in vivo*. There was no consistent dose-dependent effect of glucose infusion on components of the ubiquitin system. Moreover, addition of EAA to glucose infusions did not further enhance the anti-proteolytic response of insulin, suggesting that EAA are not key suppressors of the ubiquitin proteasome pathway in skeletal muscle.

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Larbaud D, Debras E, Taillandier D, Samuels SE, Temparis S, Champredon C, Grizard J & Attaix D (1996) *American Journal of Physiology* **271**, E505–E512.
Sadiq F, Wallace MC, Struthers J, Scaife JR, Birnie LM & Lomax MA (2001) *Proceedings of the Nutrition Society* 60, 28A.

Comparison of the effects of different ratios of *n*-6 and *n*-3 polyunsaturated fatty acids on faecal egg output and mucosal mast cell numbers in a mixed model nematode infection in pre-ruminant calves. By K.N. MUTURI[1], M. WALLACE[1], J. STRUTHERS[1], J. SCAIFE[1], M.A. LOMAX[1], F. JACKSON[2], E. JACKSON[2], J. HUNTLY[2], A. MACKELLAR[2] and B. COOP[2], [1]*Dept. of Agriculture and Forestry, University of Aberdeen, Aberdeen AB24 5UA and* [2]*Moredun Research Institute, Pentlands Science Park, Penicuik, Edinburgh EH26 0PZ*

Ruminant parasites are increasingly resistant to treatment with anthelmintics (Jackson & Coop, 2000). Increasing the effectiveness of the immune system of young animals to resist parasitic infections may offer a protective mechanism and therefore a reduction in the requirement for anthelmintic. Diet-induced changes in the PUFA content of immune cells, particularly the ratio of *n*-6 to *n*-3 PUFA, have been shown to affect the functions of the various components of the immune response such as secretion of cytokines, antibodies, antigen reception from the antigen presenting cells, lymphocyte transformation and contact lysis (Calder, 1998). The aim of this study was to establish the extent to which inflammatory and immune responses in the intestinal mucosa of the calf, stimulated by nematode parasite antigen, may be modified by manipulation of the composition of the dietary PUFA available for incorporation into mucosal immune and epithelial cell membranes.

Twenty-four pre-ruminant calves, aged 17–80 d were allocated, according to age, to two treatment groups which received, as a supplement to milk replacer, 25 g/d of either an *n*-3 PUFA-rich fish oil (*n*-3) or a mixture of palm oil and rape seed oil (normal) designed to supply the same fatty acid profile as found in milk replacer. Within each treatment group, eight calves were infected with 2000 stage 3 larvae (L3) of an abomasal nematode (*Ostertagia ostertagi*) and intestinal nematode (*Cooperia onchophara*) three times/week. The remaining four animals in each group received the PUFA supplement but were not infected. Faeces were collected twice weekly from days 21–56 post-infection and faecal egg counts (FEC) were measured. At the end of the experiment, abomasal and intestinal samples were collected for immunohistological analysis. Mucosal mast cells were identified and counted after staining with toluidine blue. The counts were performed on ten different areas of view and expressed as mean cell numbers per 0.2 mm^2. Data were analysed by one-way (ANOVA) using Minitab.

Parasite infection was established by 28 d post-infection in both treatment groups and reached a peak, as judged by FEC, by day 35. There were no significant differences in weekly FEC between the two infected groups, but numbers in the *n*-6 supplemented/infected group tended to remain higher. There were significant differences in mucosal mast cell numbers between the *n*-3 and *n*-6 supplemented groups ($P<0.05$). In intestinal samples, mast cell numbers were significantly increased by infection and were higher in animals fed the *n*-6 supplement.

		n-3 infected (*n* 8)		normal infected (*n* 8)		*n*-3 not infected (*n* 4)		normal not infected (*n* 4)	
		Mean	SEM	Mean	SEM	Mean	SEM	Mean	SEM
Faecal egg counts	Week 1	815[a]	166	932[a]	163				
	Week 2	1664[a]	300	1478[a]	327				
	Week 3	884[a]	262	1402[a]	590				
	Week 4	490[a]	96	1240[a]	528				
	Week 5	552[a]	129	743[a]	340				
Mast cells	Abomasal	23[a]	2.0	27[a]	2.7	24[a]	1.2	45[b]	3.0
(Mean cells/10	Duodenal	37[a]	2.3	46[b]	2.7	27[c]	1.2	32[d]	1.7
areas of view)	Mid-gut	42[a]	2.3	50[b]	2.8	18[c]	0.7	28[d]	4.3

Values are means and SEM. Means with different superscripts on the same line are significantly different ($P<0.05$).

The results demonstrate that the source of oil has no effect on the faecal egg counts in pre-ruminant calves, but has effects on the numbers of mucosal mast cells. The reduction in mucosal mast cell numbers in the fish oil-supplemented calves could be an indicator of suppression in the migration or proliferative capacity of mast cells from their precursors, into the epithelial and mucosal surfaces. Why this was not translated into an advantage in terms of immune response (FEC) for the normal group which had higher mast cell numbers remains to be elucidated.

Our thanks to Harbro Farm Sales Ltd (Turriff) for the gift of palm oil and fish oil supplements.
N. Muturi is an Aga Khan Foundation scholar.

Calder PC (1998) *Proceedings of the Nutrition Society* **57**, 487–502.
Jackson F & Coop RL (2000) *Parasitology* **120**, S95–S107.

The effect of rumen-protected protein on plasma insulin, growth hormone, IGF-1 and glucose in sheep. By K.N. MUTURI, L.M. BIRNIE, J. STRUTHERS, M. DONALD and M.A. LOMAX, *Department of Agriculture and Forestry, MacRobert Building, University of Aberdeen, Aberdeen AB24 5UA*

Ruminants fed at maintenance energy intake have been shown to deposit lean tissue, as long as adequate protein is supplied in the diet (Fattet *et al*. 1984). The mechanisms underlying this may be partially regulated by the endocrine system via modulation of anabolic hormonal signalling, since circulating levels of insulin and insulin-like growth factor-1 (IGF-1) are increased by the supply of high levels of protein in energy-restricted diets (Kriel *et al*. 1992). The primary aim of this study was to examine the extent to which growth hormone (GH), IGF-1 and insulin concentrations in plasma are modulated by the inclusion of a rumen-protected, soya-based protein (amino green 98) in lambs restricted in metabolizable energy (ME) intake.

In a cross-over design, six Suffolk-cross wether lambs, weighing 45.0±2.2 kg live weight were fed isocaloric diets formulated to supply 8.6 MJ/ME and either 65 g (low protein) or 200 g (high protein) of metabolizable protein (MP) per day. The diets were composed of (kg DM/d): low protein: 0.47 hay, 0.25 barley, 0.08 amino green 98; high protein: 0.46 hay, 0.1 barley, 0.29 amino green 98. Dietary adaptation periods were 14 d in duration and blood sampling was performed over 8 h, at 15-min intervals on the last day of each dietary period. Plasma samples were analysed for GH, insulin glucose and IGF-1. Data were analysed by two-way ANOVA.

There was a significant effect of the high protein diet on plasma insulin concentrations ($P<0.05$) and this was particularly marked post-feeding. No significant effects of diets on plasma GH and IGF-1 were observed. Glucose concentrations were significantly different between the high and low protein diets at 60 (4.1 (SD 0.19) *v.* 3.5 (SD 0.11 mM) , 90 (4.4 (SD 0.18) *v.* 3.6 (SD 0.17) mM)and 120 (4.5 (SD 0.24) *v.* 3.6 (SD 0.19) mM) min post-feeding ($P<0.05$) but there were no significant differences between the diets over the entire sampling period of 8 h.

	Low protein		High protein		Significance
	Mean	SEM	Mean	SEM	
Insulin (ng/ml)	0.32	0.03	0.42	0.06	*
GH (ng/ml)	5.17	2.59	6.07	2.44	NS
IGF-1 (ng/ml)	163.5	14.5	170	12.4	NS
Glucose (mM)	3.2	0.19	3.7	0.34	NS

Values are means averaged over 8 h with their SEM. Significantly different, *$P<0.05$; NS, not significant.

The present results clearly demonstrate that a rumen-protected vegetable protein is effective in stimulating an increase in insulin response to feeding in this study, without altering plasma GH and IGF-1 values. The insulin response post-feeding may be stimulated by the absorption of increased amino acids from the small intestine and could provide a mechanism to improve the nitrogen balance of ruminants at maintenance levels of feeding.

Our thanks to Northeastern Farmers, Turriff for the contribution of amino green, Diagnostic Scotland for IGF-1 and insulin antibodies, Dr A.F. Parlow (National Institute of Health) for GH antibody and Tana & Athi Rivers Development Authority (Kenya). This work was funded by the Department of Agriculture and Forestry, University of Aberdeen.

Fattet I, Hovell FDDeB, Ørskov ER, Kyle DJ, Pennie K & Smart RI (1984) *British Journal of Nutrition* **52**, 561–574.
Kriel GV, Bryant MJ & Lomax MA (1992) *Journal of Endocrinology* **132**, 195–199.

Programming of blood pressure and renal structure in rats exposed to nitrogen-supplemented maternal low protein diets. By M.C. MARCHAND[1], R.L. DUNN[2], A.A. JACKSON[2] and S.C. LANGLEY-EVANS[1], [1]*Human Nutrition and Metabolism Group, Centre for Healthcare Education, University College Northampton, Northampton NN2 7AL and* [2]*Institute of Human Nutrition, Southampton General Hospital, Southampton SO16 6YD*

A large and consistent body of experimental evidence suggests that exposure of the rat fetus to a maternal low protein (MLP) diet induces growth retardation, elevated blood pressure and renal dysfunction (Langley-Evans, 1999). At present our understanding of what constitutes an optimal maternal diet for fetal growth and development is limited. Observations of low nephron number, albuminuria and abnormal renal metabolism of vasoactive prostaglandins are all suggestive of a role for the kidney in the prenatal programming of hypertension (Langley-Evans, 1999). In order to further evaluate the relationship between fetal diet, later blood pressure and renovascular function, MLP diets were supplemented with a range of different nitrogen sources.

A total of twenty-nine virgin Wistar rats (200–225 g) were mated and allocated to experimental diets. Of these, twenty-six successfully completed pregnancy, producing 182 offspring. As previously described (Langley-Evans, 1999), six pregnant rats were fed an a control diet (CON) (18% casein) and six were fed a diet containing 9% casein (MLP). In addition five rats were fed 9% casein + 3% glycine (MLPG), four were fed 9% casein + 1.5% urea (MLPU), and five were fed 9% casein + 3% alanine (MLPA). At littering, all animals were returned to standard laboratory rat chow. At 4 weeks of age, systolic blood pressure (SBP) and pulse were determined in all offspring using the tail cuff method. The rats were then culled for tissue collection. Nephron number was determined in ten randomly selected formalin-fixed kidneys from each group, following sectioning (5 μm) through the hilar plane and staining with haemotoxylin and eosin (Langley-Evans *et al.* 1999).

Maternal diet	SBP (mmHg)			Pulse (beats/min)		Nephrons/kidney		
	n	Mean	SE	Mean	SE	Mean	*n*	SE
CON	39	109[a]	3	400[a]	6	49451[a]	10	2541
MLP	41	120[b]	3	398[a]	6	32311[b]	10	5306
MLPU	32	124[b]	3	358[a]	7	51567[a]	10	5407
MLPA	36	120[b]	2	351[b]	7	41273[a]	10	3876
MLPG	34	100[a]	3	383[a]	6	45656[a]	10	4954

Data represent mean and SEM for *n* observations. Groups with different superscript letters are significantly different (P=0.05).

Compared to the CON diet the feeding with the MLP diet induced significant elevation of SBP, as previously noted (Langley-Evans, 1999). The addition of glycine, however, completely abolished the hypertensive effect of the MLP diet. The addition of urea and alanine to the MLP diet at levels which gave equivalent nitrogen as the added glycine had no significant effect on SBP. All three groups of pregnant animals fed nitrogen-supplemented MLP diets generated offspring with a nephron capacity equivalent to the CON group. In particular, urea appeared to remove the effect of MLP on nephrogenesis, but not upon SBP. The effect of glycine on SBP appeared to be specific and is not explained by the addition of amino nitrogen or a non-essential amino acid carbon skeleton. Given the central role of glycine in a number of different metabolic pathways (Jackson, 1991), it is possible that this amino acid is limiting in the maternal low-protein diet, and that this may influence renovascular structure, functional development and other key metabolic processes. The data indicate that dietary interventions, which correct deficits in nephron reserve, may not exert a similar influence on SBP. This suggests that mechanisms responsible for fetal programming of blood pressure in rats may operate independently of the kidney.

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Relationships between polyunsaturated fatty acid (PUFA) composition of umbilical cord, brain and liver in new-born piglet born to sows fed diets differing in *n*-3 PUFA composition. By J.A. ROOKE, A.G. SINCLAIR, L.M. BIRNIE and M. EWEN, *Animal Biology Division, SAC, Craibstone Estate, Aberdeen AB21 9YA*

In a previous experiment (Rooke *et al.* 1999), the PUFA proportions of entire (tissue plus blood vessels) umbilical cord samples were concluded to be poor predictors of piglet tissue PUFA composition. The objective of the present study was to investigate, using a range of maternal *n*-3 intakes, whether umbilical artery or vein fatty acid composition might be a superior predictor of piglet tissue PUFA composition at birth than entire umbilical cord.

Samples of cord (one per litter) were obtained at farrowing from sows (five or six per diet) fed each of six diets from two experiments. The cord samples were separated into artery, vein and remaining tissue and stored at –20º. In Expt 1 (Cordoba *et al.* 2000), salmon oil (17.5 g/kg diet) replaced vegetable oil, giving two diets, whereas in Expt 2 (Rooke *et al.* 2001), palm oil (20 g/kg diet) was progressively replaced by 0, 5, 10 and 20 g salmon oil/kg to give four diets. The ingredient composition of the basal diets also differed between experiments. Brain and liver fatty acid compositions were determined on samples obtained from piglets killed prior to colostrum ingestion and selected to represent the mean weight of each litter.

Umbilical artery and vein contained (g/100 g total fatty acids) significantly lower proportions of 18:2 *n*-6 (P<0.001) than cord tissue and greater proportions of C20 and C22 long-chain PUFA including 20:3 *n*-9 (0.61 *v.* 0.38; P<0.05). Increasing proportions of long-chain *n*-3 PUFA in the maternal diet resulted in linear increases in 20:5 *n*-3 (P<0.001) and 22:6 *n*-3 (P<0.01) and curvilinear decreases (P<0.001) in 20:4 *n*-6, 22:4 *n*-6, 22:5 *n*-6 proportions in cord, artery and vein samples; there were no diet × tissue interactions. When correlation analyses were used to explore the relationships between piglet brain and liver PUFA proportions and umbilical cord, artery and vein PUFA proportions, the correlation coefficients obtained were significantly greater for artery than for vein (P 0.003) or umbilical tissue (P<0.001). Relationships were best explained by simple linear relationships. The Table shows significant relationships between artery PUFA (x) and piglet tissue PUFA (y); relationships for long-chain *n*-3 PUFA were non-significant.

Piglet tissue	Brain			Liver		
Fatty acid (g/100 g total fatty acids)	Intercept	Slope	r^2	Intercept	Slope	r^2
20:4 *n*-6	121	0.15	0.09^{NS}	70	0.38	0.24**
22:4 *n*-6	40.4	0.32	0.18*	1.5	0.14	0.31**
22:5 *n*-6	8.6	0.91	0.33***	5.3	0.55	0.36***
22:5 *n*-6/22:6 *n*-3	5.0	1.23	0.35***	3.8	0.99	0.21**

In conclusion, the PUFA composition of umbilical cord, artery and vein was influenced by maternal *n*-3 PUFA intake. Artery PUFA measurements were the best predictors of piglet brain and liver PUFA composition. However, predictive relationships between artery PUFA and piglet tissue PUFA were not strong. To improve the relationships, it may be necessary to analyse tissue from more than one umbilical cord per litter.

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Maternal docosahexaenoic acid supplementation and fetal accretion. By C. MONTGOMERY[1], B. SPEAKE[2], A. CAMERON[3], N. SATTAR[4] and L.T. WEAVER[1], [1]*Department of Child Health,* [3]*Department of Fetal Medicine,* [4]*Department of Clinical Biochemistry, University of Glasgow and* [2]*Scottish Agricultural College, Auchencruive, Ayr KQ6 5HW*

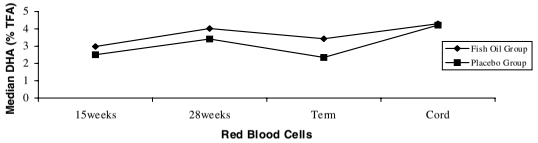
Docosahexaenoic acid (DHA) (C22:6*n*-3) is a polyunsaturated fatty acid that is an essential constituent of membranes, particularly those of the nervous system. DHA is obtained in the diet largely from fish and fish products. DHA status correlates with fish intake, even in the west of Scotland where consumption is low (Berry *et al.* 2001). Maternal supplies of DHA are transferred to the fetus via the placenta, and to the neonate via breast-milk. Babies who are inadequately supplied with DHA may be at a disadvantage both biochemically and developmentally (Jamieson *et al.* 1999). To test the hypothesis that maternal supplementation enriches maternal DHA status and increases the amount available to the fetus and neonate, 100 mothers were randomly assigned to receive either fish oil (*n* 50) or placebo (*n* 50) capsules from 15 weeks gestation until term; sixty-two mothers remained in the study at term (thirty-one in each group). The fish oil capsules contained 40% DHA (200 mg/day); the placebo consisted of high oleic sunflower oil containing 81% oleic acid (400 mg/day).

Maternal venous blood samples were obtained at 15, 28 and 40 weeks. Umbilical cord blood, umbilical cord tissue and placental tissue were collected at term. Breast-milk was obtained soon after birth. Total fatty acids in plasma, red blood cells (RBC), cord/placental tissue and breast-milk were extracted and derivatized; fatty acid methyl esters were analysed by GCMS. Results were expressed as percentage total fatty acids (% TFA) and concentrations (nmol/ml for RBC, plasma and breast-milk, nmol/g wet tissue for placenta). Differences between groups were detected using Mann-Whitney two-sample rank tests ($P<0.05$). The Table shows median concentrations of DHA in plasma and RBC.

Conc (nmol/ml)	RBC				Plasma			
	15weeks	28weeks	Term	Cord	15weeks	28weeks	Term	Cord
Fish oil Group	144	194*	168*	221	132	215*	172	460
Placebo Group	134	171	118	263	134	176	135	397

* Significantly higher than placebo group.

There were no significant differences between groups in baseline samples at 15 weeks. Both groups exhibited an increase in the concentration and percentage of DHA in maternal RBC and plasma between 15 and 28 weeks, followed by a decrease between 28 weeks and term. However, fish oil-supplemented mothers had concentrations of DHA that were 22% higher in plasma (P 0.02) and 13% higher in RBC (P 0.02) at 28 weeks, and 42% higher in RBC at term (P 0.02) compared with the placebo group. DHA accounted for a similarly higher % TFA in RBC of fish oil-supplemented mothers at 28 weeks (P 0.003) and term (P 0.01). There were no significant differences between groups in DHA as % TFA or concentration in cord blood, placental tissue, cord tissue, or breast-milk (data not shown). In both groups, DHA was higher in cord than in maternal RBC and plasma (% TFA and concentrations); cord levels were most closely related to maternal DHA status at 28 weeks (see Figure).



Maternal DHA status is maximal in mid-trimester and declines to term. The extent of this decline is limited in supplemented compared with unsupplemented mothers. The relationship between mid-trimester maternal and term cord blood DHA suggests that the timing of maternal supplementation is important, and is most likely to be beneficial if it begins before mid-gestation. Maternal DHA supplementation enhances maternal DHA status and may aid preferential transfer of DHA from mother to fetus. Maintenance of a higher DHA status at term may also enhance maternal DHA status in subsequent pregnancies.

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Maternal nutrient intake, educational level, smoking and birth weight in a cohort of pregnant women. By G. McNEILL[1], J. BAKER[1], D.M. CAMPBELL[2], S. JOHNSTON[3], C. BODNER[3] and A. SEATON[3], *Departments of* [1]*Medicine & Therapeutics,* [2]*Obstetrics & Gynaecology and* [3]*Environmental & Occupational Medicine, University of Aberdeen, Foresterhill, Aberdeen AB25 2ZD*

In view of the interest in the relationships between maternal nutrition, birth weight and risk of disease in adult life, we studied a cohort of 2000 pregnant women recruited prospectively from the ante-natal clinic at Aberdeen Maternity Hospital between 1997 and 1999. Weight, height, parity, educational level and socio-economic status (SES) of the mother were recorded at around 12 weeks of pregnancy. The SCG (formerly Aberdeen) Food Frequency Questionnaire (FFQ) was sent to the women at 34 weeks of pregnancy to collect information on diet, supplement use and smoking in the preceding 2–3 months. The validity of this FFQ has been reported elsewhere (Masson *et al.* 2001).

Complete data on diet and delivery were collected for 1597 of the 1958 women who had singleton live births. Mean birth weight was 3524 (range 1500–5180) g for boys and 3399 (range 1480–4940) g for girls. A corrected standardized birth weight score (CSBS) was calculated as the *z*-score for birth weight adjusted for sex, gestational age, parity and maternal height and weight (Campbell *et al.* 1993). Dietary intake and total nutrient intake (diet plus supplements) were calculated for protein, iron, zinc, vitamin A, vitamin C and folate. The percentages of women taking supplements containing these nutrients were 40% for iron, 7% for zinc, 4% for vitamin A, 9% for vitamin C and 19% for folate. Nutrient intakes were log-transformed before analysis, and relationships between variables were assessed by one-way ANOVA or linear regression.

CSBS was not related to SES but was related to smoking and mother's educational level (both $P<0.001$). These variables were considered as possible confounders, since smoking in pregnancy was associated with lower dietary and total vitamin C intake and higher dietary and total vitamin A intake (all $P<0.01$) and higher educational level was associated with higher dietary iron and dietary and total vitamin C and folate intake (all $P<0.01$). The Table shows regression coefficients for the equations predicting CSBS from nutrient intake before and after adjustment for potential confounding variables:

		Protein	Iron	Zinc	Vitamin A	Vitamin C	Folate
Unadjusted	Diet	0.058	0.129	0.072	–0.069	0.150***	0.089
	Total	N/a	0.075***	0.103	–0.031	0.151***	0.110
Adjusted[1]	Diet	0.325*	0.536***	0.310**	–0.105	0.205***	0.277*
	Total	N/a	0.079***	0.252**	–0.034	0.191***	0.148**
Adjusted[2]	Diet	0.219	0.349*	0.200	–0.095	0.138*	0.167
	Total	N/a	0.077***	0.163	–0.039	0.129*	0.096

[1]Adjusted for energy intake; [2] for energy intake, smoking and mother's education. *$P<0.05$, **$P<0.01$, ***$P<0.001$.

In the unadjusted models there were strong positive associations ($P<0.001$) between CSBS and total iron and dietary and total vitamin C intake, which remained after adjustment for energy intake. Dietary iron, dietary and total zinc and total folate intake also showed positive associations ($P<0.01$) with CSBS after adjustment for energy intake. When maternal smoking and education were included in the models, iron and vitamin C intakes were still positively associated with birth weight. The results suggest that iron supplements increase birth weight but that maternal diet does not have a strong influence on birth weight in this population.

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Day-to-day variation in concentrations of iron, zinc and copper in the breast-milk of Guatemalan mothers. By M. VOSSENAAR[1], R.A.M. RUTTEN[2], C.E. WEST[2], K. SCHÜMANN[3], J. BULUX[4] and N.W. SOLOMONS[4], [1]*Centre for Public Health and Nutrition Research, University of Dundee, Dundee DD1 9SY,* [2]*Division of Human Nutrition and Epidemiology, Wageningen University, Wageningen, The Netherlands,* [3]*Walther Straub Institute for Pharmacology and Toxicology, Ludwig-Maximilians-University, Munich, Germany and* [4]*Center for Studies in Sensory Impairment, Aging and Metabolism (CeSSIAM), Guatemala City, Guatemala*

Having accurate data on the concentrations of trace elements in human milk throughout lactation is important for determining the nutritional requirements of infants and for understanding the physiology of milk secretion (Casey *et al*. 1989). It has been shown that the trace-element concentration of human milk varies throughout the day and from day to day. If human milk is to be used as a standard for trace element requirements, it is important to establish the extent of the variation and possible factors influencing it. From this information it will be possible to determine how many milk samples may be considered necessary for a reliable estimate of trace-element concentrations in milk.

Concentrations of iron, zinc and copper in the milk of mothers from Guatemala were measured using inductively-coupled plasma/atomic emission spectrometry (ICP/AES). Lactating women (*n* 56) who had delivered a healthy infant in the previous 1–6 months, living in a low-income, suburban or rural area, participated in this cross-sectional study. Women infested with either *Ascaris lumbricoides* or *Trichuris trichiura* received a single dose of albendazole (400 mg) or a placebo. Two weeks after treatment, milk samples were collected on three or four consecutive sampling days.

Trace element	Concentration (mg/l)	Within-subject variation		Between-subject variation	
		SD	CV (%)	SD	CV (%)
Iron	0.28 (0.06–0.76)	0.13	46.1	0.17	61.2
Zinc	2.03 (0.47–6.19)	0.37	18.2	0.98	48.3
Copper	0.29 (0.09–0.60)	0.07	22.8	0.09	31.7

Mean concentration (range) (based on single analysis of 166 milk samples obtained from 47 mothers on 3 or 4 consecutive days), within-subject and between-subject variation of iron, zinc and copper in milk of mothers.

The instrumental error of the ICP/AES-method, expressed as SD, was 0.04, 0.27 and 0.02 mg/l for iron, zinc and copper, respectively. Concentrations in samples of milk collected from forty-seven mothers on three or four consecutive days, expressed as mean (SD), were 0.28 (0.13), 2.03 (0.37) and 0.29 (0.07) mg/l for iron, zinc and copper, respectively. The within-subject CV was 46.1, 18.2 and 22.8%, and the between-subject CV was 61.2, 48.3 and 31.7% for iron, zinc and copper, respectively. Stage of lactation, presence of intestinal parasites and residential area had a significant influence on milk zinc, copper and iron concentrations.

Element	CV														
	10 %					20 %					30 %				
	Sampling days (*n*)					Sampling days (*n*)					Sampling days (*n*)				
	1	2	3	4	5	1	2	3	4	5	1	2	3	4	5
Iron	58	48	44	42	41	15	12	11	11	10	6	5	5	5	5
Zinc	27	25	24	24	24	7	6	6	6	6	3	3	3	3	3
Copper	15	13	12	11	11	4	3	3	3	3	2	1	1	1	1

Number of subjects required for a reliable estimate of iron, zinc and copper concentration according to number of consecutive sampling days and at varying levels of reliability.

One sample of milk is sufficient to give a reliable estimate of the zinc and copper concentrations in milk; whereas more samples, taken on consecutive days, are needed for a reliable estimate of iron concentration.

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Dietary interventions in general dental practice: formative research for the development of a dietary intervention pack. By K.L. BARTON[1], A.S. ANDERSON[1], C.M. PINE[2] and M.G. PATERSON[2], [1]*Centre for Public Health Nutrition Research, University of Dundee, Ninewells Hospital and Medical School, Dundee DD1 9SY and* [2]*Section of Dental Public Health & Health Psychology, University of Dundee, Dundee DD1 4HR*

Nutrition in the early years is a major determinant of growth and development; it also influences adult health. Young children are solely dependent on their parents or other carers with respect to their diet; it is therefore vital that parents/carers recognize the importance of an appropriate diet to the future well-being of their children (Scottish Office, 1996). If good eating patterns are established in childhood, it is more likely that these will continue into adulthood. The diets of young children need to be relatively more nutrient-dense than those of older children, because requirements for nutrients need be met within relatively small quantities of food (British Medical Association, 1999). Unfortunately, the diets of young children are often more calorie-dense than nutrient-dense, which contributes to the development of obesity and tooth decay. Child dental health in Scotland is amongst the worst in Europe (Bolin, 1997). Annual surveys conducted within Health Boards have shown no overall improvement in the dental health of 5-year old children since surveys began in 1987 (SHBDEP, 2000). Sugar in drinks and foods is the leading cause of Scotland's poor dental health (Scottish Office, 1999) and, whilst the majority of people are aware of this link, many people find restriction or abstention difficult to initiate and maintain.

This study aimed to develop a dietary education pack for use in dental practice to change sugar-related dietary behaviours in pre-school children. Four main stages of development were undertaken prior to pilot testing within general dental practice: (1) a literature review on current dietary behaviours of concern related to caries development; (2) two focus groups of mothers and carers of preschool children to identify contemporary local beliefs and practices related to infant feeding habits; (3) a literature review on effective patient communications; (4) acquisition and critical assessment of the patient materials on diet and caries in children currently used in the UK in relation to current dietary messages, relevant eating patterns (especially bedtime drink consumption) and reading indices.

Published work and responses from the focus groups suggest that the three main dietary behaviours of concern with regard to dental health, which could usefully be addressed, are frequent snacking on sweet foods, consumption of sugar-containing drinks, and bedtime routine. Preliminary work suggested that the intervention pack should be tailored to each behaviour separately, should include information sheets relating to each behaviour for the dental health educator, and should include 'take-home' leaflets for each behaviour to reinforce the verbal advice given.

Following the developmental process, written material was drafted, focusing on frequent snacking on sweet foods, consumption of sugar-containing drinks, and bedtime dietary behaviours, using a range of educational strategies (e.g. highlighting sources of sugar), motivational strategies (e.g. "a happy child with a lovely smile is what every parent wants to see"), and behavioural strategies (e.g. tactics to replace giving high-sugar foods). The drafted materials were pre-piloted with colleagues and a dental health educator to assess comprehension and their suitability as educational tools. Preliminary work suggests that the dietary education pack could usefully be incorporated into a structured programme tailored to meet the specific dietary habits of individual children. The pack will now be pilot-tested for face validity and comprehension with parents of young children.

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A 'Fast Food' school meal service: the contribution to meeting nutritional requirements. By J. ARMSTRONG, L. GOLDBERG and R. HEWITT, *School of Biological and Biomedical Sciences, Glasgow Caledonian University, Cowcaddens Road, Glasgow G4 0BA*

Improving the nutrition of children is fundamental in addressing Scotland's health problems by immediately affecting nutritional status and influencing their health into adulthood (McCormick, 2000). The school environment is one arena where education on food and nutrition can be supported by practical example through the provision of nutritious food. The school meal service is one of the largest public catering providers in the country and in some areas is a highly commercialized system. Food is 'sold' to children through advertising and other marketing techniques, with considerable investment in these activities. The nutritional quality of school meals is variable and there is limited opportunity to share good practice through national co-ordination and dissemination. Regulations on Nutritional Standards for School Lunches in England came into force in April 2001 (Education England, 2000).

Glasgow City Council introduced the catering system 'Fuel Zone' in all secondary schools in 1999 and plan to expand the service to the primary school sector. Although there appear to be clear commercial benefits (an increase in attendance at school meals) there has been no formal evaluation of the nutritional consequences. The aim of this study was to assess the food choices and nutrient intakes of 100 secondary and fifty primary school children within a Fuel Zone system and to evaluate their contribution to the daily nutritional requirements of the children. For each child the food chosen at their school lunch was recorded, leftovers were deducted and portion sizes were measured at the servery. Nutrient intake was estimated using food composition database (coded in Compeat).

Nutrient intake (expressed as % of the DRV provided)	Male pupils Aged 12–14 (*n* 50)		Female pupils Aged 12–14 (*n* 50)		Male pupils Aged 4–11 (*n* 25)		Female pupils Aged 4–11 (*n* 25)		Guideline (% of DRV to be provided by school meal) (Sharp, 1992)
	%	SD	%	SD	%	SD	%	SD	
Energy (E) kJ	25.7	8.5	27.5	7.2	19.3	8.9	26.4	9.8	30% estimated average requirement
% E Carbohydrate	46.8	5.1	53.6	6.6	50.0	12.5	48.0	8.3	Not less than 50% of food energy
% E Sugar	19.5	4.4	20.3	8.6	23.0	5.7	19.0	4.1	No more than 11% of food energy
% E Fat	39.2	8.0	36.2	6.2	36.0	10.5	39.0	7.7	No more than 35% of food energy
% E Saturated fat	16.1	5.2	14.5	4.8	12.0	5.6	11.2	4.5	No more than 11% of food energy
Total protein	52.0	34.1	45.0	25.1	51.5	33.4	61.0	36.6	Not less than 30% of RNI
NSP	11.6	9.1	12.3	6.5	8.8	0.4	13.0	7.5	Not less than 30% of DRV
Calcium	14.7	13.0	17.3	14.4	41.4	24.6	49.6	23.8	Not less than 35% of RNI
Iron	25.1	10.2	14.2	7.1	18.9	21.7	22.7	18.1	Not less than 40% of RNI
Vitamin C	8.5	18.5	12.8	8.2	3.1	16.0	15.0	12.2	Not less than 35% of RNI
Sodium	55.1	22.5	47.5	22.7	66.0	30.0	70.0	32.5	RNI = 30–70mmol
Folate	11.2	6.6	11.2	6.1	17.1	16.3	21.3	13.4	Not less than 40% of RNI

The percentage of children taking one or more portions of fruit or vegetables was 7% in secondary and 20% in primary school. In secondary-school children the meal provided a quarter of daily energy requirements, less than 15% of the RNI for folate, vitamin C and non-starch polysaccharides, and less than 20% of the RNI for Ca. Energy and nutrient intakes were similarly poor in primary-school children. Na intakes were over 45% of the RNI for all ages. This significant nutrient imbalance leaves these children the implausible task of obtaining the large majority of their nutrient requirements from the other foods consumed within the day in order to redress the balance. There is urgent need for a national policy on the provision of food to children in school.

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School meals: an intervention to improve food choices and nutrient intakes. By B. MILLER and J. ARMSTRONG, *School of Biological and Biomedical Sciences, Glasgow Caledonian University, Cowcaddens Road, Glasgow G4 0BA*

The expansion of health promoting schools is identified as a national priority in Scotland (Scottish Executive, 2000). Improving the nutritional status of children through education on food and nutrition and the provision of food in schools is one aspect of this approach. The Scottish Diet Action Plan sets out guidelines for public sector catering (Scottish Office, 1996). However, the variable nutritional quality of food provided in schools is a major problem across Scotland with emphasis in some areas for a quicker meal service with 'fast food'.

Current trends indicate that 45% of Scottish schoolchildren take school meals, with 20% entitled to free school meals. In some areas, such as Glasgow, the proportion of children entitled to free school meals is 42%. School meals make a significant contribution to the total nutrient intake of children and guidelines suggest they should provide 30–40% of their nutrient requirements for the day.

This study aimed to improve nutritional intake by primary-school children at their school meal by altering the food provided in the school canteen. Alterations in the food service focused on the key areas of base ingredients, preparation, presentation and promotion of food. Alterations included introduction of reduced fat mayonaise, withdrawal of fizzy drinks and introduction of fruit juice increasing the content of vegetables in the home made soups and the introduction of a chopped-fruit cup. The food choices and nutrient intakes of forty-six primary-school children aged 9–10 years were measured before and again 2 months into the intervention. Average portion sizes and leftovers were weighed and the results of nutrient intakes compared with guidelines for school meals.

Intakes	PRE- INTERVENTION 46 Pupils Male =13, Female =33 Age 9–10			POST – INTERVENTION 46 Pupils Male =13, Female =33 Age 9–10			Guideline (% of DRV to be provided at meal) (Sharp, 1992)
	Intake	SD	% of DRV	Intake	SD	% of DRV	
Energy (E) (MJ)	M 2.6 F 2.2	0.9 0.5	M 32% F 30%	M 2.4 F 2.1	0.8 0.7	M 29% F 29%	30% of estimated average requirement
% E Carbohydrate	48	10		*54	12		Not less than 50% of food energy
% E Sugars	18	9		**22	13		Not more than 11% of food energy
% E Fat	41	9		†34	11		Not more than 35% of food energy
% E Saturated fat	16	8		†10	6		Not more than 11% of food energy
Protein (g)	15.5	5.4	55%	15.9	7.5	56%	Not less than 30% of RNI
NSP (g)	2.5	1.2	16%	3.0	1.6	19%	Not less than 30% of DRV
Calcium (mmol)	5.5	3.2	40%	4.4	4.2	32%	Not less than 35% of RNI
Iron (μmol)	38	18	23%	43	16	28%	Not less than 40% of RNI
Vitamin C (mg)[a]	3	1–17	10%	†18	8–78	60%	Not less than 35% of RNI
Sodium (mmol)	34	22–44	68%	30	20–41	60%	RNI= 50 mmol (1200mg)
Folate (μg)[a]	40	26–56	27%	**48	35–70	33%	Not less than 40% of RNI

[a]Values given are median and interquartile range. M = Male, F = Female. $^{\dagger}P< 0.001$, $^{*}P<0.01$, $^{**}P< 0.05$.

Comparison of nutrient intakes at baseline and after 2 months indicated no change in energy and protein intakes, a significant reduction in energy from fat and saturated fat, significant increases in energy from carbohydrates and sugars and significant increases in Vitamin C and folate intakes. The proportion consuming any fruit and vegetables increased from 20 to 50%. The number of children consuming one or more portions of fruit and vegetables tripled. The proportion of children consuming carbonated drinks decreased from 45 to 17%. Important supportive factors include the enthusiasm and interest of the pupils, commitment of catering management, motivation of catering staff, and co-operation of teaching staff.

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Results from a school-based nutrition education intervention aimed at increasing fruit and vegetable intake in primary-school aged children. By A.S. ANDERSON[1], A. ADAMSON[3], M.M. HETHERINGTON[2], E. FOSTER[3], L. PORTEOUS[1] and C. HIGGINS[2], [1]*Centre for Public Health Nutrition Research,* [2]*Dept of Psychology, University of Dundee, Dundee DD1 5HT and* [3]*Human Nutrition Research Centre, University of Newcastle upon Tyne, Newcastle upon Tyne NE1 4LP*

A whole-school intervention ("5-a-day the Bash Street way") aimed at increasing consumption of fruits and vegetables in primary-school aged children, was designed and implemented for 511 children in two intervention schools (with a further 464 children from two schools acting as controls) in Dundee, Scotland. The intervention focused on the message of increasing consumption of fruit juice, fruits and vegetables (excluding potatoes) to five portions per day.

The intervention programme provided increased provision of fruits and vegetables in schools (through innovative tuck-shop initiatives and increases in school lunch options), tasting opportunities, a range of point of purchase marketing (posters and quizzes), newsletters for children and parents, and teacher information sessions (delivered in school assemblies, training sessions and classroom presentations). Curriculum materials at age 6 and 11 years (largely focusing on practical food preparation and tasting, promoted through hands-on activities, written work, videos, self monitoring materials and story books) were also developed and utilized.

Evaluation was directed at a 'younger' sample (aged 6–7 years) and an 'older' sample (aged 10–11 years) and included 3-d dietary records with interview and psychological measures at baseline with follow-up at 9 months in intervention and control schools.

Variable	Baseline		T2		Intervention effect *P* value*
	Mean	SE	Mean	SE	
Energy intake (KJ)					
Intervention	7922	207	7926	213	0.327
Control	8268	257	7920	236	
Energy as fat (%)					
Intervention	35.4	0.0065	34.9	0.0056	0.929
Control	36.9	0.0051	36.3	0.0063	
Energy as carbohydrate (%)					
Intervention	51.3	0.0066	51.8	0.0053	0.368
Control	49.8	0.0060	51.2	0.0062	
Energy as protein (%)					
Intervention	13.1	0.0029	13.1	0.0028	0.097
Control	13.0	0.0028	12.2	0.0030	
Intake of starch (g)					
Intervention	128	4.4	131	4.0	0.980
Control	131	4.5	134	4.3	
Intake of sucrose (g)					
Intervention	55.1	17.5	54.6	19.4	0.578
Control	56.7	20.0	52.7	22.7	

* Values shown are the significance of the difference in change in intake from baseline to T2 between the intervention and control groups from a multiple regression model which included age and sex.

Results showed that children in the intervention schools (*n* 64) had an average increase in daily fruit intake of around 0.5 portions per person (from 133±11.9 g to 183±17.0 g per d) which was significantly greater ($P<0.05$) than the increase (from 100±11.7 g to 107±14.2 g per d) estimated in children (*n* 65) in control schools. No other changes in food or nutrient intake were detected. Increases in scores for variables relating to knowledge about fruits and vegetables and subjective norms (Ajzen & Fishbein, 1980) were also greater in intervention compared with control groups. No changes were detected in taste preferences for fruits and vegetables but preferences for snack foods had significantly diminished. It is concluded that, in this small study, a whole-school approach to increasing intakes of fruits and vegetables had a modest but significant effect on cognitive and attitudinal variables and on fruit intake.

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Cystic fibrosis-associated liver disease: a case-controlled study of patients in the Republic of Ireland. By C. CORBETT[1], S. KELLEHER[1], M. ROWLAND[1], G. CANNY[2], P. GREALLY[3], L.E. DALY[4] and B. BOURKE[2], [1]*Department of Paediatrics, Conway Institute, University College Dublin and The Children's Research Centre,* [2]*Our Lady's Hospital for Sick Children, Crumlin, Dublin 12, Republic of Ireland,* [3]*National Children's Hospital, Tallaght, Republic of Ireland and* [4]*Department of Public Health and Epidemiology, University College Dublin, Republic of Ireland*

Cystic fibrosis (CF) is an inherited disease, which affects the function of the exocrine glands in humans. Predominant features of the disease are: recurrent respiratory tract infections (which generally determine outcome), pancreatic insufficiency and liver disease, *inter alia*. It has been observed that patients with CF whose nutritional status is impaired have a poorer outcome than those who are adequately nourished. A small group of CF patients develop liver disease; however, risk factors for the development of liver disease are unknown. The aim of the current study was to determine whether poor nutritional status is a risk factor in the development of CF-associated liver disease (CFALD). For the purposes of this study, CFALD was deemed to be present if there was (i) histopathologic evidence of fibrosis/cirrhosis or (ii) radiologic and/or endoscopic confirmation of portal hypertension.

Forty paediatric patients (twenty-four boys) were identified with CFALD in the Republic of Ireland. They were matched for age and sex with CF patients not suffering from liver disease. Clinical score, X-ray score, forced expiratory volume in one second % predicted (FEV_1 %), and biochemical indicators of current clinical status were recorded. Biochemical and anthropometric indicators of current nutritional status of all patients were collected. Historical weights and heights were available for thirty pairs. All patients with CFALD had significantly elevated circulating liver enzymes. Plasma vitamins A and E were lower in patients with CFALD, whereas plasma vitamin D was not. No difference in clinical score or X-ray score was observed between the groups. A greater proportion of patients with CFALD had an FEV_1 % in the moderate range (40–70%) than their controls ($P<0.01$). Current weight centiles were significantly lower in those children suffering from CFALD. Current height centiles were lower (although not significantly) in the CFALD group. There was no significant difference between paired mid-parental heights and paired sum of skin-fold thickness.

		CFALD		Control		
	n	Median	Range	Median	Range	*P*
Age		14.24	5.6–18.8	14.13	6.96–19.6	NS
Birth weight (kg)	40	3.44	1.55–4.35	3.1	1.19–4.46	$P<0.05$
Height centile at 5years	30	32.5	85–3	45.4	75–3	NS
Weight centile at 5years	30	25	97–3	25	95–3	NS
Height centile at 8–9years	30	30	70–3	52	75–3	$P<0.05$
Weight centile at 8–9years	30	20	97–3	40	97–3	NS
Current height centile	40	20	97–3	30	97–3	NS
Current weight centile	40	15	3–80	30	97–3	$P<0.05$
Sum of skin-folds (mm)	36	30.9	18.2–67.9	33.69	14.8–106.7	NS
Mid parental height (cm)	40	168.3	153.5–187	168.2	160–180.5	NS
Vitamin A (μmol/l)	40	0.9	0.32–2.67	1.4	0.5–2.89	$P<0.05$
Vitamin D (μg/l)	40	14.2	4.1–26.5	14.3	5–26.5	NS
Vitamin E (μmol/l)	40	16.4	2.5–36.1	22	6.1–41.2	$P<.05$

Although at birth the CFALD group were slightly heavier, by 5 years they were on a similar weight centile to controls. By 8–9 years (generally the time of diagnosis of liver disease), there was a trend towards lower weight centile in the CFALD group. At 5 years those with CFALD tended to be shorter than their controls whereas by 8–9 years they were significantly shorter. These data suggest that for some years prior to diagnosis, children who develop liver disease (although not genetically shorter) seem to display a greater degree of weight and height faltering than their controls.

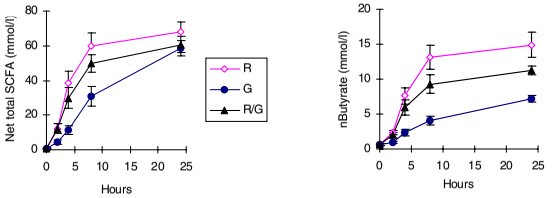
Production of short-chain fatty acids from mixtures of guar gum and Raftilose™ *in vitro*. By M.K. KHAN and C.A. EDWARDS, *Department of Human Nutrition, Glasgow University, Yorkhill Hospitals, Glasgow G3 8SJ*

Most fermentation studies have considered a single source of carbohydrate. It has been shown that the presence of ispaghula affected the fermentation of lactulose *in vitro* (Khan & Edwards, 2001). In contrast to ispaghula, guar gum may lose its viscosity during fermentation and may influence fermentation in a different manner.

The present study was designed to determine the production of short-chain fatty acids (SCFA) from Raftilose™ (R; Raffinerie Tirlemontoise, Brussels, Belgium) and low molecular weight guar gum (G; P30 Meypro Zaandam, Netherlands) alone and in mixture, using 50 mg of each to give a total 100 mg substrate/10 ml, in an *in vitro* model (Adiotomre *et al.* 1990). Cultures were incubated anaerobically (37°) with human faecal slurry (3.2%, *n* 8), in a shaking water bath (50 strokes/min). SCFA were measured in the supernatant fraction at different times (Spiller *et al.* 1980). The results are reported as mean (SE).

Cultures containing R/G mixtures produced significantly more net SCFA than those with G alone ($P<0.02$, *t* test), but not significantly less than R alone at 8 h. Cultures of R/G mixtures produced significantly more *n*-butyrate than G alone ($P<0.02$, *t* test).

The initial rapid SCFA production in R cultures was preserved in R/G mixtures, although these contained only 50 mg R. The bacteria may have rapidly fermented R before the fermentation of G in R/G cultures.



Raftilose™ dominated the fermentation pattern in R/G mixed cultures without a significant reduction in the production of net SCFA and *n*-butyrate by the fermentation of R. The combination of these carbohydrates in mixtures may yield the same benefits as raftilose alone with fewer side-effects from large doses of a single carbohydrate.

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Bioavailability and efficacy of iron supplemented orally as ferric-amino acid chelates in the rat. By A.A. ABDEL-GAYOUM[1], M.M. EL-AJAILY[2] and B.S. BAIO[2], [1]*Department of Biochemistry, Faculty of Medicine;*[2]*Department of Chemistry, Faculty of Science, University of Garyounis, Benghazi, Libya*

A simple and effective way of improving iron status of a deficient individual over a short period of time is through supplementation with iron tablets. Tablets containing 60 mg elemental iron as ferrous salts are commonly used in several countries. Previous studies have shown poor compliance of the targeted individuals with the iron supplementation, mainly because of the negative side effects. Iron supplements, which are usually in the form of ferrous salts, have been shown to be toxic to the gastrointestinal mucosa, and iron supplemented as ferric-maltol chelates were found to be less toxic, with a lower incidence of negative side-effects to patients intolerant of ferrous salts (Harvey *et al.* 1998). In the present study we investigated whether iron supplemented orally in the form of ferric-amino acid chelates would have comparable bioavailability and efficiency in the rat compared with the ferrous formate supplement.

Ferric-alanine, ferric-aspartate and ferric-histidine chelates were prepared in our laboratory. Ferrous formate tablets were purchased from a pharmacy. Weighed amounts of finely ground powders of the three chelates and the Fe-formate tablets were suspended in physiological saline under vortex mixing. Five groups of adult female Sprague-Dawley rats, each comprising seven animals, were used in the experiment. Animals in three groups were supplemented measured volumes of the ferric-amino acid chelate suspensions, by gastric intubation, to give 0.5 mg elemental iron/kg body weight per d for ten consecutive days. Animals in the fourth group were given the ferrous formate suspension to supplement a similar amount of iron for 10 d. The control group received a similar volume of saline for the same period. All animals were killed and serum and whole blood were collected and used for the assay of serum iron concentration and the haematological measurements using commercial kits.

	C		Fe-Formate		Fe-Ala		Fe-Asp		Fe-His	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Serum iron (μmol/l)	7.85	(1.37)	31.67	$(2.05)^{a***}$	36.72	$(2.61)^{a***}$	33.13	$(3)^{a***c*}$	31.7	$(2.8)^{a***c*}$
Haemoglobin (g/l)	88.43	(11.1)	111.57	$(9.5)^{a*}$	129.0	$(17.7)^{a**}$	113.0	$(14.3)^{a*}$	108.0	$(13.2)^{a*c*}$
Transferrin (Sat.%)	18.3	(2.7)	37.11	$(1.46)^{a***}$	39.1	$(2.98)^{a***}$	37.37	$(1.7)^{a***}$	36.78	$(2.7)^{a***}$
RBC count ($\times 10^{-6}$)	5.21	(0.64)	6.54	$(0.6)^{a*}$	7.12	$(0.86)^{a**}$	6.35	$(1.05)^{a*}$	6.25	(0.64)

[a]Significantly different from C; [c]significantly different from Fe-Ala. **P*<0.05; ***P*<0.01; ****P*<0.001.

The results revealed that rats given Fe-alanine had their serum iron levels raised 3.7-fold over that of control and by 15.9% compared with ferrous formate-treated animals. Animals supplemented with iron in the form of Fe-Asp or Fe-His were not significantly different from those given ferrous formate. The haematological parameters were significantly improved in animals supplemented with the ferric-amino acid chelates, with Fe-Ala showing the greatest effect. The results indicate that in the rat oral iron supplementation as ferric-alanine complex may have a better bioavailability and is probably more efficient in improving the haematological parameters than the conventional ferrous formate. Supplementation with ferric chelate may avoid the risk of negative side effects.

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Seasonal differences in vitamin D status in elderly people. By J.P. VYVYAN, B. O'HANRAHAN, H.S. PEACE and G. McNEILL, *Department of Medicine and Therapeutics, University of Aberdeen, Aberdeen AB25 2ZD*

Low vitamin D (Vit D) status is often reported in elderly people. Vit D is obtained largely from exposure of the skin to ultraviolet (UV) irradiation in sunlight. The ability to synthesise Vit D declines with age, particularly if people become less mobile. There is no UV radiation of the appropriate wavelength (290–310 nm) in the UK from the end of October until the end of March. For other months of the year 60% of effective UV radiation occurs between the hours of 11 a.m. and 3 p.m., and this is lowest in the north of the UK (DoH, 1991). Seasonal differences in Vit D status were seen in adults over 65 years in the National Diet and Nutrition Survey (Finch *et al.* 1998).

This study examined the prevalence of Vit D deficiency amongst elderly people living in the community in the city of Aberdeen (58°N) and hypothesised that biochemical levels of Vit D would be lower in the winter months than during the summer months. A sample of men and women aged 75 years and over were randomly selected from the Community Health Index. These participants were initially identified as part of a larger study of micronutrient deficiency in the elderly. A fasting blood sample was taken for analysis of 25-hydroxy Vit D (25-OH Vit D). The method of analysis used was a diasorin radio-immune system and all assays were performed in duplicate. Participants who were seen between June and September 1999 had a repeat blood sample taken in February or March 2000. Details of Vit D supplements taken and whether subjects had taken a holiday in the sun during the previous 6 months were recorded on both occasions. Low Vit D status was defined as 25-OH Vit D <25 nmol/l. The data were log-transformed to correct for skewness and a paired *t*-test was used to test differences between the two seasons. χ^2 tests were used to determine the association of Vit D supplements, or of taking a holiday in a sunny place, with low Vit D status.

Fifty-five participants completed this study (response rate 72%). Thirty-one (56%) were male (median age 79, range 75–93 years) and twenty-four (44%) were female (median age 84, range 75–90 years). During the summer period, 35% reported taking a sunny holiday compared with 22% in the winter; 26% took Vit D supplements in the summer and 40% in the winter. The present study identified 26% of the subsample as deficient during the summer and 51% during the winter, as compared with 33% in the 398 subjects in the original study. The median values of 25-OH Vit D during the summer and winter months were 31 nmol/l and 24 nmol/l, respectively. For men, the median values (ranges) were 38 nmol/l (19–78) for the summer months and 26 nmol/l (12–92) for the winter months; for women, these values were 27 nmol/l (9–45) and 21 nmol/l (7–67), respectively. There was a significant difference between the seasons in 25-OH Vit D ($P<0.001$), but there was no significant difference in the amounts of supplement taken in summer and in winter. There was no association between 25-OH Vit D levels and supplement intake in the summer months but a significant association was found during the winter months ($P<0.001$). No association was found between Vit D status and taking a sunny holiday in either season.

In conclusion, 25-OH Vit D levels were found to be significantly lower during the winter months and there was also a significant association between Vit D supplementation and biochemical level in winter. Vit D supplementation is an important consideration for elderly people living in the north-east of Scotland, particularly for women and especially during the winter months.

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