# Seasonal variations of total lipid and n-3 polyunsaturated fatty acid contents in two Victorian farmed abalone species

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The total lipid and n-3 polyunsaturated fatty acid (PUFA) contents of two farmed abalone species, blacklip (Haliotis rubra, BL) and greenlip (Haliotis laevigata, GL) collected from Port Phillip Bay, Victoria, were investigated through spring (SP), summer (SU) and autumn (AU) seasons. No seasonal profiles of total lipid content and n-3 PUFA level have been previously published for farmed abalone in this region. The total lipid content in both species varied significantly through the seasons (P < 0.01) with SU samples having the highest level (2.7% wet muscle weight in BL and 2.5% in GL) and AU samples having the lowest level (0.9% in BL and 0.8% in GL). The total lipid content was relatively low in SP with 1.2% in both BL and GL.

There were eight major fatty acids in both species, namely 16:0, 17:1, 18:0, 18:1, 18:2, 20:5n-3 (EPA), 22:5n-3 (DPA) and 20:4n-6 (AA). The predominant fatty acid in n-3 PUFA series was DPA in both species with the mean  $\pm$  S.D. concentration of  $40.7 \pm 9.9$  mg/100g wet muscle in BL (n = 31) and  $57.4 \pm 12.1$  mg/100g in GL (n = 32). The level of EPA was slightly lower than that of DPA in BL with the mean concentration of  $35.6 \pm 6.9$  mg/100g, while in GL the mean concentration was only  $29.5 \pm 6.5$  mg/100g. In both species the concentration of 22:6n-3 (DHA) was low through the three seasons with the mean of  $6.7 \pm 2.3$  mg/100g in BL and 7.1 ± 4.1 mg/100g in GL. The major n-6 PUFA were AA and 18:2 with the mean concentration of AA being  $22.1 \pm 5.0$  mg/100g in BL and  $16.8 \pm 5.2$  mg/100g in GL. The concentration of DPA was higher, and that of AA was lower, than that reported previously in Tasmanian nonfarmed BL (1).

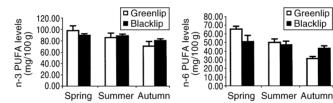


Figure 1. Seasonal variations of n-3 PUFA level

Figure 2. Seasonal variations of n-6 PUFA level

□ Greenlin

■ Blacklip

Figures 1 and 2 above show the seasonal variations of n-3 and n-6 PUFA levels in two abalone species. In GL, both n-3 and n-6 levels varied significantly (P < 0.05) with the total n-3 PUFA level increasing from 69.7mg/100g in AU to 98.0mg/100g in SP. The total n-6 PUFA level in GL ranged from 30.6mg/100g in AU to 65.2mg/100g in SP. Although there were similar variations on both n-3 and n-6 PUFA levels in BL through the three seasons, these variations are not statistically significant (P > 0.05).

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# The n-3 polyunsaturated fatty acid content of commonly available green vegetables in Australia

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Diet has long been considered to play a critical role in human health, with green vegetable consumption being claimed to have health benefits mainly due to the vitamins, minerals and phytonutrients (such as vitamin C, folate, antioxidants etc). Additionally green vegetables are known to contain a relatively high proportion of omega-3 polyunsaturated fatty acid in the form of  $\alpha$ -linolenic acid (18:3n-3). However, there are no data available on fatty acid composition and concentration of the commonly consumed green vegetables in Australia.

The present study determined fatty acid content in eleven commonly available green vegetables in Australia: spinach (Spinacea oleracea), watercress (Nasturtium officinale), parsley (Petrolelinum crispum), Chinese cabbage (Brassica chinensis), brussel sprouts (Brassica oleracea var. gemmifera), bok choy (Brassica chinensis), cos lettuce (Lactuca sativa), broccoli (Brassica oleracea), Chinese broccoli (Brassica alboglabra), baby bok choy (Brassica chinensis) and mint (Mentha viridis, M. spicata, M. Crispa). For all samples in this study, only the leaves or heads were analysed which contain the chloroplasts whereas the stems do not appear to contain many chloroplasts since they usually had little green colour. Prior to analysis the samples were blotted to remove adhering moisture and then the leaves or heads were chopped and blended. To determine any variation in lipid content, six sub-samples each weighing approximately 10 g were analysed for each of eleven vegetables. Lipid was extracted with 50.0 mL of methanol-chloroform (2:1 v/v) containing 10 mg/L of butylated hydroxytoluene and 0.2 mg/mL of tricosanoic acid (C23:0) as an internal standard. The fatty acid methyl esters of the total lipid extract were prepared by saponification of using KOH (0.68 mol/L in methanol) followed by transesterification in BF<sub>3</sub> in methanol. Fatty acids were identified by comparison with standard mixtures of fatty acid methyl esters and the results were calculated using respone factors derived from chromatograph standards of known composition. Silver ion TLC was used to identify any peaks on the GC traces that could not be identified using the standards.

Total fatty acid concentration of 11 green vegetables ranged from 44 mg/100g wet weight in Chinese cabbage to 372 mg/100g in watercress. There were three polyunsaturated fatty acids in all vegetables analysed 16:3n-3, 18:2n-6 and 18:3n-3. Green vegetables contained a significant quantity of 16:3n-3 and 18:3n-3, ranging from 23 to 225 mg/100g. Watercress and mint contained highest 16:3n-3 and 18:3n-3, and parsley had a highest 18:2n-6 in both percentage composition and concentration. Mint had a highest concentration of 18:3n-3 with a value of 195 mg/100g, while watercress contained a highest concentration of 16:3n-3 with 45 mg/100g. All eleven analyzed green vegetables contained a high proportion of PUFA, ranging from 59 to 72% of total fatty acids. The omega-3 PUFA composition in 11 analyzed vegetables ranged from 40 to 62% of total fatty acids. Monounsaturated fatty acid composition of the 11 analyzed vegetables was less than 6% of total fatty acids. The proportion of saturated fatty acid ranged from 21% in watercress and mint to 32% of total fatty acids in brussel sprouts. No eicosapentaenoic and docosahexaenoic acids were detected in any of the samples in the present study. Consumption of green vegetables would contribute to the 18:3n-3 PUFA intake, especially for vegetarian populations. The data obtained could contribute to the Australian food composition database to provide information for further research and to the general public.

# Biobalanced livestock feeding systems

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There are five major problems causing waste in intensive livestock production systems: (a) the high energy cost of milling, mixing and pelleting feeds; (b) environmental pollution from excess nutrients in manure; (c) overuse of high protein feeds; (d) the belief that vitamin and mineral and other supplements must be fed; and (e) the cost of changing to more welfare friendly systems.

A model was developed and its components tested in a series of feeding trials with laying hens.

# (c). Adding enzymes (2, 3) (b). Formulation of diets to minimum specifications (2) (a). Free choice feeding (1) (e). Utilise natural environment (nutrient, exercise, immunity) (d). Cheaper feed, reduce pollution (c). Release bound nutrients

### BIOBALANCED LIVESTOCK FEEDING SYSTEM

The results show that feed costs and wasted nutrients can be reduced by careful formulation of diets that meet the nutrient requirements of the animal but do not contain mineral, vitamin, amino acid or yolk pigment supplements. These diets also help to decrease pollution (6).

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# Progress in the elimination of iodine deficiency as a cause of brain damage by the year 2000

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Iodine deficiency is now recognised to be the most common preventable cause of brain damage in the world today with a global population of 2 billion at risk (WHO).

Since 1990 a global programme has proceeded with remarkable momentum for the elimination of iodine deficiency disorders (IDD) as a cause of brain damage by the year 2000 using the technology of universal salt iodization (USI) by the addition of iodine (20–40 mg/kilo) as potassium iodate to all salt for human and animal consumption.(1)

Following an earlier Nutrition Society lecture (1991) progress is now reported with special attention to the significant role of the International Council for Control of Iodine deficiency Disorders (ICCIDD), an international NGO founded in 1986 which now comprises an international multidisciplinary network of 600 professionals from 100 countries with a majority from developing countries first supported by Australia (AusAID) followed by UNICEF & WHO.

From its foundation the ICCIDD accepted technical assistance to national programmes as the first priority. This led to a close working relationship with the leading agencies WHO and UNICEF and with governments of countries with significant IDD public health problems. Subsequently the salt industry has been involved in a global partnership together with a World Service Club, Kiwanis International, which has raised US\$50 million for UNICEF for national IDD control programmes.

A WHO/UNICEF/ICCIDD Report (1999) revealed that of the 130 IDD affected countries, 105 had National Intersectoral Coordinating Bodies, 102 had Plans of Action for IDD control and 98 had Legislation in place. Of 5 billion people living in countries with IDD, 68% had access to iodized salt through universal salt iodization (USI).

A spectacular example of progress is provided by the People's Republic of China.

National Monitoring Results	1995	1997	1999
The proportion of households consuming iodized salt 20 mg/kg (%)	39.9	81.1	88.9
Urinary iodine level in children aged 8–10 median (ug/L)	164.8	330.2	306.0
Total goitre rate (%) by palpation	20.4	10.9	8.8
The production of iodized salt (10,000 tons)	539	620	753

Further progress can be anticipated, but sustainability requires regular monitoring of salt iodine and urine iodine. Salt iodine levels should be in the range of 20–40 mg of iodine per kilo and median urine iodine should be in the range of 100–200 ug/L. Levels below 200 ug/L are necessary to minimise the occurrence of transient iodine induced hyperthyroidism (IIH) in the iodine deficient population. Increase in iodine intake (200 ug) is required in pregnancy to provide for the needs of the growing fetus. Recent data from Sydney and Tasmania indicates that iodine deficiency has recurred in Australia.(2)

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# Raw brown onion consumption reduces plasma triglycerides and has other health benefits

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Compounds in garlic and onions have been implicated as providing putative health benefits, such as reducing the risk of coronary heart disease and atherosclerosis (1). However, the effects of different onion varieties and level of intake have not been studied. The aim of the present study was to evaluate the potential health benefits of two onion varieties fed at two levels of intake, using the pig as a human model.

Twenty-five female (Large White x Landrace) pigs (initial weight  $41.5 \pm 4.23$  kg) were used in a  $2 \times 2+1$  factorial designed experiment. The treatments consisted of a white onion (WO) and brown onion (BO) fed at 10 or 24 g/MJ DE and no onion, respectively. Onion varieties were selected on the basis of the level of cysteine-sulfoxides, WO being low and BO high. The WO and BO varieties were grown in Queensland and Tasmania, respectively. Onions were homogenised in a blender prior to being mixed with dry feed formulated to contain 16.7 MJ DE/kg and 10% (w/w) of tallow to simulate the saturated fatty acid content of a western human diet. Pigs were fed approximately 90-95% of *ad-libitum* (1.67 MJ DE/kg<sup>0.75</sup>) for 6 weeks. Blood samples were obtained by venipuncture immediately before feeding at weeks 1, 2, 4 and 6 and at three hours post-feeding at weeks 4 and 6. Plasma or serum, were analysed for total cholesterol (TC), HDL-cholesterol, LDL-cholesterol, triglycerides (TG), clotting factors such as prothrombin (PT), activated prothrombin (APPT) and thromboxane B<sub>2</sub> (TXB<sub>2</sub>) and cell counts which included the ratio of segmented neutrophils to lymphocytes (N:L).

	TC (mmol/L)	HDL (mmol/L)	LDL (mmol/L)	TG (mmol/L)	Platelets (10 <sup>9</sup> /L)	PT (sec)	APTT (sec)	TXB <sup>2</sup> (ng/mL)	N:L
Control	2.70	1.07	1.80	0.56a	428	13.8	21.5	23.59a	0.81a
WO	2.62	0.94	1.71	$0.61^{b}$	467	14.0	21.8	$29.44^{b}$	0.64  ab
BO	2.46	0.96	1.64	$0.44^{c}$	392	14.3	23.3	23.01a	$0.49^{b}$
LSD	0.291	0.123	0.217	0.149	95.34	0.423	1.795	5.612	0.252

Superscript letters a, b and c indicate significance (P < 0.05) within column.

BO was more effective than WO onions in lowering blood TC (9%, P = 0.059), LDL (10%, P = 0.13) and TG concentrations (21%, P = 0.082). BO reduced TC in a dose dependent manner (linear relationship P = 0.028). Pigs fed BO tended (P < 0.10) to have higher PT and APPT times whereas these variables were unaffected in pigs fed WO. Concentrations of  $TXB_2$  were higher in pig's fed WO onion, but were unaffected in pigs consuming BO onions. Serum fibrinogen and platelet counts were similar across all treatments. The N:L, an indicator of stress intensity, was significantly reduced in pigs fed the BO onions. There was significant difference between weeks (P < 0.05) and between pre-feeding and post-feeding (P < 0.05) for most variables, except for platelet count and cholesterol fractions (data not shown). In conclusion, the consumption of brown onions is effective in lowering plasma lipid levels and increasing clotting time in pigs.

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# Monosodium glutamate and asthma – what is the evidence?

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The flavour enhancer, monosodium glutamate (MSG) was first implicated in causing adverse reactions in people with asthma in 1981, when two doctors wrote a letter to the New England Journal of Medicine proposing a possible association between MSG and asthma.

Since this time seven clinical trials to determine the relationship between MSG and asthma have been conducted throughout the world. Two of these trials have shown an association between MSG and asthma (1,2). However five trials, involving 45 subjects with a positive history of MSG-induced asthma, have shown no such association (3–7). A further trial, which assessed a range of food chemicals in adults with asthma, demonstrated MSG-induced asthma in one out of the eight subjects studied (8).

Attempts to clarify this issue have been limited due to methodological deficiencies, including the small number of subjects studied, inadequate blinding procedures, inappropriate withdrawal of asthma medications, poor dietary control and the use of effort-dependent measures of lung function. After reviewing the evidence that is currently available, it would appear that a causal connection between MSG and asthma has not been conclusively established.

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# Time course of incorporation of 1-14C-α-linolenic acid into various rat tissues

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In a previous study we showed that an oral dose of  $1^{-14}\text{C}$ - $\alpha$ -linolenic acid is found in skin and fur of guinea pigs 48 hours after dosing (1). The aim of this study was to determine the time course of labeling of various tissues in rats following an intraperitoneal dose of  $1^{-14}\text{C}$ - $\alpha$ -linolenic acid. Twenty, 3-wk old, male Sprague-Dawley rats were each given 1.85 mCi of  $1^{-14}\text{C}$ - $\alpha$ -linolenic acid (mixed in olive oil) by intraperitoneal injection. Rats were then sacrificed 5, 10, 25, 50 hours after the dose (n = 5 for each time point). The dpm and concentration of omega-3 polyunsaturated fatty acids (PUFA) were determined by scintillation counting and gas liquid chromatography, respectively.

The tissues with the highest specific activity (dpm/mg omega 3 ) were the liver, spleen, kidney, fur and lung. The fur label declined over time starting from being high at 5 hours, which might indicate possible contamination from the intraperitoneal dose. However, the specific activity stabilized over the next 45 hours which might point to  $^{14}$ C-labelled  $\alpha$ -linolenic acid being deposited onto the fur. The maximum specific activity was different between tissues, maximum specific activity was at 10 hours for the liver, epididiymis and heart, while the label did not reach a maximum for the testis, skin (head) and brain areas (cerebellum, basal forbrain and cortex) over the period examined. Analysis by silver nitrate TLC at 25 hours time point showed that the main fractions containing  $^{14}$ C were the 6 double bond fraction for all tissues, except for epididymis and adipose where it was in the 3 double bond fraction, the skin and fur where it was in the 3 and 6 double bond fraction and the carcass where it was in the 3, 5, and 6 double bond fractions. These data are in contrast to the guinea pig where after 48 hours of dosing, almost no  $^{14}$ C from labelled linolenic acid was found in the 5 or 6 double bond fractions.

In this study, different tissues followed a different time course with regard to the uptake and metabolism of the  $^{14}$ C-labelled  $\alpha$ -linolenic acid. The finding that the epididymis had a relatively high specific activity is novel and may indicate an important function for this essential nutrient. The labelling of fur support the findings previously reported (1), however it is still possible that the fur could have been contaminated from the intraperitoneal injection site.

Based on the results in this experiment, it is possible to speculate that  $\alpha$ -linolenic acid may have a function in relation to fur, perhaps as a secreted lipid from sebaceous glands to protect the fur from damage by water, light or other agents. This speculation is consistent with the use of linoleic acid in dogs to maintain their coats in good condition (2).

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# Blood pressure and dietary sodium reduction in normotensive subjects

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Intervention studies with sodium supplementation in hypertensive and mildly hypertensive subjects support the hypothesis that higher sodium intake is associated with higher blood pressure. However, reductions in blood pressure in normotensive subjects with reduced sodium intake, within the usual range of sodium intake, has not been consistently demonstrated.

The aim of this study was to determine the effect of alterations of dietary sodium (Na) intake on blood pressure (BP) in normotensive subjects. Twins and family members were recruited to a double-blind, randomised crossover design where all subjects followed a low sodium diet (LS) (50mmol/day) for 8 weeks. Subjects took a placebo for 4 weeks and Na supplement (NaSup) for 4 weeks. All subjects provided one 24hr blood pressure measurement (AMBP) at baseline and at the end of each phase and 2, 24-hr urine collections. Home blood pressure measurements were conducted daily in the last week of each intervention phase.

One-hundred-and eight individuals (57 females, 33 males (mean age 45.1(8.9)(SD) years, not taking antihypertensive therapy commenced the study. Of these 89 completed the study (10 dropped out due to side affects from tablets, 9 from inability to comply with study demands). At baseline the mean AMBP was  $122.4 \pm 1.0$  ( $\pm$  SEM) mmHg systolic (SBP)/75.6  $\pm$  0.9 mmHg diastolic (DBP) and home BP was  $117.8 \pm 1.6$ /  $73.5 \pm 1.0$ , and the mean urinary sodium excretion  $138.0 \pm 6.0$  mmol/day. Na excretion with NaSup was similar to baseline (Na  $137 \pm 4.2$  mmol/day) and Na excretion was lower during LS phase  $51.6 \pm 4.3$  mmol/day P < 0.001). Home systolic BP was lower in LS phase  $114.7 \pm 1.3$  mmHg versus  $116.3 \pm 1.3$  with Na Sup (P < 0.05). There was no difference in AMBP between LS ( $119.4 \pm 1.3/73.9 \pm 0.8$  mmHg) and the NaSup phase ( $119.4 \pm 1.3/73.9 \pm 0.8$  mmHg). Na excretion was positively associated with 24-hr SBP at baseline ( $119.4 \pm 1.3/73.9 \pm 0.06$ ). Na excretion was phase ( $119.4 \pm 1.3/73.9 \pm 0.06$ ).

SBP measured at home was 1.6 mmHg lower on a low sodium diet (50mmol/day) compared to a usual sodium intake (137 mmol/day). This effect was not seen with 24hr blood pressure measurement. This small reduction in blood pressure, seen in normotensive subjects within the normal range of sodium intake indicates the potential health benefits of a low sodium diet. These results are in agreement with the recent US study(1) which demonstrated a graded blood pressure reduction with a low sodium diet of 67 mmol/day in normotensive and hypertensive subjects.

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# Sodium intake and self-efficacy

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High sodium intakes contribute significantly to the development of cardiovascular disease, and Australian intakes are substantially above recommended levels (1). In developing strategies to encourage reduced intakes, it is useful to compare the characteristics of those who have lower and higher Na intakes, especially characteristics that are potentially modifiable by education/counselling.

One such characteristic is self-efficacy, a person's confidence that they could perform certain behaviours if they so chose. Self-efficacy is not generic, but needs to be evaluated in relation to specific behaviours. A nine-item instrument has been developed (2) to measure self-efficacy for reducing salt intakes. It assesses the subject's confidence that they could persist with certain low-salt dietary habits (eg, buy fewer high-salt snacks, keep the salt shaker off the table, eat low-salt cereals) if they decided to. Possible scores range from 9 (minimal confidence) to 63 (maximal confidence).

As part of a study on Na intakes on 194 Hobart adults (87 males, 107 females, ages 20–69 years), we asked participants to complete the salt self-efficacy instrument and also assessed their Na intakes from 24h urinary Na excretion. Data were noticeably skewed, necessitating use of non-parametric statistical methods.

Among women, the median salt self-efficacy score was 60, and the median Na intake was 112 mmol/day. The two showed a Spearman coefficient of -0.27 (P = 0.005). Median Na intakes were 121 mmol/day for subjects in the lowest quartile of self-efficacy scores (i.e., < = 54), and 94 mmol/day in the highest quartile (i.e, a score of 63).

Among men, the median salt self-efficacy score was 54, and the median Na intake was 172 mmol/day. The two showed a Spearman coefficient of -0.19 (P = 0.09). Median Na intakes were 189 mmol/day for subjects in the lowest quartile of self-efficacy scores (< = 54), and 151 mmol/d in the highest quartile (> = 58).

We conclude that greater salt self-efficacy is linked to lower Na intakes. Further study is needed to assess whether intervention programs aimed at increasing salt self-efficacy would help patients lower their Na intakes, but our results suggest that such interventions might potentially lower Na intake by up to 30–40 mmol/day.

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# Fat distribution and blood pressure: a twin study

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Recent research has emphasized the importance of central abdominal fat as a predictor of cardiovascular disease. Furthermore, systolic (SBP) and diastolic (DBP) blood pressure may be differently associated with fat distribution (1).

This cross-sectional study was undertaken to explore the relationship between body composition and blood pressure in a sample of 22 males, 48 females (44 monozygous (MZ), 17 dizygous (DZ) twins and 9 family members who had participated in a dietary study. The mean age was 45.8 (8.9)(SD) yrs, BMI: 25.2 (4.0) kg/m<sup>2</sup>) and only those not taking anti-hypertensive therapy were included.

	S	BP	DBP	BP
	$\mathbb{R}^2$	β (se)	$\mathbb{R}^2$	$\beta$ (se)
BMI (kg/m <sup>2</sup> )	0.10**	0.75 (0.36)	0.11**	0.82 (0.23)
Total fat (g)	0.001	0.001 (0.000)	0.004*	0.001 (0.000)
Abdominal fat (g)	0.06*	0.004 (0.002)	0.06*	0.003 (0.001)

<sup>\*</sup>P < 0.05 \*\* P < 0.01.

Blood pressure measurements, using a mercury sphygmomanometer, were taken 4 times after 5 minutes seated. Body composition was determined by a Lunar DPX-L X-ray densitometer. The relationship of body composition to blood pressure (BP) (age-adjusted) was assessed using univariate regression. In the sub-group of same sex twin pairs (22 MZ, 9 DZ pairs), the within twin pair difference in body composition was assessed in relation to the within pair difference in BP (regression through the origin).

The within twin pair difference in abdominal fat was associated with the within pair difference in DBP 0.004(0.002) ( $\beta(s.e)(P < 0.05)$ ). The within pair difference in BMI and total fat was not associated with within pair difference in SBP or DBP.

BMI and abdominal fat were associated with both SBP and DBP crossectionally, however within twin pairs only abdominal fat was positively associated with DBP. These associations are evident within a group of adults who are not hypertensive and agree with a recent study, which found that body fatness, especially central abdominal fat is associated with DBP in normotensive middle-aged men and women (2).

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# Effects of the glycemic index on the insulin-like growth factor system

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Increased intake of refined carbohydrates has been associated with secular increases in height, weight and growth in groups such as the Eskimo (1). We hypothesised that acute postprandial hyperinsulineamia following the consumption high GI foods may cause changes in the insulin-like growth factor system that favour accelerated growth. Insulin-like growth factor-1 (IGF-1) is an important stimulator of growth and metabolism, and insulin-like growth factor binding protein-1 (IGFBP-1) is suppressed by acute and chronic elevation in insulin (2).

Two groups of young, lean, healthy subjects, 10 Caucasians and 10 South East Asian, were studied. The mean ( $\pm$  SD) age and BMI were 24  $\pm$  4 y and 21  $\pm$  2 respectively. They fasted overnight and consumed a low and high GI meal (50 g carbohydrate portions of pearled barley or instant potato respectively) in random order on separate occasions. On a third occasion they fasted over the same period. Finger prick blood samples were taken at regular intervals over four hours and analysed for glucose, insulin, free IGF, total IGF and IGFBP-1-3.

In all twenty subjects, IFGBP-1 levels were significantly decreased by 4 h post consumption of the low GI food ( $-44 \pm 17$  ng/mL) compared with little change after the high GI food ( $0 \pm 16$  ng/mL). However, in Caucasians, there were significantly greater increases in IGFBP-3 4 h after consumption of the low GI compared with the high GI food ( $0.3 \pm 0.1$  vs  $0.1 \pm 0.1$  µ/mL, p < 0.05). No significant differences were found in serum IGFBP-2, free IGF-1 or total IGF- 1 levels in response to the two foods.

We also noted interesting racial differences during the extended fast. In SE Asian subjects, mean fasting levels of free IGF-1 over the 4 h were significantly higher than in Caucasian subjects (0.9  $\pm$  0.01 vs 0.7  $\pm$  0.02 ng/mL). Correspondingly, mean IFGBP-1 levels were lowest in SE Asian subjects (40  $\pm$  3 vs 96  $\pm$  5 ng/mL, p < 0.01). Fasting glucose levels were higher in the SE Asian groups (5.4  $\pm$  0.1 vs 5.1  $\pm$  0.03 mM, respectively, p < 0.01).

These results provide equivocal support for the hypothesis that the ingestion of high GI foods leads to alteration in the IGF system that collectively favours increased growth. Changes in IGFBP-3 were remarkable and unexpected and may indicate increased free IGF-1 available in the tissues. Changes in IGFBP-1, however, were the opposite of those hypothesised, suggesting that a low GI food would promote higher free IGF-1 levels. Racial differences in the glucose metabolism and the IGF system during extended fasting may be relevant to the documented differences in the prevalence of type 2 diabetes.

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# The acute effect of resistant starch on postprandial satiety in an overweight population

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**Introduction:** Obesity and overweight are now common health problems all over the world. Successful maintenance of long-term weight loss is of considerable benefit in this group as it lowers the risk of certain chronic diseases. Diets high in resistant starch may be a suitable strategy for weight loss, since foods high in resistant starch have a reduced digestibility in the small intestine. This slower rate of starch digestion and gastric emptying may have a positive effect on satiety sensation. The aim of this study was to compare in an overweight population the postprandial satiety ratings in response to meals containing high or minimal levels of resistant starch.

**Method:** Nine males and 10 females aged  $42.4 \pm 13.2$  y with a mean body mass index (BMI, in kg/m²) of  $29.5 \pm 3.39$  were recruited. Subjects consumed two main meals (breakfast and lunch) containing either high-amylose resistant starch (R) or non-resistant starch (N) on two separate days in a crossover design in a comfortable laboratory setting. The N meal challenge day contained a negligible amount of resistant starch, while the R meals contained 9.53 g and 15.21 g of high-amylose resistant starch in breakfast and lunch respectively. Subjective satiety ratings were obtained by using visual analogue scales. Measurements were taken at 60-minute intervals for 10 consecutive measures on each day. Results were expressed as mean  $\pm$  SD. Comparison of means was performed by Student's t test and statistical significance was set as P < 0.05.

**Results and Discussion:** The satiety ratings from both meal-types showed an early increase after the two test meals followed by a subsequent gradual decrease and remained above the basal values 4-h postprandially. Minor differences in satiety ratings were found after the two test meals, with a significant difference occurring immediately following the lunch meal (P < 0.05). Mean satiety scores were slightly higher for subjects fed the R meal compared with those fed the N meal but these were not statistically significant. Higher intakes of resistant starch may be needed to show any effects. In addition, overweight individuals may be relatively insensitive to the satiety response (1), suggesting the need for further research in this area.



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# Dietary vitamin E modulates immune responses to Salmonella typhimurium in chickens

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Supplementation of poultry diets with Vitamin E (VE) can enhance the immune response and improve resistance to disease. In chickens VE supplementation has stimulated increased macrophage phagocytosis and increased production of immunoglobulin G (IgG) and IgM (1,2). However, the effect of dietary VE on IgA antibody, which acts as the first line of defence of the intestinal mucosa, has not been evaluated. Recent work by the authors identified increased IgA antibody production at the intestinal site in birds immunised with tetanus toxoid and receiving diets supplemented with VE (3). The present study was designed to determine whether improved antigen-specific IgA antibody production could be stimulated in birds receiving VE supplemented diets and immunised with killed *Salmonella typhimurium*, which commonly colonises the chicken through the intestinal mucosa and, poses a serious public health risk.

From the day of hatch chicks were placed on a maize-based diet containing 50 mg VE/kg which was supplemented with either 100, 250, 2500 or 5000 mg VE [BASF, Lutavit E 50 Special]/kg. At day 21 all chickens were intraperitoneally immunised with killed whole *Salmonella typhimurium* in a vegetable oil based adjuvant. Two weeks later they received an oral booster of killed whole *Salmonella typhimurium*. Samples of serum, intestinal scrapings (IS) and bile were collected on the end of the experiment, day 42, and *Salmonella typhimurium* specific IgA antibody titres were determined by enzyme-linked immunosorbent assay (4).

On day 42 birds receiving 250 mg VE supplementation /kg had significantly higher mean anti-S. typhimurium IgA antibody titres in serum (P < 0.05) and IS (P < 0.02) and, notably higher *anti-S*. typhimurium IgA titres in bile, compared to birds receiving the basal diet. Birds receiving 2500 mg VE supplementation /kg had a significant increase (P < 0.04) in serum anti-S. typhimurium IgA antibody, but there was no notable alteration in the IgA antibody titre in either the IS or bile.

These results demonstrate the capacity for vitamin E supplementation of poultry diets to enhance the immune response in chickens and, in particular, anti-Salmonella typhimurium IgA antibody titres at the intestinal mucosa following immunisation with killed whole Salmonella typhimurium. The potential for vitamin E supplementation to enhance the immune response when included in the diet for periods less than 42 days is being investigated.

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# Effect of monounsaturated fat in the diet on the serum lycopene levels

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Epidemiological data suggest that populations with higher serum/tissue levels of carotenoids have a lower risk of coronary heart disease (CHD) (1,2). Lycopene, a carotenoid mainly found in tomatoes, has been suggested to have the greatest antioxidant capacity of the carotenoids found in fruits and vegetables. Carotenoids are fat-soluble compounds and their absorption from the diet into the body may depend on the amount of dietary fat ingested. For years there has been debate about what energy source should replace the saturated fat in the diet, to give the optimum serum lipid profile to reduce CHD risk. Studies have investigated the effect of different amounts of total fat on the serum levels of carotenoids especially  $\beta$ -carotene and lutein, but to our knowledge no study has looked at the effect of different amounts of fats in the diet on the serum lycopene levels.

A randomised crossover dietary intervention study, partially funded by Grains Research and Development Corporation, Canberra, Australia and Meadow Lea Food Ltd, Mascot, Australia was conducted in 13 healthy men aged 20 to 70 years. The aim of the study was to compare the effects of monounsaturated fat enriched (MUFA) diet (38% of energy from fat) and high carbohydrate low fat (HCLF) diet (15% energy from fat) with controlled lycopene content, on serum lycopene levels. Main sources of lycopene in the diet were tomato paste and tomato soup (donated by Heinz Watties, Melbourne, Australia). The lycopene content of the diet was 20.3 mg/day. The diets were designed to be low in other carotenoids. The diets were of 14 days duration with a washout period of six weeks. Before the start of the two dietary periods, subjects were asked to take low carotenoid diet (LCD) for two days to avoid the acute peaks in serum lycopene levels which may occur with a high intake of lycopene rich food 10-12 hrs before the blood sample (3).

Compared to baseline (after two days of LCD) serum *trans*, *cis* and total lycopene levels increased after the MUFA and HCLF diet periods. There was no significant difference in *trans*, *cis* and total lycopene levels at the end of two diets. This study indicated that 38% of energy from fat in the diet compared to 15% of energy from fat with a modest amount of lycopene in the diet has no differential effect on the serum lycopene levels.

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# An overview of gene-nutrient interactions

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We are near the end of human structural genetics. The Millennium draft sequence identifies over 90% of the 3 billion base pairs of DNA carried in every cell: the total assemblage of genes, or *genome*. International partners in the Human Genome Project are now working to eliminate gaps and ambiguities, to produce a 'gold standard' sequence by 2003. The genome sequence will be an immensely valuable resource, and its high publicity has produced a revolution in nomenclature: omes are the new isms and ologies. Nutritionists, who thought they were studying metabolism or physiology, are now told that the basic information of the genome turns nutrients and their metabolites into living cells or organs (the *metabolome*) which, in turn, are integrated into a living human being (the *phenome* or *physiome*).

Actually, the structural genome by itself does not tell all that much. We need functional genomics to tell us what gene products do: and which may not be obvious from their sequence. Also, the genome is the same in all cells but the subset of genes expressed is not. Much more needs to be known about the control of transcription, whereby the information encoded in our genes is copied onto messenger RNA (mRNA), forming the transcriptome (the complete set of mRNA). One method of control is DNA imprinting by methylation; the methylome is the complete set of DNA methylations in a cell type. After transcription, mRNA changes before translation to proteins can take place. Non-coding regions (introns) are removed from between the coding regions (exons) by splicing. Often, the same initial transcript can be spliced in many different ways (the current record, for a neuroprotein gene, is about 50,000 permutations). Editing of mRNA can sometimes remove a base encoded by DNA, and replace it by another. Thus, the final members of the transcriptome are not simple copies of the genome. Also, topping and tailing of the end regions of mRNAs influences rates of protein translation. Proteins, once translated, can be cleaved or have their constituent amino-acids significantly modified. The complete set of protein molecules in a cell (the proteome), therefore, is at least an order of magnitude greater than the complete set of genes (about 30,000). International proteomic consortia are already in place, but technical problems (proteins will not form convenient paired strands, as nucleic acids do) will ensure that progress is much slower than with genomics.

Genomics and proteomics are 'big science'. Nutritionists can intelligently choose small important genetic items to generate hypothesis-led research. Of course, as non-reductionist scientists, we have much experience in elucidating aspects of the metabolome and physiome, with cell cultures, animal models and nutritional interventions in humans. But now we can maximise our research potential by a systematic, genome-up approach to the study of nutrition.

Indeed, it could be argued that nutrition is not at the edge of the gene, but rather is centre stage. Dietary components make substantial contributions to the stability of DNA, can affect the regulation of gene expression, and may have roles in genetic imprinting; the methylome is a function of folate status. Moreover, nutritional requirements are influenced by variability within the genome; mostly by single nucleotide polymorphisms (alternative bases), which occur about once every 1000 base pairs. These are defined as polymorphic variants or alleles (alternative forms of a gene) when they occur in at least 1% in the population. Such mutations close to or within a gene may influence the amount, structure and function of the gene product; this in turn can influence nutritional requirements, and susceptibility to degenerative disease.

The objectives of this overview are to delineate some of the complex each-way gene-nutrient interactions, to provide examples of nutrients that are involved in such interactions, and to show some of the opportunities available to nutritionists to advance the discipline of nutrition in the post-genomic era.

# Folate, gene expression and genomic stability

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Folate deficiency has been known to be a cause of megaloblastic anaemia since 1930. However, in the past two decades it has become evident that various methylated/reduced forms of this vitamin play a key role in DNA metabolism, specifically maintenance methylation of CpG sequences and in the synthesis of thymine, one of the four bases in DNA. Inadequate maintenance methylation of CpG has important consequences which include (a) altered methylation of CpG islands which impacts on gene expression, (b) altered structure of centromeric DNA leading to chromosome loss and aneuploidy during cell division and (c) expression of parasitic (viral) DNA sequences. These events lead to important changes in the phenotype of cells and are an initiating step in cancer. The other important role of folate (5,10-methylenetetrahydrofolate) is the synthesis of dTTP (deoxythyminetriphosphate) from dUMP (deoxyuracilmonophosphate). This reaction is important because folate deficiency increases the dUTP/dTTP ratio which results in uracil being incorporated into DNA instead of thymine. Uracil in DNA is highly mutagenic, and the cell dedicates four of eight known human DNA repair glycosylases to remove this base. Incorporation of uracil in DNA leads to excessive excision repair sites and the subsequent formation of DNA double stranded breaks which are similar to the DNA lesions caused by ionising radiation. The formation of DNA double stranded breaks leads to chromosome breakage, chromosome rearrangement and gene amplification, important events in the initiation and progression of cancer. The capacity to utilise folate is dependent on dietary intake and also on polymorphisms that affect the activity of proteins/ enzymes involved in the deconjugation, conjugation, reduction, methylation and receptor transport. When vitamin B12 is oxidised or concentration is low, activity of methionine synthase is reduced, lowering SAM concentration and trapping folate as 5-methyltetrahydrofolate making folate unavailable for synthesis of dTMP and methylation of DNA. In view of the above we have dedicated our research efforts in defining the optimal concentration of folate and vitamin B12 for minimising DNA damage in human cells. To date our research suggests that chromosome damage in human lymphocytes in vivo is minimised when RBC concentration of folate exceeds 600 nmol/L, plasma B12 exceeds 300 pmol/L and plasma homocysteine is less than 7.5umol/L (1). These concentrations are achievable at above RDA intake of folic acid (700ug) and vitamin B12 (7ug) (1). In vitro studies suggest that the optimal concentration of folic acid in medium for minimising DNA damage is in excess of 60nmol/L which is greater than the normal range of folate concentration in human plasma (15-40 nmol/L) (2). These studies suggest that current RDAs for folate and vitamin B12 may not be adequate for minimising DNA mutation. This has led to the concept that RDAs should be designed to minimise DNA damage rate because genomic instability is a causative factor in degenerative diseases such as cancer, Alzheimer's disease and accelerated ageing (3).

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# Nutrition as an evolutionary force

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Until recently, evolutionary biologists have not been much interested in nutrition. Although food ultimately provides the energy for survival growth and reproduction, many physiologists have been frustrated by the difficulty of defining exactly what contributes to variance in nutritional quality of different diets. Instead they have focussed on other aspects of animal performance such as thermal and locomotory capabilities that are easier to define and measure.

However, many animals face prolonged periods without food and the realization of the extraordinary and rapid responses of the gut in some species have resulted in a renewed emphasis on how the gut of wild species has evolved to match fluctuating food supplies, energetic demands and different food types that are the of wild vertebrates.

The most spectacular changes in gut anatomy and physiology have been observed in large sit and wait predators such as pythons. In those species the absorptive capacity of the gut can double within hours of ingesting a meal along with up-regulation of metabolic rate, digestive enzymes, nutrient transporters and other organs such as kidneys. However other snakes that feed more regularly don't show the same magnitude of regulatory response. Many other species such as migratory birds must up- or down-regulate their nutrient uptake capacity to meet requirements for activities and changing food availability.

These findings illustrate that maintenance of gut tissues is a large energetic and nutritional burden on animals and one that is borne only when there is food to process. But if the gut is so flexible when food is not available how does it respond when greater quantities of food must be processed? Other major evolutionary questions have asked whether the gut ever sets the ultimate limit to animal activity. Studies of animals in cold and those lactating at maximal capacity show rapid enhanced capacity of the gut to digest and transport nutrients to meet higher energy requirements. Peripheral processes (e.g. capacity of the mammary glands to make milk) have been argued to limit energy expenditure by animals under these conditions rather than the capacity of the gut to transport nutrients.

In wild species, periods of under-nutrition provide the best means of observing selection on nutritional status in wild species. However, there are surprisingly few examples of the fitness consequences of variation in foraging traits, despite their importance for survival of wild species. The best example is in Soay sheep where broader incisor width is strongly selected during population crashes, as is resistance to parasites. Individual variation in parasite resistance is in turn under genetic control but there is much uncertainty about the mechanisms and stability of these selective forces.

Overall, our ability to ask specific questions about the evolutionary impact of nutrition in wild species is hampered by a lack of understanding of what constitutes nutritional quality. There are many examples of wild animals with restricted or unusual diets (e.g. toxic constituents) but understanding how the interaction between consumer and diet has evolved is challenging.

# Refugee camps – a food security, livelihood and childhood nutrition assessment at Sinje, Cape Mount County, Liberia, West Africa

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Introdution/Objectives – Liberia and Sierra Leone are two West African countries ravaged by civil war and rebel takeovers. The human cost of more than a decade of killings, civil unrest, and associated miseries is huge and especially vulnerable are the women and children. WFP (World Food Program) are responsible for food aid to most refuge camps in Africa, and inherent in their rationing is the expectation that after a few months, refuges will cultivate, work or barter for some of their food. Sinje, is an established Liberian border refugee camp for 17,000 refugees from Sierra Leone (6,000 settled more than 9 months – Camp 1). WPF wished to reduce the food ration for 11,000 recently arrived refugees (Camp 2) to that of Camp 1 (4,400 kJ/day/person) which is 4000 kJ/day/person below WFP/UNHCR recommendations. Young children are at greatest risk of malnutrition and their rates are a marker for food insecurity. Many refugees have endured months of forced relocation due to unrest, before arriving at Sinje. The aim of this study was to determine the impact of the recent arrivals on food security and livelihood and childhood malnutrition and assess the effect that the proposed ration reduction may have on Camp 2.

**Design** – Cross sectional study involving systematic sampling of 318 households (eating from the same pot), utilisation of the food ration (bulgur wheat, pulses and oil), access to other sources of food and income. Anthropometric measurements of every child (6 months to 5 years) in the household, and rations supplied were recorded. Structured interviews were conducted by Liberian nationals predominantly trained and employed by Save the Children UK (responsible for monitoring food security in this region). Open ended questions concerning family savings, assets and wages, as well as intentions of returning to Sierra Leone were also posed.

**Outcomes** – Camp 1 refugees were food insecure if relying just on their rations (< 4400 kJ/person daily), but most households had developed livelihood strategies to gain more food, whilst Camp 2 was food secure on the current ration of 8800 kJ/person daily, but with few livelihood opportunities to supplement their ration in the long term. Other measures were not significantly different.

	Camp 1	Camp 2	
Number of households sampled	112	206	
Female headed households	47%	46%	
Childhood malnutrition – global	4.4%	2.7%	
Stunting in children (high: > 30%, WHO)	38%	39%	

Conclusions and Recommendations – Female headed households are particularly vulnerable to food insecurity, and most likely to resort to distress strategies eg sale of meagre assets and high interest loans. Refugees need job creation, mini marketing, and micro-banks. Farming as a livelihood is unattractive, as most refugees want to return home and the huge influx has reduced availability of land. Stunting rates, a result of long term food insecurity, may be reduced by improving other livelihood opportunities. Reduction in ration in Camp 2, whilst important to promote self reliance, should be postponed until other livelihood strategies are in place.

# Consumption of bangun-bangun leaves (*Coleus amboinicus* Lour) to increase breast milk production among Batakneese women in North Sumatra Island, Indonesia

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Bangun-bangun leaf (*Coleus amboinicus* Lour; CA) is an herb that is traditionally consumed by Batakneese women in North Sumatra Island Indonesia whilst nursing. Batakneese women believe that this herb can stimulate the production of their breast milk. The present study aimed to gather information about the beliefs and experiences of Batakneese women in consuming this herb, using a focus group discussion method.

Sixty Batakneese women, who used CA leaves whilst nursing, were invited to participate in focus group discussions conducted in three villages of Simalungun District in North Sumatra Island, Indonesia. One half of the participants were recent mothers (aged 35–51 yr) and the other were elderly mothers (aged 51–91 yr). Each discussion group consisted of 6–12 participants, either recent or elderly mothers, and was moderated by midwives (ND, SS) from the district hospitals. Topics included the knowledge about CA leaves and experience in consuming CA leaves. The duration of each discussion was about 60–90 minutes and it was recorded audio-visually.

'Bangun-bangun' is the name given by Batakneese people, especially in Simalungun, for the *Coleus amboinicus* Lour plant. In the Simalungun language, 'bangun' means 'wake-up'. Traditionally, women who have just given childbirth are given this plant in order to recover. It is believed that delivery upsets the balance achieved during pregnancy and brings about weakness. A special diet of bangun-bangun soup, considered nourishing, is given to the mother 'in order to return her to a state of balance'. The diet is also intended to ensure that the mother can take care of the newborn properly, especially by breastfeeding. All participants considered the effects of consuming bangun-bangun soup during their nursing period to have been beneficial. In general, the women felt fit (not tired but, rather, fresh) and healthy after consuming CA leaves. They felt their breasts become full with breast milk. Moreover, most participants found that consuming CA leaves helped control postpartum bleeding and 'acted as a uterine cleansing agent'.

All participants commenced CA consumption on the second day after giving birth, and most of them consumed a bowl of bangun-bangun soup three times a day for 30–40 days, whilst others did so for only 14–21 days. To make the soup more delicious, slices of chicken meat or fish are added. According to the elderly mothers, there was no restriction to or required frequency with which to consume this soup. The husband or the mother or mother-in-law usually cooks the soup at home. They obtain the CA leaves from their home garden or the local market.

The focus group discussions indicated that Batakneese people consider that the consumption of CA leaves can stimulate the production of the breast milk whilst nursing. CA leaves may be consumed at any time and as much as possible without known adverse effects.

# Compliance with the dietary regimen in a five-year trial of the primary prevention of asthma

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The Childhood Asthma Prevention Study is a randomised controlled trial designed to measure the effectiveness of house dust mite allergen reduction and supplementation with omega-3 fatty acids, both separately and in combination, for the primary prevention of atopy and asthma (1). Poor compliance may compromise the successful outcome and validity of CAPS results.

Pregnant women whose unborn children were at high risk of developing asthma due to family history were randomised prenatally into active and placebo groups (n = 616). The active dietary intervention requires mothers to add tuna oil capsules to the infant's food from six months, and use Canola margarine and oil. The placebo diet involves the use of Sunola oil capsules, polyunsaturated margarine and sunflower oil. Data are collected quarterly in the first year and then half yearly until five years. Compliance is assessed by self-rating (all visits) and plasma phospholipids at 18 months. Data are currently available for 251 children (41% of total).

This study sought to 1) assess differences in plasma phospholipids between active and placebo groups, as a biomarker for compliance with the dietary regimen 2) evaluate the validity of self-reported compliance compared with plasma phospholipids.

The active group had significantly higher plasma omega-3 fatty acid levels and significantly lower omega –6 fatty acid levels than the placebo group.

Fatty acid	Active (n = 125) mean (%) (95%CI)	Placebo (n = 126) mean (%) (95%CI)	P value	
Total omega-3	7.07 (6.71-7.42)	5.05 (4.83-5.26)	< 0.001	
Total omega-6	32.7 (32.20-33.24)	35.21 (34.69-35.72)	< 0.001	

Self-reported compliance was related to plasma phospholipids in the expected direction, that is, omega-3 fatty acids were significantly higher among self-rated good compliers with capsule taking than in self-rated poor compliers (P < 0.001). Nearly half of the subjects were correctly classified into tertiles of plasma omega-3 fatty acids according to self-rated compliance. Only 12.5% were grossly misclassified (Kappa = 0.18).

Significant differences between plasma fatty acids in the intervention groups reflected high compliance with the dietary regimen of CAPS. Self-reported compliance was significantly associated with plasma fatty acids. However, self-reported compliance was not an accurate basis for classification into tertiles of omega-3 fatty acids.

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# Calculating resting energy expenditure in men with HIV/AIDS

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Previous research investigating the role of resting energy expenditure (REE) in the aetiology of metabolic abnormalities and weight loss in HIV has been conflicting (1). This conflict in the literature may be a result of inadequate adjustment for body composition as the fat free mass is the primary determinant of REE and abnormalities of body composition. Both wasting of the fat free and fat mass ('lipodystrophy') are common in people with HIV/AIDS.

The aims of this cross sectional study were to:

- 1. To determine if resting energy expenditure accounting for fat free mass (FFM) is elevated in HIV positive males when compared with healthy controls in the era of highly active antiretroviral therapy.
- 2. To examine the accuracy of prediction equations for estimating REE in people with HIV.
- 3. To determine if REE accounting for FFM is significantly different between those HIV positive subjects reporting lipodystrophy (LD), weight loss (≥ 5%) and those who are weight stable when compared with controls.

This research was conducted in both a tertiary referral hospital HIV unit and an outpatient clinic specialising in HIV care. Seventy HIV positive males were recruited and the results compared with those from sixteen healthy age matched male control subjects.

REE was measured after an overnight fast using indirect calorimetry (Deltatrac II metabolic monitor, Helsinki, Finland). Body composition was assessed using bioelectrical impedance analysis (SEAC BIM 4, Uniquest, Brisbane).

The main findings were:

- 1. REE when adjusted for FFM using regression residuals was greater in HIV positive subjects than controls (1735  $\pm$  194 kcal n = 70 vs 1581  $\pm$  166 kcal n = 16, P < 0.05).
- 2. The Harris Benedict, Schofield and Cunningham equations significantly underestimated REE in the HIV positive subjects when compared with controls and the two equations published by Melchior and colleagues in HIV positive patients overestimated REE. Therefore a new prediction equation was developed. The accuracy of the published equations to predict REE differed in the different HIV positive subgroups which reflects the heterogeneity in body composition.
- 3. When divided into subgroups REE adjusted for FFM was significantly greater in the weight stable HIV patients (n = 23,  $113 \pm 13$  kJ/kg) than the healthy controls (n = 17,  $100 \pm 11$  kJ/kg, P < 0.05). The differences for the groups with lipodystrophy (n = 30,  $109 \pm 12$  kJ/kg) and weight loss (n = 17,  $106 \pm 11$  kJ/kg) were not significant.

In conclusion, REE is significantly higher in HIV positive males when compared with healthy controls. Body composition abnormalities common in HIV make the use of standard prediction equations for estimating REE invalid.

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### Malnutrition in children with cancer in Pakistan

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**Background:** Malnutrition is common among the majority of cancer patients, including children with cancer. In underdeveloped countries such as Pakistan, various degrees of undernutrition are prevalent in the normal population. Malnutrition is recognised to have profound effects on tolerance of anti-cancer therapy, survivability and treatment outcomes. In previous studies, malnutrition was shown to be a negative prognostic factor at Shaukat Khanum Cancer hospital.

**Method:** Two hundred and fifty children admitted to the hospital were assessed for nutritional status. Both anthropometric and biochemical parameters were used as the basis for assessment Patients were further classified into Grade-1 (mildly malnourished), Grade-II (moderately malnourished) and Grade-III (severely malnourished) on the basis of weight for age using the physical growth scales of the National Centre for Health Statistics (1).

**Results:** Of the 250 children, only 17% were well-nourished and 83% were malnourished to some degree. Of those who were malnourished, 19% were mildly malnourished, 29% were moderately malnourished and 35% were severely malnourished. Using biochemical parameters, 71% patients were hypoalbuminemic.

**Conclusion:** Malnutrition is prevalent in children with cancer in Pakistan. Pre-existing malnutrition in the community may be partly responsible. Serum albumin appears to be potentially useful in assessing malnutrition in these patients. Malnutrition will adversely affect treatment outcome, quality of life and increase mortality and morbidity. Aggressive nutrition therapy to correct nutritional status should therefore be initiated as early as possible.

1. National Centre for Health Statistics. Growth of healthy children. Am J Clin Nutr 1979; 32: 607-629.

### Food insecurity in Somali women living in Australia

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Migration and economic transition are associated with dietary change. Australia accepts both migrants and refugees from developing countries. Paradoxically these entrants may be vulnerable to both obesity and food insecurity. The current study aimed to assess changes in food habits, physical activity and body weight in Somali women who have come to Australia as refugees. The sample recruited was a convenience sample of 46. The method was a questionnaire administered by a bilingual interviewer in the subject's home. Twenty-four hour dietary recall was assessed with confirmation of portion size using models. Usual intake in both Australia and Somalia were assessed with a food frequency questionnaire (picture and photo) (1). Weight and height were measured using a portable scale and stadiometer.

The women had an average (SD) age of 35.9 (11.5) years. The majority were married (55%). The women had spent on average 2 years in Australia, they had spent at least 4 years in transit from refugee camps. Eight percent of the sample had a tertiary education, 25% had no formal education, 26% primary only and 35% had completed high school. The mean BMI was 27.4 (5.4) kg/m² (range 18.3–43.4). Fifty-four percent of the sample had a BMI > 25. Seventeen percent stated that they had lost weight since arrival in Australia, 38% reported that they had gained weight and 43% that they had maintained weight.

The mean (SD) energy intake was 4431 (1509) kJ, protein intake was 46.9 (21.2) g. Mean intakes of the micronutrients; iron 6.27 (2.9) mg, folate142 (69.9) ug and zinc 6.2 (3.1) mg fell below the Australian RDI. Using a EI/BMI < 1.5 (2), 44 subjects could be classified as under-reporting dietary intake. But was this undereating or under-reporting? It is possible that the women, influenced by cultural norms of slimness in Australia or not wishing to be stimatised, under-reported. The alternate explanation is under-eating, particularly as 60% reported either losing weight or staying the same. Undereating may be due to deprivation mentality influenced by the refugee experience. It could also be that the intakes reflect food insecurity. Refugees and newly arrived migrants may have low incomes and financial family obligations in their countries of origin. The women may curtail their intakes to provide more food for their families. These findings suggest that migrants and refugees are a population vulnerable to food insecurity.

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# Supplement usage in women entering the menopausal transition in Brisbane

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There is an increasing number of supplements on the market which purport to assist in the relief of menopausal symptoms, with over 38 different products readily available in Brisbane. Yet little published data are available on the extent of use. These supplements include non-prescription medications, herbal therapies and nutritional supplements designed to complement inadequate dietary intake or provide preventative or therapeutic benefits. This study focussed on the extent and pattern of usage of supplements purported to assist with the menopausal transition. Reasons for usage were also assessed.

Subjects were women participating in The Longitudinal Study on Ageing in Women (LAW), a multi-disciplinary study being undertaken in SE Brisbane, with 500 women aged 40–80 yr randomly selected from the electoral roll. Data on reported supplement usage over the previous 6 mo were collected by interview as part of year 1 baseline assessment of overall dietary and supplemental intake of phytoestrogens. Results are presented for the cohort of 158 women aged 40–55 yr who were likely to be entering the menopausal transition.

The overall prevalence of usage of one or more supplements was 58%: 36% reported using vitamins, 27% used herbal therapies (excluding menopausal supplements), 24% used minerals, 6% used supplements for premenstrual symptoms and 11% used supplements for relief of menopausal symptoms, with a significant increase in the older group aged 50–55 yr (P < 0.05). Of the women who reported taking supplements for menopausal symptoms, products included Evening Primrose oil (42%), phytoestrogens (38%) eg soy, isoflavones, Red Clover or linseed, and herbal preparations (20%) eg Chinese herbs or wild yam. Usage of supplements according to age group is summarised in the table.

Reported use of supplements for premenstrual symptoms or menopausal transition	40-44  yr (n = 21)	45-49  yr $(n = 61)$	50-55  yr $(n = 76)$	40-55  yr (n = 158)
No supplements	85.7%	85.2%	78.9%	82.3%
Premenstrual symptoms	9.5%	8.2%	3.9%	6.3%
Menopausal symptoms	4.7%	6.5%	$17.1\%^{1}$	11.4%

<sup>&</sup>lt;sup>1</sup>significantly higher than corresponding values in other age cohorts, P < 0.05.

The results of this population-based study indicate that 11.4% of women aged 40–55 yr reported taking supplements for relief of menopausal symptoms over the previous 6 mo. Preliminary analyses in the same cohort indicate that the prevalence of use of hormone replacement therapy (HRT) was 10–20%. Therefore it appears that about 70 to 80% of women were not taking either prescribed or non-prescribed medications. This cannot be attributed entirely to lack of need, as studies indicate that up to 80% of Western women experience adverse menopausal symptoms (1). Whether our findings reflect an absence of severe symptoms or use of alternative strategies, remains to be clarified. Possible concerns regarding the safety of HRT or uncertainty about the efficacy of herbal or nutritional supplements may also partly explain the observed discrepancy.

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# An investigation into the association between eating environment and food intake of residents with dementia at an aged care facility

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**Objective** – Anecdotal reports have suggested that small dining rooms improve behavior at meal times in those with dementia and that aromatherapy can have a calming influence. This study aimed to determine whether the type of dining area (canteen style versus domestic style) or the use of aromatherapy influenced food consumption of residents with dementia in an aged care facility.

**Design** – Food consumption of residents with dementia was measured in a large, canteen style dining area over 15 days at lunch and dinner and compared to food consumption of residents with dementia in a small, domestic style dining area. Due to the nature of the research and the need for minimal disruption to residents, individual intakes were unable to be measured. Intakes were estimated from total food offered to the groups minus food returned. Aromatherapy was also used in the small room and food consumption measured for a further 15 days at lunch and dinner. Analysis of variance was used to determine the effect of meal, room and aromatherapy on mean intakes and mean proportion eaten per person. Descriptive statistics were used for reporting frequency of consumption of individual meals and likes and dislikes.

**Outcomes** – Ten residents participated in the study, nine female and one male. In the first stage six residents were present in the small room and four were present in the large room. In the second stage (aromatherapy) there was five residents present in the small room.

	Mean total daily intake (g) per person (SD)	Lunch	Dinner	Significance (lunch vs dinner)
Large room	494 (101.0)	246 (70.5)	244 (56.3)	NS
Small room	448 (98.3)	240 (67.3)	209 (46.4)	0.02
Small room with aromatherapy	488 (58.0)	261 (61.5)	228 (37.7)	0.02
Significance aromatherapy	NS	NS	NS	

The mean intake of residents in the small room was significantly more (P = 0.02) at lunch (240 g  $\pm$  67.3) than dinner (209 g  $\pm$  46.4), and this occurred whether or not aromatherapy was used. Mean intakes were slightly higher at lunch (21g more per person) and dinner (19 g more per person) when aromatherapy was used, but did not reach significance. Examining the proportion of food eaten revealed that even though total daily intake (lunch plus dinner) increased in the small room with aromatherapy (74.6% versus 71.2% of total food offered), this was not significant. A significantly (P = 0.011) greater proportion was eaten in the large room compared to the small room (78.1% versus 71.2% of total food offered).

**Conclusions** – Residents with dementia ate a larger amount at lunch than dinner and those residents in the large room ate more when compared to those in small room. Aromatherapy did not alter food consumption in residents with dementia. Further research with a larger sample size may modify this conclusion.

# Depression in malnourished children with cancer

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**Background:** Cancer is considered as the most feared of all the diseases. The stress of dealing with an illness like cancer can cause many uncomfortable feelings such as depression. Malnutrition and depression show close relationship with each other. Depression is closely associated with malnutrition.

**Objective:** To assess depression in malnourished cancer patients.

Setting: Shaukat Khanum Memorial Cancer Hospital and Research Center, Lahore-Pakistan.

**Method:** The sample of 46 admitted paediatric cancer patients in pediatric oncology ward of Shaukat Khanum Memorial Cancer Hospital and Research Centre. Thirty-six males and 10 females were assessed by a trained clinical psychologist and clinical nutritionist by using psychological assessment form. Nutrition assessment of children were based on weight for age with the help of growth charts (1).

**Results:** Of 46 malnourished pediatric cancer patients 37% (n = 17) were depressed. Malnourished patients were categorised into three categories on the basis of anthrometry, mildly malnourished, moderately and severely malnourished. The incidence of depression in mildly malnourished patients was 4% (n = 2), moderately and severely malnourished patients was 13% (n = 6) and 17% (n = 8) respectively.

**Conclusion:** This study shows that the depression is closely linked with the day by day deteriorating nutritional status in children with cancer.

1. Physical growth: National Centre for Health Statistics percentiles. Am J Clin Nutr 1979; 32: 607–629.

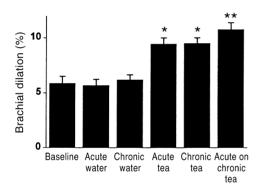
# Short and long term black tea consumption reverses endothelial dysfunction in patients with coronary artery disease

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Epidemiological studies suggest that tea consumption decreases cardiovascular risk, but the mechanisms of benefit remain undefined. Endothelial dysfunction has been associated with coronary artery disease (1). Some antioxidants have been shown to reverse endothelial dysfunction (2) and tea contains antioxidant flavonoids.

To test the hypothesis that tea consumption will reverse endothelial dysfunction, we randomized 66 patients with proven coronary artery disease to consume black tea and water in a cross over design. Short-term effects were examined two hours after consumption of 450 mL of tea or water. Long terms effects were examined after consumption of 900 mL tea or water daily for four weeks. Vasomotor function of the brachial artery was examined at baseline and after each intervention with vascular ultrasound. Fifty patients completed the protocol and had technically suitable ultrasound measurements. Both short and long term tea consumption improved endothelial-dependent flow-mediated dilation of the brachial artery, whereas consumption of water had no effect (P < 0.0001 by repeated-measures ANOVA). Tea consumption had no effect on endothelium-independent nitroglycerin-induced dilation. An equivalent oral dose of caffeine (200 mg) had no short-term effect on flow-mediated dilation. Plasma flavonoids increased after short- and long-term tea consumption.



In 50 patients with coronary artery disease, beverage consumption significantly affected flow-mediated dilation (P < 0.001). Post hoc analysis demonstrated that flow-mediated dilation was higher after short and long term tea consumption versus baseline and water consumption (\*P < 0.001). Furthermore, short on long term tea ingestion resulted in additional improvement (\*\*P = 0.02).

In conclusion, acute and chronic tea consumption reverses endothelial vasomotor dysfunction in patients with coronary artery disease. This finding may partly explain the association between tea intake and decreased cardiovascular disease events.

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# Regular ingestion of tea does not inhibit in vivo lipid peroxidation in humans

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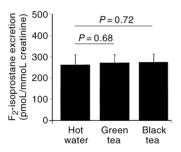
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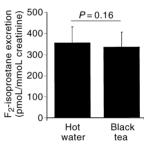
Results of prospective studies suggest that tea may protect against cardiovascular disease. A potential mechanism for such an effect involves inhibition of lipid peroxidation by polyphenolic antioxidants derived from tea. Our objective was to determine if regular ingestion of tea could inhibit  $in\ vivo$  lipid peroxidation. Two controlled intervention studies assessed the effects of regular ingestion of tea on lipid peroxidation determined by measurement of urinary  $F_2$ -isoprostane excretion, which is currently regarded as one of the best available markers of  $in\ vivo$  lipid peroxidation.

Study 1. The effects of five cups/day of green tea and black tea were compared to hot water containing the same concentration of caffeine in 13 otherwise healthy subjects with raised blood pressure using a randomised three-period (seven days each) crossover study.  $F_2$ -isoprostane excretion was not altered following regular ingestion of green tea (273  $\pm$  48 pmol/mmol creatinine) or black tea (274  $\pm$  39 pmol/mmol creatinine) in comparison to hot water (263  $\pm$  47 pmol/mmol creatinine) [Figure 1].

Study 2. The effects of five cups per/day of black tea were compared to hot water in 22 otherwise healthy subjects with mildly raised serum total cholesterol concentrations using a randomised two-period (four weeks each) crossover study.  $F_2$ -isoprostane excretion was not altered by regular ingestion of black tea (334  $\pm$  71 pmol/mmol creatinine) in comparison to hot water (355  $\pm$  75 pmol/mmol creatinine) [Figure 2].

These results do not support the hypothesis that polyphenolic antioxidants derived from tea inhibit *in vivo* lipid peroxidation.





**Figure 1.** Urinary  $F_2$ -isoprostanes following five cups/day of hot water containing caffeine, green tea and black tea for seven days each in random order in subjects with raised blood pressure (mean  $\pm$  SEM).

**Figure 2.** Urinary  $F_2$ -isoprostanes following five cups/day of hot water and black tea for four weeks each in random order in subjects with mild elevations in serum total cholesterol concentrations (mean  $\pm$  SEM).

# The impact of xenoestrogens in the diet: feminizing agents or functional foods?

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Xenoestrogens are synthetic or naturally-occurring chemical compounds in the environment that are able to mimic the action of the female hormone,  $17\beta$ -estradiol (estrogen). This wide range of chemicals share a common mechanism involving occupancy of the estrogen receptor site to form a complex which may then bind to a specific region of a target gene, initiating protein synthesis and cell division. The estrogenicity of a wide range of compounds has been tested by measuring relative binding affinities, gene expression or cell proliferation.

International interest and concern about the significance of these compounds to human health has arisen from wildlife effects including the feminization of marine snails, reduced penis size in alligators, the thinning of egg shells and impaired reproductive function of seals. Possible human health effects include reduced sperm count and quality, cryptorchidism, hypospadias, male breast and testicular cancer. On the other hand, some groups of xenoestrogens, in particular the isoflavones and flavonoids, have beneficial effects which may reduce the risk of breast cancer in women, help to alleviate postmenopausal symptoms, and reduce the risk of cardiovascular disease, atheroscelerosis and cancer generally.

Food is a major route of exposure to xenoestrogens and we have assessed the daily intake of 20 naturally-occurring (soy isoflavones, lignans, coumestans, flavonoids, and resorcylic lactones) and synthetic xenoestrogens (organochlorine pesticides, PCB congeners, alkylphenols) known to occur in food. Dietary exposure of the wider New Zealand population was estimated from either New Zealand or international reports of concentrations of xenoestrogens in food and New Zealand consumption data (1,2). For an adult male, the estimated daily intakes were 0.015 mg estrogen equivalents/day on the basis of binding affinity to the receptor site and 0.003 mg estrogen equivalents/day on the basis of resulting cell proliferation. More than 98% of total estimated intake was from isoflavones and flavonoids.

When bioavailabilty is taken into account by factoring intake estimates with plasma concentrations, the estimated circulating blood level from all xenoestrogens combined, for an adult male, is approximately half the circulating level of endogenous estradiol. This would appear pharmacologically significant.

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# Dietary n-3 and n-6 fatty acids alter the molecular species profile of avian breast muscle phospholipids

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We have previously shown that dietary n-3 and n-6 polyunsaturated fatty acids (PUFA) reduce abdominal fat pad mass, plasma triglycerides and cholesterol in broiler chickens when compared to feeding saturated fatty acids (1). These changes may be a consequence of alterations in the fluidity of the plasma cell membrane composition (2) and this in turn may influence processes involved in energy metabolism. We investigated the effects of these dietary fats on the distribution of subclasses of choline (PC) and ethanolamine (PE) phospholipids in the breast muscle of these same broilers.

Day-old broiler chickens were reared in a brooder and fed a commercial starter diet for three weeks. They were then randomly divided into three groups (n = 10) and were fed the experimental diets for six weeks. The diets were isonitrogenous and contained 80 g/kg of either edible tallow sunflower oil or fish oil giving diets enriched in saturated fatty acids, n-6 PUFA or n-3 PUFA respectively. At end of feeding, samples of breast muscle were taken and later analysed for phospholipid molecular species.

Supplementation with the different fatty acids (FA) had no effect on the distribution of phospholipid subclasses. Sunflower oil and tallow resulted in a similar molecular species profile. For the diacyl PC phospholipids the principal species were 16:0-18:1(n-9) and 16:0-18:2(n-6) whereas, for the alkyl-enyl PC phospholipids the predominant species were 16:0-18:1(n-9) and 16:0-20:4(n-6). Of the diacyl PE phospholipids the dominant species was 18:0-20:4(n-6) and of the alkyl-enyl PE phospholipids the major species were 16:0-18:1(n-9), 16:0-20:4(n-6) and 18:0:20:4(n-6). Supplementation with fish oil significantly increased (P < 0.01) levels of both eicosapentaenoic acid (20:5n-3) and docosahexaenoic acid (22:6n-3) into their PC and PE phospholipids compared to the other diets. Increased n-3 PUFA incorporation was associated with decreased arachidonic acid (20:4n-6) in both PC and PE phospholipids.

Broilers fed the n- 3 and n-6 enriched diets have similar energy metabolism and this is different to tallow feeding (1). The present data indicates that membrane composition is similar for broilers fed sunflower oil and tallow but different for broilers fed fish oil. Taken together, these results suggest that changes in energy metabolism are not related to membrane phospholipid composition.

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# Diet containing cocoa powder with flavanols and procyanidins inhibits platelet function

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Flavanols and their related procyanidins are flavonoids found in foods such as tea, wine and cocoa powder, and are powerful antioxidants *in vitro* (1). The consumption of a high intake of a cocoa beverage, containing 897 mg total flavanols and oligomeric procyanidins, inhibited platelet activation and function in an acute study (6 hours) in humans (2).

The current study investigated the long-term effect of a lower dose of flavanols and procyanidins from cocoa powder using a double blind, randomised, placebo-controlled study with 32 subjects. Subjects were stratified into active and placebo groups based on plasma vitamin C levels prior to the study. Subjects on the active diet consumed 234 mg of flavanols and procyanidins (CocoaPro<sup>TM</sup>, Mars Inc) per day for 4 weeks, while subjects on the placebo tablet consumed an identical tablet made from cocoa powder with a low level of flavanols and procyanidins (< 1mg) for 4 weeks. Dietary restrictions were implemented to control the amount of flavonoids from the diet. Weighed food records, anthropometric measurements and fasting blood tests were performed at day 0 and 28. Plasma was analysed for F<sub>2</sub>-isoprostanes, TBARS, TRAP, the flavanols catechin and epicatechin, vitamin C, E, A, carotenoids and uric acid to determine the effect of oxidative damage. Plasma was also analysed for lipids and lipoproteins, while whole blood was analysed for platelet aggregation and platelet activation using flow cytometry.

Results showed that the plasma levels of epicatechin, catechin and vitamin C were significantly increased in the active group at day 28 and that platelet aggregation and activation (% of activated platelets) was significantly lower in the active group (p < 0.05) compared with the control, using two different agonists, at day 28. There were no significant differences between groups for vitamin E, A, the carotenoids nor plasma lipids and lipoproteins. In terms of antioxidant protection, there were no significant differences in TBARS, TRAP and F2-isoprostanes between groups. These results with a relatively low intake of cocoa flavanols and procyanidins over a 4-week period support the short-term data showing benefits on platelet function. *In vitro* data suggest that flavonoids inhibit platelet function by reducing  $H_2O_2$  production, and in turn, phospholipase C activation in the platelet (3). Further investigations with different levels of supplementation are recommended.

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# Lycopene concentration and antioxidant capacity after consuming tomatoes with olive oil

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Lycopene is a carotenoid found in high concentrations in tomatoes and tomato products and it is the most efficient singlet oxygen quencher of all the carotenoids (1). High lycopene concentrations have been found to be protective against myocardial infarction (2). Consumption of tomato juice, tomato paste and fresh tomatoes with corn oil or olive oil, increases plasma/ serum lycopene concentrations (3,4), although it is not known if this increase is associated with increased antioxidant capacity of the plasma.

This study examined plasma lycopene concentrations after a 9 d dietary intervention where tomatoes were cooked with extra virgin olive oil. Subjects (n = 10) were aged between 20–35 yr, of Anglo Celtic origin and in good health. They completed a 5 d diet avoiding dietary sources of lycopene, then consumed 4 tomato meals on consecutive days: two on the first and one on each of the second and third days. Each tomato meal contained 500 g of tomatoes and 20 mL of extra virgin olive oil. Fasting blood samples were taken at baseline, 24 h after the completion of a low lycopene diet, the morning following the two lycopene meals and 24 h after the third and fourth meals. Plasma carotenoids and vitamin E were measured using HPLC. The antioxidant capacity of the plasma was measured by ORAC, TBARS and a singlet oxygen assay.

Results indicated that avoiding foods containing lycopene led to a significant decrease in total plasma lycopene. Results also showed a significant increase in plasma total lycopene, *trans*-lycopene and *cis*-lycopene concentrations, when tomatoes were cooked and consumed with olive oil. There was no change in antioxidant capacity of the plasma as assessed by the ORAC assay, which assesses peroxyl radical scavenging ability.

	trans-Lycopene <sup>1</sup>	cis-Lycopene¹ (μg/100 mL plasma)	Total Lycopene <sup>1</sup>
Baseline	$20.39 \pm 3.62$	12.51 ± 2.47	$31.88 \pm 6.10$
Avoiding lycopene	$11.80 \pm 1.94$	$8.67 \pm 1.44$	$20.45 \pm 3.347$
2 meals	$19.79 \pm 2.37$	$13.45 \pm 1.37$	$33.33 \pm 3.53$
4 meals	$18.92 \pm 2.44$	$11.98 \pm 1.18$	$32.26 \pm 2.64$
P-value	0.0033	0.0359	0.0084

<sup>1</sup>mean ± SEM.

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# Are probiotics effective?

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Recent research has provided sound clinical evidence of the effectiveness of some defined strains of probiotic bacteria in helping to control several human disease conditions. Summaries of the present evidence have been presented in recent reviews (1–4). There are four strains with substantial published clinical data: *Lactobacillus rhamnosus* GG (Valio), *Saccharomyces cereviseae* Boulardii (Biocodex), *Lactobacillus paracasei* Shirota (Yakult), and *Bifidobacterium lactis* BB12 (Chr Hansen Labs). There are only 10 other strains with any peer-reviewed recently-published clinical data. These include: *Lactobacillus reuterii* (Biogaia), *Lactobacillus johnsonii* La1 (Nestle), and *Enterococcus faecium* SF68 (Cernelle).

There is now strong evidence that specific probiotic strains can alleviate antibiotic-associated diarrhoea, Clostridium difficile diarrhoea, rotavirus diarrhoea in children, other bacterial infections causing diarrhoea, and constipation. Exciting new findings are occurring in the use of probiotic bacteria (L-GG and BB12) to delay the development of food allergies and atopic eczema in young children (5). This could prevent the development of asthma in later life. Lactose intolerance is lessened by yoghurts and other fermented dairy products, and this effect is assisted by use of probiotic strains containing active β-galactosidase. There is proof that some strains may lower cholesterol levels, but this effect does not seem to be sustained. Many strains will promote immune responses, but the direct effect of such modulation on health is not clear in most cases. Evidence that probiotics may reduce the incidence and duration of travellers' diarrhoea is variable, and seems to depend on age group and cause of diarrhoea. Animal models provide evidence that development of bowel cancers may be prevented by probiotics, but the evidence is inconsistent and it is not yet possible to relate probiotic intake to prevention of the development of bowel cancer in humans (6). Bio-markers such as faecal enzyme, β-glucuronidase, show consistent reductions when humans consume many probiotic strains.

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### The intestinal microflora in Australian breast-fed and formula-fed infants

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Previous studies have found that breast-fed (BF) infants have an intestinal microflora dominated by bifido-bacteria possibly caused by bifidobacterial growth factors present in human milk which protect infants against bacterial pathogens. In contrast, formula-fed (FF) infants have more *Bacteroides*, enterobacteria and clostridia. In this study, we compared the effect of the type of feeding on the composition of the faecal microflora in 10 full-term, healthy Australian infants (five BF and five FF) aged 4 to 12 weeks.

Faecal samples were placed in an anaerobic chamber within 3 h of collection. Faeces (1 g) were homogenised and diluted 10-fold (10<sup>-1</sup>–10<sup>-8</sup>) g/ml in Wilkins-Chalgren anaerobe broth. One hundred microlitres of each dilution were plated in duplicate and incubated anaerobically at 37°C on Wilkins-Chalgren anaerobe blood agar (2 days, total anaerobes), and supplement (2 days, *Bacteroides*), Reinforced Clostridial agar (preparations were heat treated for 10 mins at 90°C to select for clostridial spores) (2 days, clostridia), Rogosa agar (2 days, lactobacilli) and raffinose bifidobacteria agar (3 days, bifidobacteria). Plates which contained the following media were incubated aerobically on nutrient agar (1 day, total aerobes), and MacConkey agar (1 day, enterobacteria). After incubation, colonies were counted and identified by colony morphology. Bacterial counts were calculated as log 10 of colony-forming units/g of faeces. Faecal pH was measured with a digital pH meter.

	Faecal bacterial counts <sup>1</sup>			
	Breast-fed $(n = 5)$	Formula-fed $(n = 5)$		
Total anaerobes	11.21 ± 0.39	$10.08 \pm 0.71$		
Bacteroides	$8.88 \pm 0.91$	$9.86 \pm 0.62$		
Bifidobacteria	$9.35 \pm 0.33$	$7.82 \pm 0.79$		
Lactobacilli	$7.94 \pm 0.90$	$2.72 \pm 0.792$		
Clostridia	$1.98 \pm 1.22$	$4.76 \pm 2.05$		
Total aerobes	$8.49 \pm 0.10$	$9.06 \pm 0.17$		
Enterobacteria	$7.03 \pm 0.71$	$9.22 \pm 0.052$		

<sup>&</sup>lt;sup>1</sup>mean ± SEM. Colony forming units.

The composition of the intestinal flora was found to be different between BF and FF infants. Breast-fed infants had higher faecal bacterial counts of lactobacilli than FF infants (P < 0.05). Lactobacilli were present in the faeces of all BF infants but only three of the five FF infants. Formula-fed infants had higher counts of enterobacteria than BF infants (P < 0.05). Bifidobacteria were the predominant faecal bacteria in BF infants. Conversely, *Bacteroides* were the predominant faecal bacteria in FF infants. There were no marked differences between the groups in counts of *Bacteroides* or clostridia. Faecal pH was significantly lower in the BF group (5.47  $\pm$  0.06) than in the FF group (7.34  $\pm$  0.17) (P < 0.05). This study supports other research findings on the benefits of breast-feeding on the intestinal microflora of infants.

<sup>&</sup>lt;sup>2</sup>significantly different from breast-fed group, (P < 0.05).

# The effect of two years milk supplementation on bone mineral accretion in Chinese adolescent girls

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To investigate the effect of milk supplementation on bone mineral accretion during early puberty, a two-year double-blind, controlled supplementation trial with vitamin D and/or calcium fortified milk was carried out in Chinese 757 girls aged 10 years consuming plant-based diets. There were divided into three groups according to of randomly selected Beijing schools: In Group 1 schools, subjects received 330 mL UHT milk fortified with Ca as milk salts (providing an extra 560 mg Ca/day) on every school day; in Group 2 schools, subjects received the same milk additionally fortified with vitamin D (8  $\mu$ g/day); in Group 3 schools, subjects were unsupplemented controls. is the control group. Bone mineral density (BMD) of distal (DF) and proximal forearm (PF), wasere measured in all subjects, and of total body (TB) was measured in a sub-sample of 414 girls by dual X-ray absorptiometry (DXA) at baseline and end-trial.

	Group 1 (milk) n = 237	Baseline Group 2 (milk + vitD) n = 260	Group 3 (control) n = 260	
Weight (kg)	33.79 (7.24)	33.43 (7.01)	33.73 (6.94)	45.53 (9.68) 45.34 (9.26) 44.11 (8.94)
Height (cm)	140.48 (6.39)	141.13 (6.99)	141.11 (6.46)	153.73 (6.53) 154.12 (6.60) 153.25 (6.34)
DFBMD	0.233	0.233	0.233	0.277 0.276 0.280
(g/cm <sup>2</sup> )	(0.029)	(0.029)	(0.028)	$(0.047) \qquad (0.044) \qquad (0.047)$
PFBMD	0.479	0.480	0.480	0.541* 0.543* 0.492
(g/cm <sup>2</sup> )	(0.050)	(0.049)	(0.051)	$(0.084) \qquad (0.080) \qquad (0.072)$
TBBMD	0.690	0.683	0.696	0.743 0.746 0.726
(g/cm <sup>2</sup> )	(0.058)	(0.049)	(0.053)	$(0.084) \qquad (0.078) \qquad (0.083)$

Values are mean (SD); \* P < 0.001; # subject nosas 145, 136, 133 at baseline, 112, 114, 124 at end-trial for 24 months, respectively.

A total of 327 days supplementation was provided over day during the two years was 327 days. The additional Ca intakes were averaged 251 mg per day over this period. While only 56.3% of subjects had breast development either at Tanner stage 2 or 3 at baseline, 81.5% had reached Tanner stage 2 or 3, and 13.8% had reached Tanner stage 4 or 5 at 24 months. No significant differences in terms of weight, height and pubertal status wereas found between groups at baseline or end-trial 24 months. Both supplemented groups had significantly higher PFBMD than controls group at end-trial 24 months. Compared with controls group, Group 1 and Group 2 subjects had significantly higher percentage gains (mean  $\pm$  SEM) in PFBMD (13.06  $\pm$  0.92, 12.88  $\pm$  0.83 vs 2.78  $\pm$  0.90, P < 0.001), and TBBMD (7.02  $\pm$  0.59, 8.89  $\pm$  0.61 vs 3.86  $\pm$  0.58, P < 0.001). 24 months Mmilk supplementation on school days over 24 months, significantly increased bone mineral accrual in Chinese adolescent girls. If this e gain persists, the eventual peak bone mineral density should might also be increased in the study subjects. Milk supplementation (330 mL with a total of 560 mg Ca with/without 8 mg vitamin D on school days over two years) significantly increased bone mineral accrual in Chinese adolescent girls. If this gain persists, peak bone mineral density may increase in supplemented subjects, and reduce the risk of future osteoporotic fracture.

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# Do differences in nutrient intake predict differences in bone mass in boys: a co-twin control study

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Genetic factors determine a large proportion of the variance in bone traits such as size, mass and volumetric density. The proportion of variance attributable to genetic factors can be determined using the classic twin model, in which it is assumed that similarities in life style factors, such as diet, are the same within monozygotic (MZ) pairs as they are within dizygotic (DZ) pairs. The effects of home environment, however, may differ less within MZ pairs than within DZ pairs, and this may account in part for their greater resemblance. This assumption has not been rigorously tested in young males. Protein and calcium intakes are important nutrients for bone mass accrual. Protein insufficiency during growth is associated with delayed skeletal maturity, and reduced cortical and trabecular bone (1). Calcium supplementation has been associated with increased bone mass accrual in children (2). Using data from male twins, we determined the similarity in nutrient intake within MZ and DZ pairs, and the extent to which within pair differences in bone mass and anthropometry could be explained by within pair differences in nutrient intake.

We studied 36 MZ and 39 DZ male twin pairs aged  $11.3 \pm 2.9$  years (range 7–20 years). Bone mass and body composition were measured using dual energy x-ray absorptiometry (DXA). Dietary intake was assessed using 3-day weighed food diaries, and analysed using FoodWorks Nutrition Program (Version 2.10). Anthropometry was measured using standard methods. Similarities within pairs were assessed using Pearson's correlation. The extent to which within pair differences in bone mass could be accounted for by within pair differences in nutrient intake was determined using multiple linear regression through the origin. Data was analysed using StatView (version 4.51).

MZ and DZ twins did not differ in mean age, bone mass, anthropometry or nutrient intake. Age-adjusted correlations for height, sitting height, leg length and bone mass ranged from r=0.86-0.96 for MZ pairs and r=0.68-0.78 for DZ twins (all P < 0.01). Age-adjusted correlations for calcium, protein and energy intakes were r=0.89, r=0.77 and r=0.84 for MZ pairs and r=0.43, r=0.43 and r=0.52 for DZ pairs, respectively (all P < 0.01). Within pair differences in protein intake were marginally significant predictors of within pair differences in total body ( $\beta=0.3$ ) and leg BMC ( $\beta=0.3$ ) (P < 0.08), but not axial BMC. Within pair differences in calcium and energy intake did not predict differences in bone mass. Within pair differences in nutrient intake did not predict differences in height, sitting height or leg length.

MZ pairs differ less in their dietary intake than do DZ pairs. These data suggest that about 9% of the variance in within pair differences in BMC at several sites was explained by within pair differences in protein intake. Dietary calcium intake did not appear to be an independent predictor of bone mass, but this could be a type 2 error due to lack of power. Protein intake may be a more important factor in bone mass accrual at the legs than the spine. The importance of dietary protein intake in relation to bone mass accrual is becoming more apparent.

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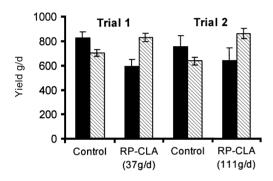
## Rumen protected conjugated linoleic acids: effects on milk composition in dairy cows

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Conjugated linoleic acids (CLA) are geometrical and positional isomers of conjugated linoleic acid having potent metabolic effects. They reduce plasma triacylglycerol, cholesterol, fat deposition and have anti-cancer and anti-inflammatory properties (1). In ruminants CLA are formed either by partial hydrogenation of C18-di and tri-unsaturated fatty acids in the rumen or are synthesised in tissues from trans-11-octadecanoic acid via the  $\triangle 9$  desaturase pathway (2). Ruminant-derived foods provide significant sources of CLA in the human diet and because of their potential health benefits, current research is directed towards increasing the CLA content of meat and milk products. Previous studies have shown abomasal infusions of CLA and dietary supplements of unsaturated oils increased the CLA content in milk but had no effect on milk protein yield (2).

The effect of feeding CLA protected from ruminal hydrogenation (RP-CLA) by encapsulation in an inert matrix of protein (3) on milk composition are presented in the figure below.



Protein yield  $\square$  and fat yield  $\blacksquare$  in Holstein cows (trial 1, n = 5; trial 2, n = 3) at pasture, supplemented with RP-CLA for 4 days.

In short term feeding trials supplements of RP-CLA significantly increased milk protein yield (P < 0.05) and reduced milk fat yield (P < 0.05); the proportion of CLA in milk increased from 1.4 to 2.2%. The CLA-induced increase in milk protein yield reflects a major re-channeling of nutrient use in the dairy cow; where protein synthesis and secretion is enhanced and lipogenesis is inhibited. Long term feeding trials are required to assess the impact of RP-CLA on lactation and reproductive performance.

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# High carbohydrate and high monounsaturated fat dietary targets produce similar outcomes in the management of type 2 diabetes mellitus with concomitant reduced saturated fat intakes

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Dietary intervention is the cornerstone of treatment for type 2 diabetes mellitus (T2DM). American recommendations are defined in terms of macronutrient energy proportions: 10–20% Protein, < 10% saturated fatty acids (SFA),  $\le 10\%$  polyunsaturated fatty acids with the remaining 60–70% to comprise carbohydrate (CHO) and monounsaturated fatty acids (MUFA) (1). The decision to include more MUFA or CHO is based on the need to control energy intake, but not overload a CHO sensitive system (2). In this study we compare the effects of two dietary approaches which fit within the current guidelines: a high CHO, low SFA diet and a high MUFA, low SFA diet.

Fifty six men and women diagnosed with T2DM in the last 2 years were recruited from the Illawarra Diabetes Service. Subjects were randomised to either a high carbohydrate (53% CHO/ 12%MUFA) or a high MUFA (43% CHO / 22% MUFA) diet, with both diets comprising < 10%SFA, ≤ 10% PUFA and 15% Protein under weight maintenance conditions. Diet histories and 3 day food records were done at baseline and 3 monthly intervals for 12 months. Outcome variables were changes from baseline in weight, waist circumference, HbA1c, and in plasma cholesterol, triglycerides and HDL cholesterol. On completion, data were available from 19 MUFA- and 23 CHO- group subjects.

There were no significant differences between groups for usual dietary intakes on entry to the study. Subjects in both groups needed to reduce their SFA intakes (mean baseline intakes  $12 \pm 2\%$  energy). By 12 months both groups had achieved reduced SFA intakes (no difference between groups), and the MUFA diet group were consuming significantly more MUFA than the high CHO group (p < 0.05). The high CHO group were consuming more CHO than the MUFA group, but this difference was not significant. There were no significant differences between groups in changes in clinical measures at 12 months. Both groups showed a significant increase from baseline in HbA1c \* (p < 0.05) indicating deterioration in metabolic control, but no change in weight, waist circumference or plasma lipids.

Change variable	High MUFA diet1	High CHO diet1
Weight (kg)	-0.69 (0.53)	-0.26 (0.71)
Waist circumference	-1.8 (0.87)	-1.4(0.76)
HbA1c (%)	+0.92 (0.42)*	+0.63 (0.28)*
Cholesterol (mmol/L)	-0.14 (0.22)	-0.03(0.14)
Triglyceride (mmol/L)	-0.10 (0.19)	-0.20(0.30)
HDL cholesterol (mmol/L)	-0.02 (0.06)	+0.06 (0.03)

<sup>&</sup>lt;sup>1</sup>mean (SEM) \* significant change at P < 0.05.

This study suggests that the current allowance for some flexibility in the CHO / MUFA component of the diet produces similar clinical outcomes, but further efforts are needed to improve glycemic control.

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## Saturated fat intake linked to risk of inflammatory bowel disease – results of a case control study

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**Objective** – To determine the pre symptomatic dietary factors which predispose to the development of inflammatory bowel disease (IBD).

**Design** – Case control study of newly diagnosed cases with IBD matched (within 5 years of age, gender and geographic location) to randomly selected (electoral roll) multiple controls. Cases were recruited within 6 months of diagnosis from NSW and ACT by referral from gastroenterologists. Diet was assessed within 2 years of onset of symptoms prior to diagnosis using a 218 item food frequency questionnaire which included vitamin supplement use. Questions on potential confounders (education, work, nationality, supplement use) or effect modifiers (smoking, breastfed, oral contraceptive use, appendectomy and tonsillectomy) were included. Energy adjustment was used in the analysis and conditional logistic regression for matching.

**Outcomes** – Data from 107 case and 308 matched controls were useable for analysis. Education, work status, tonsillectomy, vitamin supplement use and alcohol use were not associated with IBD. Having ever smoked (prior to symptoms) was significantly associated with IBD, although current smoking was not. Having been breastfed was negatively associated with IBD, while OC use was positively associated with IBD. Median energy intake was higher (P < 0.01) in cases (11.4 MJ) than controls (10.0 MJ). Utilising energy adjustment by regression analysis resulted in total fat, and saturated fat intake being higher in cases than controls. Using energy adjusted nutrient intakes, conditional logistic regression produced odds ratios that were significantly higher for total fat, saturated and monounsaturated fat. Controlling for the effect modifiers/confounders found in the univariate analysis left saturated and total fat intake as the only significant predictors of IBD (Odds ratio 2.96 and 2.23 respectively, highest versus lowest quartile).

	Quartile 1 (OR =1)	Quartile 2	Quartile 3	Quartile 4	Q 2 OR	Q 3 OR	Q 4 OR	95%C CI	Trend P
Total Fat (g)	< 81	81–95	95–106	> 106	1.39	1.79	2.43	1.22-4.82	0.010
Saturated (g)	< 32	32-37	37–43	> 43	1.30	1.91	2.64	1.34-5.19	0.003
Monounsaturated(g) After adjustment*	< 29	29–34	34–38	> 38	1.50	1.43	2.06	1.05-4.01	0.049
Total Fat (g)	< 81	81–95	95–106	> 106	1.37	1.91	2.23	1.08-4.63	0.026
Saturated (g)	< 32	32–37	37–43	> 43	1.66	2.03	2.96	1.41-6.20	0.007

<sup>\*</sup>Three confounders controlled in model – oral contraceptive use, past smoking & breastfed less than 6 weeks.

**Conclusions** – Increased consumption of saturated and total fat in the diet prior to symptom appearance is related to the subsequent appearance of IBD. Calculation of the population attributable risk from this data suggests that about one third of cases could be avoided if the population reduced saturated fat consumption below the top quartile of intake.

### How effective is the 'low fat' message?

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Public health nutrition strategies have emphasised fat reduction. Attitudinal research indicates that fat consumption is associated with weight gain and heart disease and hence fat restriction is perceived as beneficial (1). Knowledge of low fat strategies for food selection and preparation suggest that the 'low fat' message has been successfully communicated. However, there is confusion and hence little consideration of type of fat.

The Heart Foundation (HF) policy on dietary fat and cardiovascular disease (CVD), based on a rigorous review of the scientific evidence, places greater emphasis on type of fat. In particular, reducing saturated fatty acid (SAFA) and increasing polyunsaturated fatty acid (PUFA) (2). Little evidence was found to support a recommendation for total fat and CVD. A subsequent HF review on the relationship between dietary fat and body weight found that energy density, rather than fat, is a major dietary determinant of energy intake. Since energy density is affected by several factors, fat reduction alone may not reduce energy intake.

Analysis of the 1995 National Nutrition Survey (NNS) data showed that on the day surveyed, only 1% of diets complied with both SAFA and PUFA recommendations, 15% with the SAFA recommendation and 10% with the PUFA recommendation (3). High intakes of whole milk, cheese, pastries, butter and cereal-based mixed dishes prevented compliance with the SAFA recommendation and low intakes of polyunsaturated margarine and oil, the PUFA recommendation. Fat modification strategies were more effective than fat reduction strategies in shifting the diets of adults towards SAFA and PUFA recommendations (3).

Dietary modelling was conducted to ensure that public health dietary strategies and food-based recommendations reflect the scientific evidence. Manipulations of a model, based on the eating patterns of adults in the NNS, showed that butter, cheese and takeaway foods (for dinner) had the most negative effect on the ratio of PUFA to SAFA. Conversely, soybean and sunflower oils, low SAFA commercial deep-frying oil and fortified soy beverage had the most positive effect on the PUFA to SAFA ratio in the model diet. It also showed that 25 g of spreads and oils can be included in an energy restricted diet and still meet SAFA, PUFA and ALA recommendations.

The evidence suggests that the emphasis of dietary messages for CVD prevention must evolve from 'low fat' to 'type of fat' with due consideration to energy density. In addition, several foods must be targeted and specific recommendations on the type and amount of foods are required to more effectively reduce CVD.

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## Pregnancy and lactation have no long-term adverse effects on bone mass: a twin study

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Pregnancy and lactation place significant stress on maternal calcium homeostasis, and may result in substantial changes in bone mineral density. While bone loss in the period immediately following parturition is well-documented (1), there is not a clear consensus regarding long-term recovery in bone mineral from the effects of either pregnancy or lactation. We retrospectively assessed the number of pregnancies and duration of breast feeding in relation to bone mineral density (BMD) in female twins, using cross-sectional and co-twin model approaches.

Female twins and siblings (n = 1354) > 18 years of agewho were grouped according to number of pregnancies: never pregnant (NP) (n = 426), 1–2, (2P) (n = 455) and > 3, (3P) (n = 473). Of these subjects 83 twin pairs were identified where one twin within a pair had been pregnant (> 20 weeks) and the other had never been pregnant beyond 20 weeks. Information on pregnancies and breast feeding was obtained by questionnaire and bone density at lumbar spine (LS), total hip (HP), and total body bone mineral content (TBMC) by dualenergy x-ray absorptiometry (Hologic QDR 1000W).

Those who were never pregnant were younger (NP  $33.1 \pm 0.68$  years ( $\pm$  SEM), 2P  $45.1 \pm 0.53$  years and 3P  $49.8 \pm 0.48$  years (P < 0.05 ANOVA)), had a lower BMI (NP  $24.2 \pm 0.22$ , 2P  $25.9 \pm 0.24$  and 3P  $26.4 \pm 0.23$  (P<0.05)) and were taller (NP  $163.2 \pm 0.31$ cm, 2P  $162.5 \pm 0.31$ cm and 3P  $161.4 \pm 0.31$ cm (P < 0.05)). After adjustment for age, lean and fat mass, groups 2P and 3P had 3.8% higher LS BMD compared with NP (P < 0.001), and TB BMC was 2.7% higher in 2P and 3.1% higher in 3P compared with NP (P < 0.001) and HP BMD was greater in 3P compared to NP by 2% (P < 0.01). Of the Of the 928 parous individuals parous women 87% breast-fed (> one month). After adjustment for age, lean, fat mass, TB BMC was higher in those who breast-fed ( $2.30 \pm 0.34$ kg) compared with those who did not ( $2.24 \pm 0.02$  kg) (P < 0.01).

71% breast-fed and there were 58 parous twin pairs where one twin breast-fed and the other did not. There were no significant differences in height, weight, BMI, or HP BMD, LS BMD, TB BMC between breast feeding twin and non-breast feeding twin.

In 83 twin pairs (21 monozygotic, 62 dizygotic), mean age 42.2 (15.7) (SD) years, who were discordant for ever being pregnant, the parous twins had a mean of 2.3 (0.13) pregnancies and breast-fed for 8.39 (1.67) months per child. There were no significant differences in height, weight, BMI, or HP BMD, LS BMD, TB BMC between nulliparous and parous twins.

Of the parous women 70% breast-fed and there were 58 parous twin pairs where one twin breast-fed and the other did not. There were no significant differences in height, weight, BMI, or HP BMD, LS BMD, TB BMC between breast feeding twin and non-breast feeding twin.

These results indicate that there is no long-term detrimental effect of pregnancy or breast feeding on bone density. There was and some evidence from the cross sectional analysis to suggest that pregnancy may increase bone density, although no within-pair difference in bone was observed in twin pairs discordant for ever being pregnant. Therefore, although there may be acute reduction in bone mineral density with pregnancy and breast feeding, mothers appear to readily replace the bone lost after a period of time.

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## Increased dietary saturated fat intake decreases the ratio of thromboxane/prostacyclin in healthy male subjects

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The ratio of thromboxane A<sub>2</sub>/prostacyclin I<sub>2</sub> (TXA<sub>2</sub>/PGI<sub>2</sub>) plays a critical role in platelet aggregation (Moncada and Vane 1979). Evidence from dietary intervention studies has found that the ratio of TXA<sub>2</sub>/PGI<sub>2</sub> was decreased by marine omega-3 polyunsaturated fatty acid (n-3 PUFA) in humans (Ferretti et al 1998, von Schacky et al 1985). However, there is no data on the effects of the diet high in saturated fat from animal source on the ratio of urine stable metabolites of TXA<sub>2</sub>/PGI<sub>2</sub> in literature. The aim of the present study was to investigate the effect of dietary saturated fat on ratio of urine stable metabolites of TXA<sub>2</sub>/PGI<sub>2</sub>.

In the present study we investigated the effect of dietary saturated fat on the ratio of urine excretion 11-dehydro thromboxane  $B_2$  (TXB<sub>2</sub>) and 6-keto prostaglandin F 1a (PGF 1 $\alpha$ ) in 27 healthy aged 30 to 55 years free-living male subjects. Each volunteer was randomly assigned to one of the two diets for a period of 4 weeks, after which each subject resumed his usual diet for 2 weeks as a 'wash-out period', before being assigned to the other diet for a further 4 weeks. The two diets were designed to provide similar amounts of energy, protein, dietary fiber, and alcohol, differing only in the amount of fat. The high fat (HF) diet was designed to provide 10–15 % more energy from animal fat compared to the low fat (LF) diet. Twenty-seven subjects collected their 24-hour urine on the last day of each of the diets. The samples were stored at – 20°C for later analysis. The concentrations of 11-dehydro TXB<sub>2</sub> and 6-keto prostaglandin F 1 $\alpha$  in the urine was determined by using an enzyme immunoassay (EIA) method with commercially available EIA kits. Serum lipids from 12 randomly selected subjects were extracted by chloroform: methanol (1:1, v/v). Methyl esters of fatty acids of serum lipids were prepared by standard methods. Methyl esters of fatty acids were separated by gas chromatography as described.

The ratio of urine excretion 11-dehydro  $TXB_2$  and 6-keto PGF 1a was significantly lower in the HF (2.7  $\pm$  0.2) than in the LF diet (3.1  $\pm$  0.3) (p < 0.05). Serum concentration of 20:4n-6 was 6% higher in the HF than in the LF diet, while the proportion of 20:4n-6 was 5% lower in the HF than in the LF diet. Compared with the LF diet, the concentration and proportion of 14:0, 18:0, 20:0 and total saturated fatty acid in serum was significantly higher in the HF diet (p < 0.05), and 18:3n-3 and the ratio of n-3 PUFA to n-6 PUFA was significantly lower in HF diet (p < 0.05). The present result indicate that decreased ratio of urine excretion of 11-dehydro  $TXB_2$  to 6-keto PGF 1 $\alpha$  in the HF diet compared with the LF diet may be caused by decreased intake of 20:4n-6 proportion, rather than intake of absolute amount of 20:4n-6.

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## Influence of dietary stearic acid enrichment on individual platelet phospholipid fatty acid composition

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It is widely accepted that stearic acid, as opposed to saturated fats in general, is not hypercholesterolemic. In addition, stearic acid enriched diets have previously been shown to reduce platelet aggregation (1) and platelet size as measured by mean platelet volume (MPV) (1,2). This decrease in MPV, indicative of platelets in a quiescent, non-activated state, may represent a reduced thrombotic tendency.

Many cell functions are influenced by their membrane fatty acid composition and thus, it is important to determine if there is preferential distribution of stearic acid amongst specific platelet phospholipid fractions and if stearic acid incorporation affects the level and distribution of other specific fatty acids.

Five healthy male subjects aged  $44 \pm 14$  years consumed a stearic acid (C18:0) enriched diet at a level of 6.4% total energy (~ 20 g per day compared with an habitual intake of ~ 8 g per day) for four weeks. Habitual and intervention dietary intakes were measured using seven day weighed food records. Venous blood was collected for Full Blood Examination including measurement for MPV and platelet fatty acid determination at days 0 and 28. The platelet phospholipid (PL) classes, phosphatidylethanolamine (PE), phosphatidylcholine (PC), phosphatidylserine (PS) and phosphatidylinositol (PI), were separated by TLC using the solvent system: methyl acetate: propan-1-ol: choloroform: methanol:0.25% aqueous KCI (25:25:25:10:9, by vol). Fatty acid methyl esters were prepared by a standard method and identified using Gas Chromatography.

Stearic acid levels increased significantly (P < 0.05) in the PE and PC platelet PL fractions by 17% and 15%, respectively, compared with baseline levels. In the PI fraction, there was a non-significant trend to decrease stearic acid levels combined with a significant increase in the level of linoleic acid and a significant decease in the level of arachidonic acid (AA) by 248% and 13%, respectively, compared with baseline levels. No differences were observed in MPV.

	PE <sup>1</sup>	PC <sup>1</sup>	PS <sup>1</sup>	PI <sup>1</sup>
C18:0% at baseline	$15.63 \pm 0.46$	13.79 ± 1.31	41.51 ± 2.29	$38.16 \pm 0.76$
C18:0% at day 28	$18.22 \pm 1.37^{a}$	$15.92 \pm 2.25^{a}$	$41.82 \pm 1.38$	$35.81 \pm 2.33$

<sup>&</sup>lt;sup>1</sup>mean  $\pm$  SD, <sup>a</sup>significantly different to baseline (P < 0.05).

The significant decrease in AA in the PI fraction may be linked to the previously encountered lower platelet aggregation in stearic acid enriched diets (1).

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# Relationship between platelet phospholipid polyunsaturated fatty acids and dietary intake of fish, meat and polyunsaturated fat in male Melbourne Chinese and Caucasian

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Increased n-3 polyunsaturated fatty acid (PUFA) in the tissues is associated with decreased risk of cardio-vascular disease (1). The aims of this study were to investigate (1) platelet phospholipid (PL) polyunsaturated fatty acid (PUFA) composition in subjects who were the Melbourne Chinese migrants compared with those who were the Melbourne Caucasians, (2) the relationship between platelet PL PUFA and intake of fish, meat and PUFA. Ninety-seven Melbourne Chinese males aged between 25 to 55 years and 78 age and sex matched Caucasians were recruited in Melbourne. Dietary intake was assessed using a semi-quantitative Food Frequency Questionnaire. The platelet PUFA was measured by gas liquid chromatography.

The Melbourne Chinese had a significantly higher intake of fish (p = 0.012) and white meat (p = 0.0045) compared with the Melbourne Caucasians and had significantly higher proportions of platelet PL 20:5n-3 (p = 0.006), 22:6n-3 (p < 0.0001), total n-3 (p = 0.027) and 22:5n-6 (p = 0.0002). The Melbourne Chinese had a significantly lower intake of red and total meat (p < 0.0001) than the Melbourne Caucasians, and significantly lower proportions of 20:3n-6 (p = 0.023), 20:4n-6 (p < 0.002), 22:4n-6 (p < 0.0001), total n-6 (p = 0.037), 22:5n-3 (p < 0.0001) and ratio of n-6/n-3 (p = 0.011).

	Fish (g/day)		Meat (g	g/day)	PUFA	PUFA (g/day)	
	Std. Coeff.	P value	Std. Coeff.	P value	Std. Coeff.	P value	
18:2n-6	0.001	0.968	-0.146	0.135	0.128	0.190	
20:3n-6	0.030	0.701	-0.172	0.070	0.312	0.001	
20:4n-6	-0.032	0.691	0.026	0.787	0.044	0.652	
22:4n-6	-0.125	0.114	-0.076	0.424	0.242	0.012	
22:5n-6	0.034	0.664	< 0.0001	0.993	-0.311	0.039	
22:5n-3	-0.235	0.002	0.146	0.104	0.218	0.016	
22:6n-3	0.211	0.003	-0.064	0.446	-0.415	< 0.0001	
22:6n-3/22:5n-3	0.277	< 0.0001	-0.126	0.105	-0.448	< 0.0001	

Multiple linear regression result (Table) indicated that platelet PL 20:5n-3 and 22:6n-3 were positively correlated with fish intake, and negatively correlated with dietary intake of meat and PUFA, while 22:5n-3 was positively correlated with dietary meat and PUFA intake, and negatively correlated with fish intake. Dietary intake of PUFA and fish are potential confounding factors for assessing the effects of meat consumption on platelet PL individual PUFA. Dietary intake of PUFA and meat did not influence the incorporation of fish long chain n-3 PUFA into platelet PL in this study population.

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### The demographic dimension: past and future

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The human population took hundreds of thousands of years to reach 1 billion in the mid-nineteenth century, since when it has risen to 6 billion; it will probably reach some kind of equilibrium under 10 billion during the 21st century. This has been a unique historical event and is the social aspect of the Industrial Revolution. For most of this time population growth was constrained by food resources, as described by Malthus just as the period was ending. Boserup (1) argues that population growth itself was the main mechanism in increasing food resources.

We are still in a transitional period during which science and capital have greatly increased food production and, by lowering mortality, population numbers. Third World mortality fell steeply after World War II causing unprecedentedly high rates of population growth. The constraint of growth by reducing birth rates was brought about by birth control resulting from socio economic change, scientific breakthroughs and organised family planning programs. Food production has kept up with population growth, although there are still large numbers of undernourished people.

The presentation will focus on two issues, the future of population growth and the resource problems created by such growth. The United Nations population projections (2,3) will be examined to demonstrate global and regional implications. The end point of the demographic transition is no longer seen as being necessarily constituted by stationary population. In a world where 44% of the population already lives in countries with below-long-term-replacement fertility, it is quite possible that human numbers will peak at 8–10 billion during the 21st century and then begin a long period of decline. Fears that rapid population growth would impede economic growth or would outstrip increases in food production have so far proved unfounded and will probably remain so in the 21st century. The real question is the long-term equilibrium between population and resources in a world of almost 10 billion people, or sustainability in a situation where that number represents a hump preceding smaller numbers.

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### Prospects for the Third Horseman in an environmentally stressed biosphere

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Human numbers will reach an estimated 8–9 billion by 2050. This along with continued economic development in today's low-income countries, means that the total global demand for food will increase by around threefold over the coming half-century. Will a combination of high-tech precision farming, enlightened practices (such as low-till and mixed-crop farming), equitable land tenure and socially-attuned use of GM technology suffice to keep St. John the Divine's 'Third Horseman' at bay? Opinions vary (1,2).

Against this background of social changes and technical possibilities, today's emerging global environmental changes, such as climate change, will also affect food production. Other incipient large-scale environmental changes that are affecting, or will affect, food production include stratospheric ozone depletion, biodiversity losses (with knock-on effects on crop and livestock pest species), and the perturbation of several of the great elemental cycles of nitrogen, sulphur and phosphorus (3). Further, current agricultural practices are increasingly damaging to the biosphere at large, and entail deforestation, chemical pollution of soils and waterways, destruction of habitat and increased risks of infectious diseases in livestock – and their passage into human populations (4,5).

These various environmental changes will affect the production of crops and livestock on land and wild and cultivated fisheries in various and complex ways. The modelling of how global climate change is likely to affect world and regional food production is illustrative. On balance, recent modelling-based estimates indicate that, in the medium-to-longer term, if not over the next several decades, climate change will affect crop yields adversely, especially in food-insecure regions (6). An increase in climatic variability will amplify the risks to future food production.

Our capacity to maintain food supplies for an expanding and increasingly expectant world population will depend on maximising the efficiency and sustainability of production methods, using genetic biotechnologies wisely, and minimising ecologically damaging environmental changes. Resolution of the longstanding disparity between the world's rich and poor, with widespread chronic deficits in 'food entitlements' (7) alongside an unprecedentedly well-fed privileged minority, should also be part of any future sustainable solution.

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### The impact of changing world resources on food security

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It is usual to think of resources as ones to do with people, fuel (energy) and minerals, food and water (1). Each of these is highly relevant to human food security. But information resources and intelligence, now increasingly a part of information technology (IT), and systems, especially ones of governance and culture, are also crucial (2). Bringing together these resources for food security in a way that is sustainable is the greatest challenge.

New and enhanced resources include those of population (especially older people), cultural fusion, information, renewable energy and biotechnology (3). Good governance is being both strengthened and weakened depending on location, information and education systems, burden of disease, and other factors. Gravely threatened are potable water supplies. Food transport may become precarious if jet-fuel dependent and local food production has diminished – yet it has played an important role in the diversification of the human diet. The decline in eco-systems and biodiversity may make food variety and what it offers food security more uncertain – generalised food variety has been a relatively recent human achievement through food cultural exchange, agricultural and horticultural development, food trade, and economic development; it may be short-lived.

Food and nutrition policy can no longer ignore its own impact on world resources and their sustainability.

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