Expected outcomes of genomics R&D and implications for human and animal nutrition

H Oddy

Meat and Livestock Australia, Sydney, NSW, 2000

No abstract received at time of going to print.

Aqueous tomato extract inhibits platelet aggregation

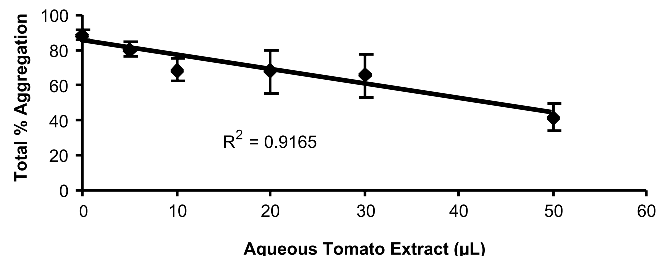
SA Lazarus[1], AK Dutta-Roy[2], ML Garg[1]

[1]*Nutrition & Dietetics, University of Newcastle, NSW, 2308*
[2]*Institute for Nutrition Research, University of Oslo, Norway, N-0316*

Epidemiological data have revealed a relatively low incidence of heart disease in Italy and other Mediterranean countries that cannot be entirely accounted for by traditional risk factors and have identified tomatoes as a component in the diet that may help explain the protective effect observed (1). Indeed, data have inversely correlated blood levels of lycopene, of which tomato products contribute approximately 80% to the diet, to incidence of acute coronary events, development of early atherosclerosis, and mortality from heart disease (2–3). Blood platelets play an important role in the development and stability of plaques. Recently, the presence of anti-platelet factor(s) in tomatoes has been reported (4). However, the active components in tomatoes and mechanism of their anti-platelet action are still unknown. To this end, the purpose of the present study was to determine the extent to which an aqueous tomato extract could inhibit collagen-induced platelet aggregation *in vitro*.

Fasting blood samples were collected into sodium citrated tubes from healthy volunteers who had not taken medications known to affect platelet function for the previous seven days. Platelet-rich plasma was obtained by centrifugation and the platelet counts (1.5-4.0 x 10^8 per mL) determined by microscopy using a hemocytometer. Platelet aggregation was measured following the addition of PBS (pH 7.4) or incubation with the aqueous tomato extract for 15 minutes using a dual channel optical aggregometer (Chronolog).

The aqueous tomato extract inhibited platelet aggregation induced by collagen (2 µg/mL) in a dose-dependent manner ($R = -0.95733$, $P = 0.003$), and the amount of aqueous tomato extract required to inhibit aggregation by 50% (IC_{50}) was determined to be 51 µL in the 500 µL reaction system (see Figure). The addition of 40 µL of aqueous tomato extract significantly inhibited collagen-induced platelet aggregation ($P = 0.007$), with the amount of collagen required to achieve 50% aggregation (EC_{50}) in the presence or absence of aqueous tomato extract being 1.05 and 0.34 µg/mL, respectively.



These data demonstrate that an aqueous tomato extract can inhibit platelet aggregation *in vitro*, and may provide an explanation for the cardioprotective effects of tomatoes observed in epidemiological studies. Further research is in progress to determine whether these effects can be demonstrated following intervention with tomato products.

References

1. Parfitt VJ, Rubba P, Bolton C, Marotta G, Hartog M, Mancini M. A comparison of antioxidant status and free radical peroxidation of plasma lipoproteins in healthy young persons from Naples and Bristol. Eur Heart J 1994; 15: 871–6.
2. Klipstein-Grobusch K, Launer LJ, Geleijnse JM, Boeing H, Hofman A, Witteman JC. Serum carotenoids and atherosclerosis. The Rotterdam Study. Atherosclerosis 2000; 148: 49–56.
3. Rissanen TH, Voutilainen S, Nyyssonen K, Lakka TA, Sivenius J, Salonen R, Kaplan GA, Salonen JT. Low serum lycopene concentration is associated with an excess incidence of acute coronary events and stroke: the Kuopio Ischaemic Heart Disease Risk Factor Study. Br J Nutr 2001; 85: 749–54.
4. Dutta-Roy AK, Crosbie L, Gordon MJ. Effects of tomato extract on platelet aggregation in vitro. Platelets 2001; 12: 218–27.

Supported by the University of Newcastle.
Key words: tomato, platelet aggregation, cardiovascular disease

Raw brown onion consumption reduces plasma triglycerides in pigs

NK Gabler[1], E Ostrowska[1], BG Tatham[1], SJ Sterling[2], RB Jones[2], DR Eagling[2], FR Dunshea[1]

Natural Resources and Environment, [1]Werribee, VIC, 3030
[2]Knoxfield, VIC, 3176

Organosulphur compounds present in garlic and onions have been shown to inhibit cholesterol synthesis in vitro and potentially reduce the risk of heart disease (1). Consumption of onions has been also inversely correlated to coronary heart disease mortality in man (2). The aim of the present study was to evaluate the potential health benefits of brown onions fed at two levels of intake, using the pig as a human model.

Fifteen female and fifteen male crossbred (Large White x Landrace) pigs (initial weight 24.4 ± 0.85 kg) were selected and pre-fed with a wheat-based control diet containing 25% (w/w) of total fat for three weeks. At the end of the three weeks period the pigs (39.9 ± 0.97 kg) were randomly allocated to one of the three dietary treatments and were maintained in individual pens throughout the study. The treatments consisted of brown onions fed at 16 or 40 g/kg body weight $(BW)^{0.75}$ and no onion. Onions were homogenised in a blender prior to being mixed with dry feed formulated to contain 19.5 MJ DE/kg, 8% (w/w) of canola oil and 15% (w/w) of tallow to simulate the saturated to unsaturated fatty acid ratio of a western human diet. Pigs were fed approximately 80% of ad libitum for 6 weeks. Blood samples were obtained by venipuncture three hours post-feeding at weeks 1, 2, 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_2 (TXB_2) and cell counts.

Onion g/kg	Female			Male				Significance[2]		
$BW^{0.75}$	0	16	40	0	16	40	SED[1]	O	S	SxOxD
TG (mmol/L)	1.02	0.87	0.83	1.02	0.81	0.94	0.071	0.032	0.81	0.31
TC (mmol/L)	2.40	2.33	2.32	2.21	2.38	2.75	0.130	0.30	0.44	0.22
HDL (mmol/L)	1.32	1.32	1.45	1.20	1.25	1.34	0.075	0.25	0.23	0.95
LDL (mmol/L)	1.78	1.81	1.98	1.59	1.62	1.82	0.103	0.23	0.077	0.91
Platelets (10^9/L)	419	353	419	498	421	449	30.8	0.14	0.061	0.59
PT (seconds)	13.1	13.1	13.3	13.0	13.3	13.4	0.16	0.15	0.62	0.82
APTT (seconds)	16.9	15.9	17.1	15.6	16.6	16.4	0.56	0.65	0.44	0.27
TXB_2 (ng/mL)	19.2	22.2	11.3	14.9	14.7	17.5	2.17	0.79	0.37	0.012

[1]SED – Standard error of differences of means for onion vs control pigs. For SED of the effect of sex divide by 1.06.
[2]O – Onion vs control pigs; S – Sex; SxOxD – Sex x onion x dose interaction.

Consumption of brown onions reduced plasma TG concentrations (15.8%, P = 0.032) regardless of sex and the amount of onion fed. TC, HDL and LDL levels were not affected by onion supplementation, while LDL concentrations tended (P = 0.077) to be higher in female compared to male pigs. While consumption of onions have been shown to have anticoagulant effects (3), blood clotting measures were largely unaffected by onion supplementation in pigs. Onion consumption did not elicit any overall inhibitory effect on serum TXB_2 concentrations, although there was a significant interaction between onions, sex and dose of onion (P = 0.012) such that TXB_2 was reduced at the highest dose of onion supplementation in females but not in males. In conclusion, consumption of raw brown onion is effective in lowering plasma lipid levels and hence may be of health benefit.

References

1. Liu L, Yeh YY. Inhibition of cholesterol biosynthesis by organosulfur compounds derived from garlic. Lipids 2000; 35: 197–203.
2. Hertog MG, Feskens EJ, Hollman PC, Katan MB, Kromhout D. Dietary antioxidant flavonoids and risk of coronary heart disease: the Zutphen Elderly Study. Lancet 1993; 342: 1007–1011.
3. Chen JH, Chen HI, Tsai SJ, Jen CJ. Chronic consumption of raw but not boiled Welsh onion juice inhibits rat platelet function. 2000; J Nutr: 130, 34–37.

Key words: onions, triglycerides, pigs

Effect of dietary supplementation of concentrated pomegranate juice on blood lipids in type 2 diabetic patients with hyperlipidemia

F Tahbaz, A Esmaill-Zadeh, I Gaeini

National Nutrition and Food Technology Research Institute, S Beheshti University of Medical Sciences, PO Box 19395/4741, Tehran, Iran

Previous epidemiologic studies suggest that a high intake of fruits and vegetables is associated with a reduced risk of coronary heart disease (1). The beneficial effect could be related to minor components, especially flavonoids, which are proposed to exert their action by inhibiting LDL oxidation and platelet aggregation (2). Pomegranate is native to Iran and it is a rich source of flavonoids. The potential cardio protective effects of pomegranate juice have rarely been investigated in humans. In healthy men, intake of 50 ml pomegranate juice/d for 2 weeks decreased LDL susceptibility to aggregation and retention and increased the activity of serum paroxonase by 20% but didn't have any significant effect on the plasma lipid profile (3).

The present study was undertaken to determine whether concentrated pomegranate juice (CPJ) beneficially alters plasma lipid concentrations in type 2 diabetic patients with hyperlipidemia (cholesterol or triglyceride>200 mg/dl). For this reason 22 patients with type 2 diabetes without any other chronic diseases, were recruited from Iranian Diabetes Society and signed the consent form to participate in this study. These patients were instructed to follow the American Heart Association step I lipid-lowering diet for 8 weeks before CPJ consumption. In this pre-study period a 24-hour food recall and a food record (containing flavonoids-rich foodstuffs) were completed every 10 days. At the end of week 8 (baseline), anthropometric measurements were performed by standard methods and biochemical indices including total cholesterol, HDL cholesterol and triglyceride were measured (Zist-Chem, Tehran, Iran). LDL cholesterol was found using the Freidwald equation, and total cholesterol/LDL cholesterol and LDL cholesterol/HDL cholesterol were calculated. Thereafter the patients consumed 40gr CPJ for next 8 weeks. CPJ was provided from Nariran Inc and was free from added sugar and any other additive. Similarly during this period, dietary intake was assessed on a regular basis every 10 days. At the end of this period anthropometric measurements and blood indices were re-evaluated. These results were compared with the baseline by Wilcoxon-signed test. P-value was considered significant at $P < 0.05$.

There were 14 women (63.6%) and 8 men (36.4%) in this study. Mean (±SD) age, weight and duration of diabetes were 52.5 (±5.2) years, 71.5 (±10.3) kg and 7.9 (±6.6) years, respectively. After consumption of PJC significant reductions were seen in total cholesterol ($P < 0.006$), LDL-c ($P < 0.006$), LDL-c/HDL-c ($P < 0.001$) and Total-c /HDL-c ($P < 0.001$). But there were no significant changes in triacylglycerol and HDL-c. Weight, level of physical activity and the intake of nutrients and flavonoid-rich foodstuffs did not change during CPJ supplementation period. This study showed that CPJ supplementation could modify heart disease risk factors in this group of type 2 diabetic patients. Therefore, its inclusion in their diets might be beneficial.

Variable	Baseline*	After CPJ supplementation*
Total cholesterol (mg/dl)	202.4 ± 27.7	191.4 ± 21**
LDL cholesterol (mg/dl)	124.4 ± 31.9	112.9 ± 25.9**
HDL cholesterol (mg/dl)	38.2 ± 8.1	38.7 ± 7.7
Total-c /HDL-c	5.5 ± 1.3	5.09 ± 1.1**
LDL-c/HDL-c	3.4 ± 1.2	3 ± 0.9**
Triacylglycerol (mg/dl)	198.5 ± 57.5	195.4 ± 52.8

*$\bar{X}$ ± SD ** Significantly different from baseline, $P < 0.05$ (Wilcoxon-Signed test).

References

1. Bors W, Heller W, Micheal C, Saran M. Flavonoids as antioxidants: determination of radical scavenging efficiencies. Methods Enzymol 1990; 186: 343–55.
2. Cook NC, Samman S. Flavonoids-chemistry,metabolism,cardioprotective effects and dietary sources. J Nutr Biochem 1996; 7: 66–76.
3. Aviram M, Dornfeld L, Roseblat M, Volkova N, Kapian M, Coleman R, Hayek T, Fuhrman B. Pomegranate juice consumption reduces oxidative stress, atherogenic modifications to low density lipoprotein and platelet aggregation: studies in human and in atherosclerotic apolipoprotein E-deficient mice. Am J Clin Nutr 2000; 71: 1062–76.

Key words: concentrated pomegranate juice, type 2 diabetes, hyperlipidemia

Improvement in plasma lipid levels (including lipoprotein (a)) after chronic soy consumption may be linked to equol

BJ Meyer, TA Larkin, AJ Owen, LB Astheimer, LC Tapsell and PRC Howe

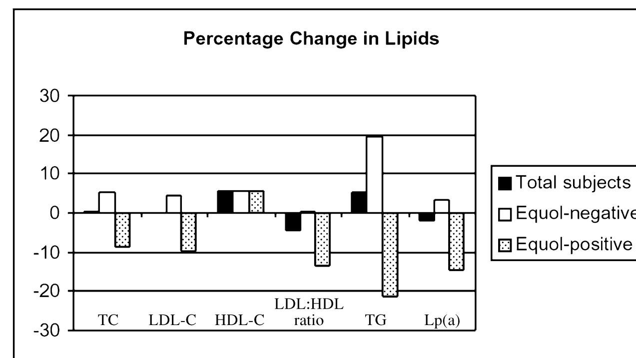
Smart Foods Centre and Department of Biomedical Science, University of Wollongong, NSW, 2522.

Dietary interventions with soy have been inconsistent, with some studies demonstrating a hypocholesterolaemic action while others have not. We examined the effect of the consumption of soy milk and soy yoghurt produced from whole soy beans (containing soy protein, isoflavones and polyunsaturated fatty acids) on lipid and cardiovascular characteristics in men and women having moderate cardiovascular risks.

The study design was a placebo controlled randomised, crossover diet intervention trial that compared the regular consumption of soy-based with a dairy-based (control) intervention. Twenty-six mildly hypercholesterolaemic (average total plasma cholesterol of 6.0 mmol/L) or mildly hypertensive volunteers were randomly assigned to one of two groups consuming a regular diet incorporating 4 serves per day of either soy or dairy foods for 5 weeks, after which the diets were reversed for a further 5 weeks. The soy diet provided at least 25 g of soy protein and 80mg of isoflavones per day. Clinical assessments included dietary interviews, height, body mass, clinic and 24 hour ambulatory blood pressure, arterial compliance and a fasting blood sample. Plasma lipids, fatty acids, and isoflavones, and 24 hour urinary isoflavone excretion were measured initially and after each 5 week period.

Following the soy intervention, plasma and urinary isoflavone levels increased 23 and 6 fold, respectively, for the 23 subjects completing the study. Polyunsaturated fat intake doubled with the consumption of the soy diet. The total amount of dietary fat consumed did not change. However, the plasma P:S ratio increased significantly with consumption of the soy diet but not with the dairy diet. Despite the large increases in isoflavone levels the consumption of the soy products had no significant effect on plasma lipids, blood pressure or arterial compliance compared with consumption of dairy products.

However, when subjects were retrospectively grouped as equol positive (n = 8) or equol negative (n = 15) based on whether equol was detected in their plasma or urine, we observed highly significant reductions in total cholesterol (8.5%), LDL-cholesterol (10%), LDL:HDL ratio (13.5%), plasma triglyceride (21%) and lipoprotein (a) (14%) after the 5 weeks of soy consumption in the equol positive group ($P < 0.05$). This effect in equol positive subjects appeared independent of any macronutrient changes.



In conclusion regular consumption of whole soy bean milk and yoghurt products resulting in raised circulating levels of isoflavones and PUFAs. However, this resulted in improved plasma lipids only in the equol-positive subjects.

This study was supported by So Natural Foods Australia.
Key words: soy isoflavones, cholesterol, equol.

Mediterranean and low fat diets are associated with similar lipid levels at 1 year in patients with coronary heart disease on statin therapy

DM Colquhoun

Core Research Group, University of Queensland, Brisbane, QLD, 4066

Background: A low fat (LF) diet is usually recommended for patients with coronary heart disease (CHD). A high fat Mediterranean (Med) diet, rich in monounsaturates (MUFA) is associated with low CHD. Relative efficacy of diets for patients on statins is unknown. This study compared LF to Med diets on lipids and lipoproteins in patients with CHD on standard therapy (including statins).

Method: 68 patients with angiographic CHD were randomised to LF (fat 20–25% energy (E), saturated fat (SFA) 8–10% E0 or Med (fat 35–40% E, >50% MUFA). Lipids were measured prior to drug therapy, at randomisation and 3 and 12 months.

Results: 86% LF patients and 80% Med patients were on statins. Similarly, 80% LF and 85% Med patients were taking aspirin. Mean fat intake in LF diet was 20% of total E (SFA 8.5% of total E) compared to Med diet with fat 34% of total E (57% MUFA).

	Chol	Trig	HDL-C	LDL-C
Pre statin	6.59	2.81	1.18	3.92
LF 12 Months	4.42	1.56	1.21	2.52
Med 12 Months	4.52	1.48	1.24	2.62

Conclusion: Med and LF diets are associated with similar lipid and lipoprotein levels at 1 year in patients with CHD on standard therapy. Dietary recommendations for CHD patients ought to consider a Med style diet in addition to a LF diet.

Key words: Mediterranean, lipid, CHD

Omega 3 LCPUFA do not inhibit the growth of preterm or term infants. A meta analysis

RA Gibson, K Ried, M Makrides

Child Nutrition Research Centre, Child Health Research Institute, Women's and Children's Hospital, and Flinders Medical Centre, Adelaide, SA

Long-chain polyunsaturated fatty acids (LCPUFA) occur naturally in breast milk and have been added to some infant formulas in forms that are not identical to those in breast milk. There have been few studies specifically designed and with sufficient power to assess the safety of these supplements. While the beneficial effects of dietary LCPUFA supplementation on early visual development to preterm and term infants have been demonstrated in several international clinical trials, the influence on the infant's growth performance has not been as clear. Early studies on preterm infants in the US have aroused international concern with the apparent association of omega-3 LCPUFA supplemented formulas and an inhibition of growth. The goal of our project was to summarise and evaluate 10 years of intensive clinical research on LCPUFA supplementation in infant nutrition and to bring clarity into the ongoing debate on its safety. This was achieved by means of a systematic review and a meta-analysis of growth measures from all published trials.

Thirteen randomised trials involving 1279 term infants met our criteria for inclusion in the meta-analysis. Infants allocated to LCPUFA supplementation were 90 g lighter (95% confidence interval, CI, –0.14, –0.04), 0.32 cm shorter (95% CI, –0.55, –0.08) and had a smaller head circumferences (–0.18 cm, 95% CI, –0.32, –0.04) at birth compared with infants allocated to the control groups. This difference, prior to randomisation, was explained by the fact the infants allocated to the LCPUFA groups were born two days earlier than infants in the control groups despite that fact that all infants were at least 37 weeks gestation at birth. Most importantly there were no significant differences in weight, length or head circumference of infants at either four or twelve months of age.

Eleven randomised trials involving preterm infants met our criteria for inclusion in the meta-analysis but data was only available from nine trials. 1097 infants were involved in these nine trials. The biggest predictors of growth at 40 and 57 weeks post-menstrual age (PMA) for these infants were their size at birth, sex and whether they received a nutrient enriched (protein and micronutrients) preterm formula. The influence of LCPUFA supplementation was small and did not reach statistical significance.

It therefore appears that LCPUFA supplementation of both preterm and term infant formulas do not influence growth of infants.

Supported by the Financial Markets Foundation for Children, Wyeth Nutritionals International, NH&MRC
Key words: long chain fatty acids, growth, infants

New Zealand green lipped mussel (NZGLM) oil can reduce pro-inflammatory eicosanoids and cytokines and oxidation markers *in vivo*

KJ Murphy[1], M Kiely[2], K Galvin[2], PA Morrissey[2], NJ Mann[1] and AJ Sinclair[1]

[1]*Department of Food Science, RMIT University, VIC, 3001*
[2]*Department of Food Science, Nutrition and Technology, University College Cork, Ireland*

Arthritis is a chronic inflammatory disease, lacking an adequate therapeutic treatment with minimal side effects. Therapy has traditionally involved the use of non-steroidal anti-inflammatory drugs which are often accompanied by severe side effects. Clinical and animal studies have demonstrated oils rich in marine derived omega-3 polyunsaturated fatty acids (n-3 PUFA) can reduce the production of eicosanoids and cytokines associated with the inflammatory response and can reduce lipid peroxidation (1, 2). Past research has shown that the lipids of the NZGLM possess equal or greater anti-inflammatory activity than regular n-3 PUFA rich fish oils (3). The aim of this study was to compare the efficacy of the NZGLM oil in comparison with a regular fish oil rich in n-3 PUFA, in reducing markers of inflammation and cardiovascular disease risk factors. This was a double blind, randomised, parallel study, with a six week dietary intervention and a two week washout period following supplementation. Twenty eight healthy subjects were randomly assigned to consume either 2 mL/day of the NZGLM oil containing 241 mg n-3 PUFA or 2 mL/day of fish oil containing 181 mg n-3 PUFA. Subjects gave fasting blood samples at day 0, day 21, 42 and 56. Dietary restrictions were implemented to control the dietary intake of n-3 PUFA from other sources. Blood was analysed for neutrophil phospholipid fatty acids and serum levels of thromboxane B_2 (TXB_2). Stimulated monocyte production of prostaglandin E_2 (PGE_2), interleukin-1β (IL-1β) and tumor necrosis factor α (TNFα) were also measured. Lipid oxidation was assessed by measuring low density lipoprotein (LDL) concentration of cholesteryl esters and cholesteryl linoleate hydroperoxide (Ch18:2n-6-OOH). Plasma antioxidant status was assessed using the ferric reducing antioxidant power (FRAP) assay, tocopherol, retinol and carotenoids.

Following six weeks of supplementation, both groups showed a small, but significant increase in neutrophil phospholipid content of eicosapentaenoic acid and docosahexaenoic acid ($P < 0.05$), a significant reduction in serum TXB_2 and a significant reduction in endotoxin stimulated monocyte production of PGE_2 and IL-1β in subjects with high baseline levels ($P < 0.05$). There was an increase in plasma antioxidant status in both treatment groups ($P < 0.05$) and a trend to decrease LDL Ch18:2n-6-OOH and free cholesterol at day 42 compared with day 0 in both groups. These results are in agreement with past research (1–4), however the novel aspect of this study is that the dose of long chain n-3 PUFA was substantially lower than in many other studies. These data provide additional biochemical evidence that the NZGLM oil is anti-inflammatory *in vivo*, and could aid in reducing the symptoms associated with arthritis. It is possible that the marine oils reduced the level of these eicosanoids and cytokines by inducing a shift in cyclooxygenase and lipoxygenase substrate specificity, reducing eicosanoid and cytokine synthesis, inhibiting the expression of cell surface adhesion molecules and modulating gene transcription and peroxisome proliferator activated receptor activation.

References

1. Lorenz R, Weber PC, Szimnau P, Heldwein W, Strasser T, Loeschke K. Supplementation with n-3 fatty acids from fish oil in chronic inflammatory bowel disease—a randomized, placebo-controlled, double- blind cross-over trial. J Intern Med Suppl 1989; 225: 225–32.
2. Higgins S, McCarthy SN, Corridan BM, Roche HM, Wallace JMW, O'Brien NM, *et al.* Measurement of free cholesterol, cholesteryl esters and cholesteryl linoleate hydroperoxide in copper-oxidised low density lipoprotein in healthy volunteers supplemented with a low dose of n-3 polyunsaturated fatty acids. Nutrition Research 2000; 20: 1091–1102.
3. Whitehouse MW, Macrides TA, Kalafatis N, Betts WH, Haynes DR, Broadbent J. Anti-inflammatory activity of a lipid fraction (Lyprinol) from the N.Z. green-lipped mussel. Inflammopharmacology 1997; 5: 237–246.
4. Kesavulu MM, Kameswararao B, Apparao C, Kumar EG, Harinarayan CV. Effect of omega-3 fatty acids on lipid peroxidation and antioxidant enzyme status in type 2 diabetic patients. Diabetes Metab 2002; 28: 20–6.

Key words: NZGLM, eicosanoid, anti-inflammatory

Nutrition in inflammatory disease: what is the evidence?

[1]DH Volker, [1]T Katz, [1]LKC Shui, [1]P Quaggiotto, [2]M Garg, [3]GAC Major

[1]*Human Nutrition Unit, Dept of Biochemistry, University of Sydney, NSW, 2006*
[2] *Department of Nutrition & Dietetics, University of Newcastle, NSW, 2308*
[3]*The Royal Newcastle Hospital, Newcastle, NSW, 2300*

Objective: To determine the efficacy of fish oil derived (n-3) fatty acid supplementation in subjects with osteoarthritis (OA) and rheumatoid arthritis (RA) compared to placebo.

Methods: Two placebo controlled, double blind, randomised studies were conducted to determine the effect of supplementation on lipid biomarkers and clinical variables in OA and RA patients[1] (n = 24 and n = 26 respectively). Fish oil and placebo were supplemented at a rate of 40 mg/kg body weight/day for 15 weeks in the RA study and 20 weeks in the OA study. Placebo for the RA subjects was a corn oil, olive oil mix and evening primrose oil for OA subjects. Eligibility protocol for OA and RA subjects required a background diet that contained <15 g n-6 fatty acids/day and low saturated fat intake (<33% of total fat intake), active disease, with stable medication for a minimum of 3 months. Background diet fatty acid content was determined by food frequency questionnaire and dietary compliance by 24 hour recall.

Results: Analysis of lipid biomarkers demonstrated a significant increase in n-3 fatty acids in phospholipid and plasma fatty acids in the supplemented RA and OA groups. Analysis of clinical variables in the RA group indicated a non-significant improvement in tender joint counts (TJC) and reduced duration in early morning stiffness (EMS). The OA group demonstrated a significant improvement in TJC and a reduced duration of EMS ($P < 0.05$).

Conclusions: These findings suggest that fish oil supplementation at an n-3 fatty acid dose of 40 mg/kg of body weight/day, in conjunction with a diet low in n-6 and saturated fat leads to a significant incorporation of n-3 fatty acids in the plasma fatty acids and phospholipids and improvement in clinical variables in Australians with inflammatory diseases such as OA and RA. These studies suggest the need for a nutritional regimen in diseases such as OA and RA.

Clinical Variable	RA[1] Intervention	RA[1] Placebo	OA[1] Intervention	OA[1] Placebo
Tender Joint Count (0 wk)	6.0 ±1.5	9.0 ± 1.2	5.0 ± 1.6	4.0 ± 1.2
Tender Joint Count (15/20 wk)	4.0 ±1.2	8.0 ± 2.3	2.0 ± 0.6	4.0 ± 0.9
Early morning Stiffness Hrs (0 wk)	1.2 ±0.5	1.5 ± 0.9	3.1 ± 1.9	0.32 ± 0.1
Early morning Stiffness Hrs (15/20 wk)	0.8 ±0.1	2.2 ± 1.2	0.9 ± 0.1	0.54 ± 0.3

[1] mean ± SEM.

Reference

1. Volker DH, FitzGerald PEB, Major GAC, Garg ML. Efficacy of fish oil concentrate in the treatment of rheumatoid arthritis. J Rheumatol 2000; 27: 2343–6.

Is there a relationship between α-linolenic acid and prostate cancer?

NM Attar-Bashi, D Li, AJ Sinclair

Department of Food Science, RMIT University, VIC, 3001

A number of studies have suggested there is a positive association between diet or plasma α-linolenic acid levels and the incidence of prostate cancer (1, 2), however other studies have not supported this association (3, 4).

The lack of consistency in findings from these studies might be due to a number of reasons including methodological limitations related to measurement error in estimating past dietary exposure of a nutrient like α-linolenic acid from food frequency questionnaires, measurement of α-linolenic acid levels in plasma which might not represent long-term dietary intake, the assumption that α-linolenic acid levels in plasma is representative of α-linolenic acid levels in the prostate tissue, and the sample size of the studies which might have been too small in some studies.

To shed light on the relationship between fatty acid exposure and the invasiveness and metastatic potential of prostate cancer, one study (4) examined prostatic levels of individual fatty acids in relation to histopathological characteristics of cancer in men undergoing radical prostatectomy for localised disease. The results of the study showed that prostatic α-linolenic acid levels tended to be lower in cases than in control subjects with significantly lower levels when tumours extended to an anatomical or surgical margin. Measuring prostatic levels of fatty acids, as in this study, offers advantages over self-reported usual dietary intake, since it provides an estimate of exposure at the target organ level, where the concentrations likely reflect long-term dietary intake.

We have been investigating the effect of α-linolenic acid and other fatty acids on the growth of PC-3 human prostate cancer cell lines. Concentrations of fatty acids tested ranged between 10-200 μg/mL. Cells were plated out at a density of 10^4/100 μl/well in a 96-well plate and allowed and overnight period for attachment. Different concentrations of fatty acids were then added in parallel and incubated for 24, 48, or 72 hours in a 37^0C and 5% CO_2 incubator. Effects of fatty acids on cells was determined using a colorimetric MTT (3-4,5-dimethylthiazol-2-yl-2,5-diphenyl-tetrazolinm bromide) assay. The results showed that α-linolenic acid and other polyunsaturated fatty acids (PUFA) such as DHA and EPA significantly ($P < 0.05$) reduced the growth rate of the tumour cells at the high concentrations. Current studies are looking at mechanisms of fatty acids effects on tumour cells which might include fatty acid peroxidation, where spontaneous oxidation of PUFA yields reactive aldehydes and other products of lipid peroxidation that are potentially toxic to cells and may also signal apoptosis. Also fatty acids can exert their effect by interfering with the lipoxygenase enzyme activity. The present results indicate that PUFA, including ALA, are effective at reducing the growth of PC-3 human prostate cancer cell lines However, mechanisms of action are not yet understood and require further investigation.

References

1. Giovannucci E, Rimm EB, Colditz GA, Stampfer MJ, AscherioA, Chute CC, Willett WC. A prospective study of dietary fat and risk of prostate cancer. J Natl. Cancer Inst. 1993; 85: 1571–1579.
2. Gann PH, Hennekens CH, Sacks FM, Grodstein F, Giovannucci EL, Stampfer M. Prospective Study of plasma fatty acids and risk of prostate cancer. 1994; 86: 281–286.
3. Schuurman AG, van den Brandt PA, Dorant E, Brants HA, Goldbohm RA. Association of energy and fat intake with prostate carcinoma risk: results from The Netherlands Cohort Study. Cancer. 1999; 86: 1019–27.
4. Freeman V, Meydani M, Yong S, Pyle J, Flanigan R, Waters B, Wojcik E. Prostatic levels of fatty acids and the histopathology of localized prostate cancer. The Journal of Urology 2000; 164: 2168–72.

Key words: prostate cancer, α-linolenic acid

Low-dose iron supplements in pregnancy prevent iron deficiency at the end of pregnancy and during the post-partum period: the results of a randomised controlled trial

M Makrides[1], CA Crowther[2], RA Gibson[1], RS Gibson[3], CM Skeaff[3]

[1]*Child Health Research Institute and Adelaide University Dept Paediatrics, Women's & Children's Hospital, North Adelaide, SA, 5006*
[2]*Adelaide University Dept Obstetrics & Gynaecology, Women's & Children's Hospital, North Adelaide, 5006*
[3]*Department of Human Nutrition, University of Otago, Otago, New Zealand*

Context: Iron deficiency anaemia (IDA) is common in pregnant women in industrialised countries and previous trials aimed at preventing IDA have used high-dose iron supplements that are known to cause gastrointestinal side effects. Current clinical practice in Australia is to screening for anaemia at 28 weeks gestation and treat if detected. There have been no reports to systematically assess the benefits and tolerance of routine low-does iron supplementation in pregnancy.

Objective: To assess the effect of supplementing the diet with 20 mg/d iron, a low-dose designed to meet the recommended intake during pregnancy on maternal iron deficiency (ID, without anaemia), IDA, and gastro-intestinal side effects.

Design: Randomised, double blind, controlled trial conducted December 1997 – October 1999 with a follow-up to six months post-partum.

Setting: Maternity hospital in Adelaide, Australia.

Participants: 430 women with singleton or twin pregnancies, without pre-existing anaemia and not taking iron supplements. 386 (89.7%) women completed the follow-up to six months post-partum.

Intervention: 20 mg daily iron supplement (ferrous sulphate) from 20 weeks gestation until birth.

Main outcome measures: Maternal IDA and ID at the end of pregnancy and at six months post-partum. Gastro-intestinal side effects assessed via questionnaire at 24 and 36 weeks of gestation.

Results: At the end of pregnancy, fewer women from the iron supplemented group had IDA than the placebo group (6/198, 3% vs 20/185, 11%; relative risk, RR 0.28, 95% confidence interval, CI, 0.12, 0.68, $P < 0.005$) and fewer iron supplemented women had ID than placebo treated women (65/186, 35% vs 102/176, 58%; RR, 0.60, 95% CI, 0.48, 0.76, $P < 0.001$). There were no differences between the groups in the numbers of women reporting any gastrointestinal side effects. At six months post-partum fewer women from the iron group had ID compared with the placebo group (31/190, 16% vs 51/177, 29%; RR 0.57, 95% CI 0.38, 0.84, $P < 0.005$). The rate of IDA between the groups did not differ.

Conclusion: Supplementing the diet of women with 20 mg of iron daily from 20 weeks of pregnancy offers a low risk strategy to prevent IDA and ID.

Supported by the NH&MRC, Channel 7 Medical Research Foundation, Gunn & Gunn Research Foundation, Perinatal Pathology Fund and Soul Pattinson Manufacturing.
Key words: iron, pregnancy, iron deficiency

Dietary strategies in the management of diabetes

J Brand-Miller

Human Nutrition Unit, University of Sydney, NSW, 2006

Diet is always said to be the 'cornerstone' of management of diabetes, yet the recommended dietary guidelines remain controversial and relatively few patients succeed in being well-controlled on diet alone. This implies that dietary treatment is not sufficient in itself or otherwise too difficult or even counterproductive. Many experts argue against the current dietary recommendations for diabetes, with both the quantity and quality of carbohydrate being at the centre of the controversy. This presentation is designed to critically address the issues of how much and what type of carbohydrate should be recommended for people with diabetes. It takes an evidence-based approach, giving greater weight to randomised controlled intervention studies.

Important questions that need to be addressed include: What is the scientific basis for recommending high carbohydrate diets? What are their potential adverse effects? What is the evidence for recommending diets high in monounsaturated fat (MUFA) instead? Are low glycaemic index (GI) diets superior to high MUFA diets? What is the optimal diet for improving insulin sensitivity? Is this different to the optimal diet for weight loss?

Prior to 1970 the prescribed diet for diabetes was low in carbohydrate (5–40% energy) because of its obvious role in raising blood glucose levels. But during the 1970s, a spate of studies showed that high carbohydrate-*very* high fibre diets (containing large amounts of legumes and wholegrains), compared to the high fat diets traditionally recommended, not only improved glycemic control but also improved insulin sensitivity. As a result, most diabetes associations around the world began to recommend high-carbohydrate (>55% energy) diets that were low in saturated fat and 'high' in fibre for people with diabetes (American Diabetes Association (ADA), 1987; Diabetes and Nutrition Study Group of the European Association for the Study of Diabetes, 1988). Unfortunately, the high fibre criterion was seen as not critical to glycemic control and the amounts recommended were only marginally higher than that generally consumed.

Not surprisingly, during the 1990s, studies began to appear showing that high carbohydrate diets containing only moderate amounts of fibre had adverse effects on blood triglyceride (TG), HDL-cholesterol and fasting glucose levels when compared with high-fat diets enriched with MUFA. Since people with diabetes are at high risk of cardiovascular disease, the findings were taken very seriously and in 1995 the ADA approved high MUFA as part of individualised dietary management. However, high MUFA diets have *not* been shown to reduce the most important measure of long-term diabetes control, ie glycated hemoglobin. Furthermore, high MUFA diets with more than 38% energy as fat have been found to be associated with insulin resistance. There is also concern that the energy dense nature of any high fat diet may predispose to weight gain.

Low GI diets for the management of diabetes have also been controversial. They are promoted on the basis that they allow a high carbohydrate intake with the least effect on postprandial blood glucose levels and without having to be exceptionally high in fibre. A recent meta-analysis comprising 14 studies in people with type 1 and type 2 diabetes (1) showed that low GI diets reduce glycated hemoglobin by 7.4%, a level comparable to many oral hypoglycaemic agents and superior to that of expensive insulin analogues. Low GI diets have been embraced in Australia and parts of Europe but not the United States where the concept is considered complex and another burden for people with diabetes.

The future is likely to see the percentage of carbohydrate in the diabetic diet 'individualised' to increase compliance and take account of usual food habits. Emphasis on changes in the *types* of carbohydrate foods and *types* of oils and margarines may be more important to overall diabetes control than the amount of carbohydrate vs fat *per se*.

Reference

1. Brand-Miller J, Colagiuri S, Hayne S, Petocz P. A meta-analysis of high vs low glycaemic index diets in diabetes. Proc Nutr Soc Aust 2002; 26: in press.

Nutrient-drug synergies to optimise therapeutic benefit

PRC Howe

Department of Physiology, University of Adelaide, SA 5005 and School of Pharmaceutical, Molecular and Biomedical Sciences, University of South Australia, Adelaide SA 5001

Food sources of bioactive nutrients may offer therapeutic benefits which are equally if not more important than their health promoting properties; however food regulations constrain therapeutic applications. The emergence of functional foods with considerable therapeutic potential, such as cholesterol-lowering spreads containing plant sterols, challenges this regulatory position.

With the growing incidence of diet-related chronic diseases, bioactive nutrients and functional foods can and should play a central role in therapy. Even if medication is available and effective, price subsidies can be a significant economic burden. Nutritional and lifestyle interventions offer consumers greater personal control of their health. Moreover, they have the potential to augment drug therapy, thus reducing dose requirements and possible side effects.

A clear example of nutrient-drug synergy is the ability of dietary sodium restriction to potentiate blood pressure reduction by most antihypertensive drugs (1). Unfortunately sodium restriction is poorly utilised as a primary intervention, let alone promoted as adjunct therapy. Trials such as DASH and DASH-sodium reaffirm the antihypertensive efficacy of decreasing the dietary intake of sodium relative to potassium. Adoption of this food-based strategy by the 13% of Australians over 25 who are treated for hypertension could significantly reduce their $700 million drug bill!

Apart from potentiating drug efficacy, nutrient supplementation can offer broader risk benefit. An example is the ability of omega-3 supplementation (ω3) to further reduce blood pressure in hypertensives treated with diuretics or β-blockers and, in addition, counteract adverse effects of these drugs on plasma lipids (2). Interactions with other nutrients may also influence the overall therapeutic effect, e.g. sodium restriction can enhance the antihypertensive effect of ω3 (2). The impressive benefit of ω3 in secondary prevention of coronary disease (GISSI-P trial) may be at least partly attributed to interaction with aspirin, producing novel antiinflammatory agents (3).

Other possibly beneficial combinations include plant sterols or soy protein with statins for the treatment of hypercholesterolaemia. Adding a cholesterol uptake inhibitor (sterol) to a synthesis inhibitor (statin) may seem obvious, yet few trials have been undertaken to substantiate this benefit (4) and, thus far, there has been no evaluation of soy in combination with statins, even though a U.S. health claim promotes soy protein for *prevention* of coronary heart disease.

Adjunct nutritional therapies could facilitate management of a wide range of chronic disorders. Unfortunately, however, there is little inducement to either evaluate or promote them in this role.

References

1. Beard TC. Salt in Medical Practice. 2nd ed. Holland Park: Queensland Hypertension Assoc. 2000.
2. Howe PR. Dietary fats and hypertension – focus on fish oil. Ann NY Acad Sci. 1997; 827: 339–52.
3. Serhan C, Clish C, Brannon J, *et al*. Novel functional sets of lipid-derived mediators with antiinflammatory actions generated from omega-3 fatty acids via COX2 nonsteroidal antiinflammatory drugs and transcellular processing. J Exp Med 2000; 192: 1197–1204.
4. Plat J, Mensink R. Effects of plant sterols and stanols on lipid metabolism and cardiovascular risk. Nutr Metab Cardiovasc Dis 2001; 11: 31–40.

The science/food industry/clinical interface

D Colquhoun

Associate Professor of Medicine, University of Queensland, Core Research Group, Wesley Medical Centre, QLD, 4066

Food choices are key factors in the causation of cardiovascular disease and many cancers. In stark contrast to the pharmaceutical industry, surprisingly there is very little traffic between the food industry and the general medical community.

The food industry includes small farmers to giant food processing companies like Nestle (food sales worldwide US$49.4 billion in 1999) and food service companies like McDonalds (sales in US$19.5 billion in 2000). In Australia and other developed countries production far exceeds demand and like other industries there is fierce competition at each level of the food industry. The nutrition and medical communities interact in many ways giving advice to industry regarding food product development (genetically modified food, nutraceuticals etc) and the promotion of foods or specific products and doing independent or commissioned research.

Withdrawal of government funding has resulted in greater reliance on industry for research and this influences what is investigated and how findings are presented (similar to pharmaceutical industry).(1) The interests of various food bodies may clash with nutritionists whose main concern is health and optimal nutrition. (2) In the USA lobbyists for certain food interests have successfully had politicians block publication of food guidelines and have them modified.(3)

The food industry has been successful in producing and delivering abundant food in Australia. To prevent lifestyle (largely good choice) disease we need to constructively engage the food industry to develop mutually respective partnership between academia, government and various levels of the food industry.

For:

(i) more research (basic and applied)

(ii) develop new products and promote healthy eating

(iii) support groups such as nutrition society and the National Heart Foundation (4–6)

In conclusion there needs to be far more activity between the food industry and the scientific community as our interests frequent coincide for the great health of our community.

References

1. Edwards G. Should industry sponsor research? BMJ 1998; 317: (7154).
2. Nestle M. Ethical dilemmas in choosing the healthful diet: vote with your fork! Proc Nutrition Soc 2000; 59: 619–29.
3. Nestle M. Food politics. University of California Press Berkeley LA 2002.
4. Inniss S, Bahlo E, Kardinaal A. Beyond implementation: the role of physicians, health care professionals and consumers in the development and use of food products. Eur J Clin Nutrition 1999; 53 (suppl): S25–9.
5. Asheley JM, Jarvis WT. Position statement of the American Dietetic Association: food and nutrition misinformation. J Am Diet Assoc 1995; 95: 705–7.
6. Crockett SJ. Food industry's role in national policy: working for the common good. J Am Diet Assoc 2002; 102: 478–9.

Resistant starch and health: from concept to products

David L Topping[1], Matthew K Morell[2], Roger A King[1], Anthony R Bird[1]

[1]*CSIRO Health Sciences and Nutrition, Adelaide, SA, 5000*
[2]*CSIRO Plant Industry, Black Mountain, ACT, 2600*

Improvements in diet and hence in public health are effected optimally through changes in the food supply rather than by individual action. Evidence for this comes from consideration of the increases in dietary fibre consumption by Australians over the past 20 years. Current intakes at the population level appear to be close to recommended and seem to be through foods available generally, not supplements (1). Of the benefits expected of fibre, improved laxation is well established but others (eg protection against colo-rectal cancer) are defined less well. It appears that starch which has escaped small intestinal digestion (resistant starch, RS) may be more important than fibre for large bowel health (2). RS appears to act through the products of its fermentation by the large bowel microflora. In children post weaning and in adults these are short chain fatty acids (SCFA). Among the benefits expected through greater SCFA supply are improved colonic blood flow and motility, lowered risk of colo-rectal cancer and improved fluid and electrolyte absorption. Butyrate is thought to be the most potent SCFA for many of these attributes although proof of a direct protective role for it in human colonic cancer is yet to be established. Some types of RS appear to favour butyrate production and there is interest in promoting RS consumption at the population level. This is a major challenge as Australian starch (and hence RS) intakes are low. In the short term it is impractical to raise total consumption for consumer acceptance and technical reasons. Attention has focussed on promoting the consumption of foods naturally high in RS eg navy beans and brown (whole grain) rice products. Consumption of these at normal serve sizes could make a meaningful contribution to RS intakes. However, caution needs to be exercised, as there is, as yet, no widely applicable method for measuring RS in foods and so estimating intakes by individuals or populations. Similarly, there are no data for the target figure for RS consumption although interventions have shown improved indices of bowel health with high RS foods (eg 3). Other studies have shown a practical route to enrich some foods with RS through the addition of a high amylose starch (4). Foods containing this product are available in Australia and elsewhere and have a range of documented effects including action as a prebiotic. A bowel health product which is a combination of psyllium and RS has also been developed. This product shows enhanced SCFA production compared with psyllium alone and is available commercially.

There seems to be an opportunity to enhance public health through increasing the consumption of foods high in RS. The approaches used so far are rather limited as the list of high RS food products is small and high amylose starches may have some technical limitations. CSIRO Plant Industry and Health Sciences and Nutrition have been collaborating on an alternative – a novel barley cultivar (*Barleyplus*™) which is under development. This is a non-GM mutant with a defect in starch synthesis leading to high levels of amylose with low amylopectin. The starch in this cultivar has a different genetic basis and distinct favourable properties when compared to alternative sources of high amylose starch. This confers resistance to amylolysis together with useful processing characteristics and offers the opportunity to produce a wide range of consumer-friendly products with enhanced nutritional attributes. Commercialisation of the novel barley is proceeding through collaboration within CSIRO and with external partners.

References

1. Baghurst PA, Baghurst KI, Record SJ. Dietary fibre, non-starch polysaccharides and resistant starch – A review. Food Aust 1996; 48: suppl S3–S35.
2. Topping DL, Clifton PM. Short chain fatty acids and human colonic function – Relative roles of resistant starch and non-starch polysaccharides. Physiol Rev 2001; 81: 1031–1064.
3. Topping DL, Illman RJ, Clarke JM, Trimble RP, Jackson KA, Marsono Y. Dietary fat and fiber alter large bowel and portal venous volatile fatty acids and plasma cholesterol but not biliary steroids in pigs. J Nutr 1993; 123: 133–143.
4. Brown IL, Mcnaught KJ, Moloney E. *Hi-maize*™: new directions in starch technology and nutrition. Food Aust 1995; 47: 272–275.

The link between nutritional science and food regulations/complementary medicine

DCK Roberts

Australian Food and Grocery Council, Canberra, ACT, 2604

Background: There is a continuum of risk associated with products regulated as foods and those regulated as medicines. These risks are associated with the composition/formulation of products and/or the uses/indications for which they are promoted. Regulation of foods involves managing whatever risks exist, with relation to quality and safety of the food and with relation to any claim about the food – is it truthful, will it mislead? The policy principles established for the regulation of health claims in Australia provides a sound risk-based management system to ensure a level of regulation commensurate with the low risk nature of most claims; and meet the need to improve market access to quality new products while maintaining public health and safety. The most important feature of a risk-based approach to regulation is the balance between pre-market evaluation or assessment of claims, and the on-going post-market monitoring and its enforcement.

Objective: To determine the appropriate evidence base for claims about foods and dietary supplements and the regulatory framework that delivers minimum effect regulation.

Outcomes: There are a number of evidence based guidelines available for assessing the quality and strength of claims that might arise from studies concerning food and nutrition. In the medicines arena, there are those promulgated by NHMRC (1) and those by the Complementary Medicines Evaluation Committee (CMEC) (2). There are draft proposals for an evidence guideline for foods by FSANZ (formerly ANZFA) (3). All of these have as a basic premise that the totality or balance of evidence should be supportive of the claim and that the quality of that evidence is high. Quality in the NHMRC guideline is based on the type of evidence that is available, thus a systematic review of a number of randomised control trials is taken to be the best and case reports, the weakest evidence. CMEC has adopted a similar best evidence guideline and has used lesser levels (medium and general) to support lower level claims for medicines. Within these levels lie the association type of evidence that can be obtained from strong quality epidemiological studies – ecological, case-control and cohort. In the foods area, it is impossible to obtain RCT type evidence for whole foods or diets, although it is possible for some food ingredients. What is far more common is strong quality evidence derived from epidemiological studies. FSANZ proposal for substantiation of health claims recognizes this and suggests a multilevel approach to evidence (A-G) with level C being cohort studies, while D is case-control. This retains the option for ingredients attaining a level A (systematic reviews etc) evidence status but accepts the best foods/dietary supplements can attain is good quality cohort studies. Given the likely nature of claims about foods being low risk and no more than the medium and general level seen for complementary medicines, the regulatory system should be the minimum necessary to ensure public health and safety, while allowing for innovation and timely market access. Pre-clearance for low risk claims should not be required, providing the manufacturer holds the appropriate evidence for the claim and can produce it for assessment on challenge.

Conclusions: A 'light touch' of regulation would permit low risk claims to proceed to market without pre-market assessment. Such a streamlined approach to assessment for low risk claims allows for timely market access. An important feature of this risk management approach is that the pre-market assessment is supported by appropriate post-market vigilance to ensure consumer confidence in the system.

References

1. http://www.health.gov.au/nhmrc
2. http://www.health.gov.au/tga/docs/html/tgaccevi.htm
3. http://www.foodstandards.gov.au/standardsdevelopment/inquiryreportproposalp153healthandrelatedclaims/section3amanagementf598.cfm

High fat diets do not increase CCl_4-induced oxidative stress in the rat

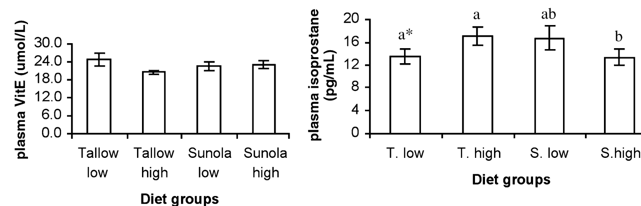
LK MacDonald-Wicks, ML Garg

Nutrition & Dietetics, School of Health Sciences, Faculty of Health, University of Newcastle, NSW, 2308, Australia

High fat diets have been associated with increased obesity and other chronic lifestyle diseases such as atherosclerosis and type II diabetes. High fat diets may promote atherogenesis by subjecting vascular endothelial cells to oxidative stress (1). Due to discoveries of potential relationships between oxidative stress, activation of oxidative stress-sensitive transcription factors and the etiology of acute and chronic diseases, the issue of the type of dietary fat and the amount of dietary fat consumed needs to be revisited (1). This study was conducted to examine the effect of high fat diets as saturated or monounsaturated fatty acids on oxidative stress.

Four groups of rats (n = 9) were fed either a high fat (200 g/Kg diet) or a low fat (50 g/Kg diet) diet based on Tallow (saturated) or Sunola (monounsaturated). The rats were fed the diet for four weeks, then gavaged with CCl_4 to induce oxidative stress. The rats were euthanised four hours later and plasma and liver collected for analysis.

Oxidative stress, as indicated by the plasma isoprostane concentrations (8-iso-$PGF_{2\alpha}$), were not affected by the increased fat in the diet. There was a significant decrease in plasma isoprostane values between the high fat Sunola and both the low fat and high fat Tallow diet groups ($p < 0.05$). However, isoprostane concentrations were unaltered by the amount and type of fat in the liver tissue. The increase in fat did not affect either plasma vitamin E, or liver vitamin E concentration. In combination these two parameters indicate that there was no increase in oxidative stress caused by the increase in the amount of fat in the diet regardless of the type of fat in the diet.



*The values without common superscript are significantly different (p<0.05)

In conclusion, there is no increase in oxidative stress due to an increase in amount of fat in the diet. It appears that when an increase in oxidative stress occurs it is probably due to the type of fat in the diet, not necessarily the amount of fat in the diet.

Reference

1. Hennig B, Toborek M, and McClain CJ. High energy diets, fatty acids and endothelial cell function: implications for atherosclerosis. J Am College Nutr, 2001; 20: 97–105.

Supported by a joint grant from Meadowlea Foods and GRDC
Key words: high fat diets, oxidative stress, F_2-isoprostane

Simultaneous measurement of tocopherols and carotenoids in oils using reversed-phase high-performance liquid chromatography

Q Su, M Daskalakis, NDH Balazs

Biochemistry Unit, Southern Cross Pathology Australia, Monash Medical Centre, Melbourne, VIC, 3168

Fresh vegetables and fruit are major sources of the antioxidant carotenoids, vitamin C and folate, while olive oil and other vegetable oils are the main source of vitamin E. Evidence is mounting for the potential protective role of such antioxidant vitamins and carotenoids in the development and progressions of cancer, occlusive vascular disease, diabetes, cataract formation and age-related macular degeneration. Absorption efficiency of carotenoids is known to be affected by formation of dietary fat, protein and bile salt concentrations. Various methods have reported determination of tocopherols in oils by using thin layer chromatography followed by spectrophotometry (1), gas chromatography (2) or HPLC (3). Several studies have reported simultaneous determination of α-tocopherol and β-carotene in olive oils (3,4).

We developed a rapid, direct HPLC method for simultaneous measurements of γ-tocopherol, α-tocopherol, lutein, lycopene, α-carotene and β-carotene in oils. Pretreatment of samples for these measurements was not required. The chromatographic system comprised a Waters 2690 separations module, 996 Photodiode Array Detector and a Spherisorb ODS-2 column (250 × 4.6 mm, 5 µm, 'Goldpak', UK). The mobile phase consisted of methanol-acetonitrile-chloroform and run as a gradient at 1.0mL/min. The methanol and acetonitrile contained 0.05% ammonium acetate and 0.1% triethylamine respectively. Run time was 26 minutes. α- and γ-Tocopherols and carotenoids were monitored at 292nm and 450nm respectively. The coefficients of variation (CV) were 6.5% for α-tocopherol, 3.4% for α-carotene and 5.9% for β-carotene. The detection limits were 0.8ng for carotenoids and 15ng for tocopherols. Oil samples were obtained from various sources.

Concentration of α–, and γ–tocopherols, lutein and β–carotene in oils (nmol/g)

Type of oil	γ–tocopherol	α–tocopherol	lutein	α–carotene	β–carotene	13–β–carotene	lycopene
Marine mussel oil	562	3079	5.1	16.9	32.1	32.8	22.8
Sunflower oil	60	3256	–	–	–	–	–
Extra light olive oil	9.5	957	–	–	1.2		–
Extra virgin olive oil	33	1016	0.1	–	–	10.4	–

The content of α- and γ-tocopherols and β-carotene in marine, olive and seed oils were very different. Sunflower oil had the highest levels of α-tocopherol and marine oil had higher γ-tocopherol, α-carotene, β-carotene and lycopene concentrations than all the other oils. The main advantage of the method described is its speed and the ability to simultaneously determine a number of lipid-soluble antioxidant compounds.

References

1. Ranalli A, Sgaramella A, Surricchio G. The new 'Cytolase 0' enzyme processing aid improves quality and yields of virgin olive oil. Food Chem 1999; 66: 443–454.
2. Giacometti J. Determination of aliphatic alcohols, squalene, alpha-tocopherol and sterols in olive oils: direct method involving gas chromatography of the unsaponifiable fraction following silylation. Analyst 2001; 126: 472–5.
3. Gimeno E, Calero E, Castellote AI, Lamuela-Raventoes RM, de la Torre MC, Lopez-Sabater MC. Simultaneous determination of alpha-tocopherol and beta-carotene in olive oil by reversed-phase high-performance liquid chromatography. J Chromatogr A 2000; 881: 255–9.
4. Su Q, Rowley KG, Itsiopoulos C, O'Dea K. Identification and quantitation of major carotenoids in selected components of the Mediterranean diet: green leafy vegetables, figs and olive oil. Eur J Clin Nutr (in press).

Acknowledgement: We thank Dr Kevin Rowley and Professor Andrew Sinclair for suggestions in preparation of this manuscript and the provision of the oils.
Key words: carotenoids, HPLC, measurement

Antioxidant tocopherol and carotenoid content of seed and marine oils

M Daskalakis, Q Su, JCG Doery, NDH Balazs

Biochemistry Unit, Southern Cross Pathology Australia, Monash Medical Centre, Melbourne, VIC, 3168

There are now numerous studies documenting the benefits of a Mediterranean diet, high in olive oil rather than saturated fats, for the prevention of atherosclerosis (1). The benefits of olive oil may be attributed to both monounsaturated fatty acid and their carotenoid content. The low mortality from cardiovascular disease (CVD) in Greek immigrants in Australia, despite a relatively high incidence of Type 2 diabetes and other recognised CVD risk factors (2), suggests a dietary protective mechanism which may be related to high olive oil intake. We have used a HPLC assay (3) for the measurement of tocopherols and carotenoids in two marine oils and 12 seed oils.

Results

a) Marine oils, which are rarely consumed by the public, have the highest concentration of carotenoids of all oils tested.
b) Sunflower oil has the highest tocopherol levels but no carotenoids.
c) Palm oil is very rich in β carotenes (but is not very palatable).
d) Family produced by traditional methods (non-commercial) olive oils has the highest carotenoid levels amongst the olive oils tested.

Concentration of tocopherols and carotenoids in various oils (nmol/g)

Oil brand	Vitamin E	Carotenoid antioxidant						
	γ-tocopherol	α-tocopherol	13-β-carotene	lutein	α-carotene	β-carotene	lycopene	total carotenoids*
Seed oils								
Caroteno palm oil (Malaysia)	7	768	1454	–	–	956	–	2433
Crisco peanut oil (Australia)	328	956	–	–	–	–	–	–
Crisco sunflower oil (Australia)								
Olive oils	60	3256	–	–	–	–	–	–
MVB oil (French)	44	524	3.0	2.0	–	–	–	5.0
Basso extra light (French)	31	1051	3.2	1.4	–	–	–	4.6
MVR oil (Spanish)	32	493	2.8	1.3	–	–	–	4.1
MORO oil (Spanish)	14	307	–	–	–	–	–	–
Bertoli extra light (Australia)	9	957	1.2	–	–	–	–	1.2
Vassiliou family 1998 (Greek)	20	836	1.5	2.6	–	–	5.9	9.3
Vassiliou family 1999 (Greek)	12	334	4.7	1.4	1.3	8.2	–	15.5
Vassiliou family 2000 (Greek)	6	688	1.9	0.9	–	–	3.4	6.2
Marine oils								
Marine mussel oil (Australia)	562	3079	32.8	5.1	16.9	32.1	10.8	97.6
Krill oil **(Australia)	76	492	2.2	2.0	1.6	1.7	3.3	10.8

* The sum of lutein, α–carotene, β–carotene, 13–β–carotene and lycopene; **from mutton bird; – not detected;

Conclusions

The health benefits of olive oil may be increased by their carotenoid antioxidant content, which varies considerably depending on source and manufacturing process. This information should be included on product labelling.

References

1. de Lorgeril M, Renaud S, Mamelle N, *et al*. Mediterranean alpha-linolenic acid-rich diet in the secondary prevention of coronary heart disease. Lancet 1994; 343: 1454–9.
2. National Heart Foundation of Australia, Heart and Stroke Facts. Canberra: National Heart Foundation of Australia 1996.
3. Su Q, Daskalakis M, Balazs NDH. Simultaneous measurement of tocopherols and carotenoids in oils using reversed-phase high-performance liquid chromatography (In preparing).

Key words: seed oil, marine oil, carotenoids

Increases in plasma lycopene concentrations change the antioxidant activity of the plasma as measured by ORAC but has no effect on two other *ex vivo* total plasma antioxidant assays

JM Fielding[1], R Stockmann[2], D Li[3], AJ Sinclair[1]

[1]*Dept of Food Science, RMIT University, Melbourne, VIC, 3001*
[2]*Food Science Australia, Werribee, VIC, 3030*
[3]*Dept of Food Science, Hangzhou University of Commerce, Hangzhou, China, 310035*

Antioxidants have been suggested to have a role in the prevention of cardiovascular disease and some cancers. Quantifying the action of antioxidants or the amount of oxidative stress of cells and tissues, before and after a dose is becoming of increasing interest in medical research. Methods have been developed to quantify the antioxidant capacity of total and fractionated plasma, such as the FRAP (ferric reducing antioxidant power), ORAC (oxygen radical absorbance capacity) and TBARS method (thiobarbituric acid reactive substances). This study investigated the effect of the addition of lycopene to total plasma, at biologically relevant concentrations, and the resultant *ex vivo* plasma antioxidant activity or production of pro-oxidants.

Lycopene (98% *trans* isomer, Hoffmann LaRoche, Switzerland; dissolved in DCM then nitrogen evaporated) was added to a pooled sample of human plasma (n=12), obtained at fasting, to give plasma concentrations between 0.28 and 1.87 µmol/L. The antioxidant capacity was measured by ORAC, a singlet oxygen assay (SOA) and lipid peroxidation was measured by TBARS, after 1 h and 24 h of incubation of the plasma at 37°C and 5% CO_2 (n = 6 at six different concentrations of lycopene). There was no change in the *ex vivo* antioxidant capacity or lipid peroxidation of the plasma at the 1 h and 24 h periods measured by TBARS (P = 0.179 and P = 0.369, respectively) and SOA (P = 0.338 and 0.311, respectively) at increasing lycopene concentrations in plasma. However, the ORAC assay showed a dose-dependant increase in the antioxidant capacity after 1 h (P = 0.002) of incubation but not after 24 h (P = 0.207).

Analysis	Incubation period (h)	R^2	Regression equation
ORAC	1	0.9216	y = 45.908x + 2384.7
ORAC	24	0.3600	y = 40.234 + 5274.1
TBARS	1	0.3983	y = -0.0041x + 1.4903
TBARS	24	0.0012	y = -5E-05x + 0.9254
SOA	1	0.2275	y = 0.0002x + 0.1222
SOA	24	0.2515	y = -0.0002x + 0.1144

We speculate that the lack of effect at 24 h was due to isomerisation of *trans* to *cis* lycopene under the incubation conditions, the failure of the exogenously added lycopene to partition into the appropriate plasma lipoprotein fractions (eg LDL) or the loss of other antioxidants in the plasma.

This data shows that addition of *trans*-lycopene to plasma across the range which could be encountered physiologically, leads to an increased antioxidant capacity but that there was no effect of the increased lycopene on lipid peroxidation or singlet oxygen quenching. This research suggests that some *ex vivo* antioxidant capacity assays may not be sufficiently sensitive for the prediction of antioxidant action *in vivo*.

This project was supported by funding from RMIT University and Food Science Australia
Key words: lycopene, antioxidant assays, ORAC

Evolutionary implications for human brain development and fatty acid intake

NJ Mann

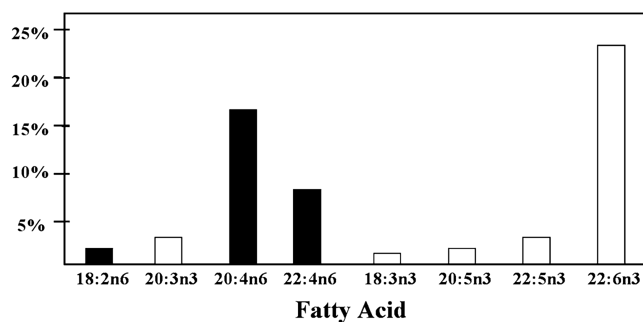
[1]*Department of Food Science, RMIT University, VIC, 3001*

With the emergence of the various species of the genus Homo at least 2–3 million years ago, a rapid increase in brain mass relative to body mass (encephalization) occurred (1). As humans fit the Kleiber equation for predictive value of resting metabolic rate (RMR) relative to body size (2), a reduction in the size and energy demand of another organ was necessary. The human gut is the only organ which shows a reduced size and energy demand relative to that expected for our body size (3). However a reduced gut size is only possible with a shift to a 'high quality' diet.

Tissue	Mass (Kg)		Metabolic	% Body
	Observed	Expected	increment	RMR
Brain	1.30	0.45	+9.5	16.1
Heart	0.30	0.32	-0.6	10.7
Kidney	0.30	0.24	+1.4	7.7
Liver	1.40	1.56	-2.0	18.9
GI tract	1.10	1.88	-9.5	14.8

The selective pressure that allowed for the increase in brain size is attributed to this improvement in dietary quality that involved both higher energy density and abundance of preformed long chain polyunsaturated fatty acids, such as docosahexaenoic acid (DHA) and arachidonic acid (AA), which dominate brain phospholipid (PL) composition as indicated in the Figure below (4).

To establish the likely range of foods leading to this process, the nutrient composition of a wide range of African ruminant tissues (brain, marrow, etc) freshwater fish, and edible wild plants were investigated. The richest source of DHA and AA was ruminant brain tissue (5). African field studies on carcass composition of large herbivores consumed by carnivores indicate that bone marrow (energy dense) and brain tissue were the items most likely left by carnivores. Hence these parts would be the most frequently available to prehistoric hominid scavengers. Freshwater fish most certainly would contribute adequate AA and DHA at sufficient levels for encephalization, however they would fail to meet hominid energy requirements (low energy density 119 kcal/100g). In conclusion it seems likely that evolving hominids consumed scavenged ruminant brain tissue as a rich source of AA and DHA and bone marrow as a principle energy source for the evolution of a large metabolically active brain.



References

1. Ruff CB, Trinkhaus E, Holliday TW. Body mass and encephalisation in Pleistocene Homo. Nature 1997; 387: 173–176.
2. Leonard WR, Robertson ML. Evolutionary perspective on human nutrition: the influence of brain and body size on diet and metabolism. Am J Hum Biol 1994; 6: 77–88.
3. Aiello LC, Wheeler P. The expensive tissue hypothesis. Curr Anthropol 1995; 36: 199–222.
4. Crawford MA. The role of dietary fatty acids in biology: their place in the evolution of the human brain. Nutr Rev 1992; 50: 3–11.
5. Cordain L, Watkins BA, Mann NJ. Fatty acid composition and energy density of foods available to Africanhominids. World Rev Nutr Diet 2001; 90: 144–161.

Do differences in the food supply explain discrepancies in epidemiology?

NJ Mann, AJ Sinclair and Y Yep

Department of Food Science, RMIT University, VIC, 3001

Epidemiological studies often show correlations within a population between specific food types and disease outcomes. However a considerable amount of variability exists between such studies in terms of level of significance (or relevance) of these correlations, particularly between different regional or national settings. These differing outcomes are often a result of three distinct shortcomings of epidemiological investigations involving diet: (i) Poor adjustment for intake (or lack of intake) of other dietary factors, (ii) Lack of adequate description or subgrouping of food types on dietary recall tools such as food frequency questionnaires, (iii) Food compositional differences at a regional or national level. The first limitation requires a case by case evaluation, however the latter two points are best illustrated by reference to a specific case, that of the ongoing debate over red meat consumption and colon cancer. For the situation regarding the recording of food intake, the recall tools are usually 'blunt' instruments that collect simplified information from which unsubstantiated conclusions are made. For instance frequency data is collected on 'meat' intake (often without quantities) where the term 'meat' includes: lean meat, fatty meat cuts and dishes and meat products (with added fat, food additives and other food types). In the case of national food compositional differences, red meat can have a broad range of nutrient, contaminant and fat content and type. The US situation from which many of these studies originate involves dramatic differences in animal feeding regimes, meat composition and preparation to Australia.

For instance the average selvedge fat on retail Australian beef and lamb cuts is 2.35 mm and 3.73 mm, respectively, with most cuts completely trimmed of fat (1). In a US study, average selvedge fat on retail beef and lamb cuts was 3.8 mm (1991) (2). Nutrition surveys also suggest that around 80% of Australians eat their meat trimmed of fat (3). In addition high red meat eaters have been shown to consume almost 40% more vegetables than non red meat eaters and 60% more than light red meat eaters (4).

The majority of Australian beef and lamb are grass-fed. Due to the extensive practice of grain-feeding (mainly corn), US beef cuts are more marbled and contain almost twice as much fat and saturated fatty acids as comparable Australian beef cuts (5). Grass-fed Australian beef contains significantly more long chain omega 3 polyunsaturated fatty acids (LCPUFA) than grain-fed US beef. Lean Australian beef rump contains approximately 100 mg of LC n-3 PUFA which is comparable to that of many species of white fish in Australia (6). Furthermore, the intramuscular fat of Australian grass-fed steers has higher levels of conjugated linoleic (7) acid and lower levels of trans-fatty acids than grain fed steers (8).

Lean red meat is more widely available in Australia than in the US. The data presented suggests that studies investigating the association between red meat consumption and cancer in US populations may not be relevant to Australians due to these differences in the red meat supply.

References

1. Cobiac L, Droulez V, Leppard P, Lewis J. Use of external fat width to describe beef and lamb cuts in food composition tables. Journal of Food Composition and Analysis 2002; in press.
2. Savell JW, Harris JJ, Cross HR, Hale DS, Beasley LC. National Beef Market Basket Survey. J Anim Sci. 1991; 69: 2883–2893.
3. Coles-Rutishauser I, Penm R. Monitoring food habits and food security: Australia 1995–1996. Canberra; Australian Institute of Health & Welfare: 1996.
4. Baghurst K, Record S, Leppard P. Red meat consumption in Australia: intakes, nutrient contribution and changes over time. Austr J Nutr Diet 2000; 57 (4 Suppl): S3–S36.
5. www.nal.usda.gov/fnic/foodcomp
6. Sinclair AJ, Oon S, Lim C, Li D, Mann NJ. The omega-3 fatty acid content of canned, smoked and fresh Australian fish. Aust J Nutr Dietet 1998; 55: 3116–120.
7. French P, Stanton C, Lawless F, O'Riordan EG, Monahan FJ, Caffrey PJ, Moloney AP. Fatty acid composition including CLA of intramuscular fat from steers offered grazed grass, grass silage or concentrate based diets. J Anim Sci 2000; 78: 2849–55.
8. Sinclair AJ, Mann NJ, Yep Y. The omega-3 content of lean red meat, MLA Project CHMN.016 Final Report, 2002.

Effects of fish oil (MaxEPA) supplementation on fatty acid profile and platelet activating factor generation in human buccal mucosal cells

M Garg, A Bencke, RJ Blake

Discipline of Nutrition & Dietetics, Faculty of Health, University of Newcastle, NSW, 2308

Platelet activating factor (PAF) has been implicated as a contributing factor in a number of conditions including heart disease, thrombosis, acute inflammation, asthma and systematic anaphylaxis, immune disorders and gastrointestinal ulceration [1]. Diets enriched with marine n-3 fatty acids have been shown to reduce the production of PAF by human monocyte [2]. The present studies were carried out to determine phospholipid fatty acid composition and PAF generation alterations in human buccal mucosal cells following supplementation with fish oil (MaxEPA). Buccal mucosal cells may prove to be a viable tissue for measurement of fatty acid and PAF changes following dietary fat manipulations as collection of these cells is a relatively simple, non invasive and easy method to perform compared to invasive, painful blood collection by conventional venepuncture techniques.

Volunteers (7 males and 7 females) between the age of 18 and 65 were recruited from the University of Newcastle population by advertisement, excluding those on significant weight reduction diets, vitamin supplements, prescription drugs or smokers. Subjects were supplemented with 12 × 1 g MaxEPA capsules, providing collectively 3.6 g n-3 fatty acids per day for a period of four weeks. Buccal cells were collected by scraping with a wooden spatula and mouth rinsing with physiological saline solution at baseline, 1 week, and 4 weeks after MaxEPA supplementation. PAF production by mucosal cells was assessed by stimulation with a calcium ionophore (A23187) using radioimmunoassy. Fatty acid composition was determined by gas chromatography.

Marine n-3 fatty acids were incorporated into buccal mucosal cells, with a significant difference observed at 1 and 4 weeks for 20:5n-3 and at 4 weeks for 22:6n-3. This was accompanied by a decrease in 20:4n-6, with significance reached after 4 weeks. A general trend in PAF production was observed in both stimulated and unstimulated cells, but the difference did not reach a statistical significance ($p < 0.05$). We conclude that buccal mucosal cells are a viable tissue to determine fatty acid changes rapidly following dietary manipulation, and that A23187 may not be the most appropriate stimulant for PAF production in these cells.

	Baseline[1]	1 week[1]	4 weeks[1]
C20:4n-6 (%)	2.52 ± 0.20	2.53 ± 0.28	2.36 ± 0.15 [a]
C20:5n-3 (%)	0.04 ± 0.04	0.35 ± 0.11 [a]	1.32 ± 0.08 [a]
C22:6n-3 (%)	0.36 ± 0.21	0.31 ± 0.15	1.43 ± 0.09 [a]

[1]mean ± SEM. [a]$p = < 0.05$.

References

1. Braquet P, Touqui L, Slen TY, Vargaftig BB. Perspectives in platelet activating factor research. Pharmacol Rev 1987; 39: 97–145.
2. Sperling RI, Robin JL, Kylander LTH, Lewis RA, Austen KF. The effects of omega-3 polyunsaturated fatty acids on the generation of PAF-acether by human monocytes. J Immunol 1987; 139: 4186–4191.

Key words: omega-3 fatty acids, buccal mucosal cells, PAF

Fatty acid and sterol composition of frozen and freeze-dried New Zealand green lipped mussel (Perna canaliculus) from three sites in New Zealand

KJ Murphy, NJ Mann and AJ Sinclair

Department of Food Science, RMIT University, VIC, 3001

Omega-3 polyunsaturated fatty acids (n-3 PUFA), particularly docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) from fish oil have been widely investigated in terms of their beneficial effect on certain risk factors for cardiovascular disease and for reducing the symptoms of inflammatory diseases in humans. Lipids from the New Zealand Green Lipped Mussel (NZGLM) have been reported to possess anti-inflammatory activity *in vitro* and *in vivo* (1–3). The anti-inflammatory activity has been reported to reside in either the free fatty acid (FFA) fraction with fatty acids containing four, five and six double bonds, or sterols, or a polysaccharide fraction (3–4). In view of previously reported anti-inflammatory bioactivity of the NZGLM, the overall lipid profile and fatty acid and sterol composition of the NZGLM from various sites in New Zealand were investigated using thin layer chromatography (TLC) and gas liquid chromatography (GLC). Samples were either frozen (F) or freeze-dried (FD) soon after collection. It was also thought prior to the study, there may be differences in the dietary sources of phytoplankton between the sites, responsible for the bioactivity, however data collected in New Zealand reported no difference in the type of phytoplankton, but a difference in the quantity. There were no major significant differences in the major components of the lipid, fatty acid and sterol composition between FD or frozen samples, nor were there any significant differences in the major composition between sites. The only major difference was between total lipid composition of the freeze-dried and frozen samples due to the removal of water during freeze-drying.

Total lipid content on a dry weight basis in FD samples was significantly higher than frozen samples ($P < 0.05$) and there was no significant site variation. Triglyceride lipid fraction appeared to be the most prominent in the frozen and FD samples. The FFA band was the next most prominent band and was visually more prominent in the frozen samples. Sterol esters were detected in higher amounts in the frozen samples compared with the FD samples.

Polyunsaturated fatty acids were the main group of fatty acids in both frozen and FD samples (45–46%), most of which were n-3 PUFA (39–41%). Saturated fatty acids accounted for approximately one quarter of total fatty acids, with little variation between FD and frozen samples. The major fatty acids of the NZGLM were DHA (19% in both FD and frozen samples), EPA (15% in both FD and frozen samples) and palmitic acid in the FD sample (15%). Cholesterol was the most prominent sterol (31% of total sterols). This study is unique as it compares the lipid composition of the NZGLM from three sites in New Zealand with the additional effect of processing. This study showed that there were no major significant differences in lipid, sterol and fatty acid composition between the FD and frozen samples of the NZGLM for three sites in New Zealand. Food chain studies and further research is warranted to investigate the presence and role of major and minor lipid components of the NZGLM.

References

1. Gibson RG, Gibson SL, Conway V, Chappell D. *Perna canaliculus* in the treatment of arthritis. The Practitioner 1980; 224: 955–960.
2. Whitehouse MW, Macrides TA, Kalafatis N, Betts WH, Haynes DR, Broadbent J. Anti-inflammatory activity of a lipid fraction (Lyprinol) from the N.Z. green lipped-mussel. Inflammopharmacology 1997; 5: 237–246.
3. Emelyanov A, Fedoseev G, Krasnoschekova O, Abulimity A, Trendeleva T, Barnes PJ. Treatment of asthma with lipid extract of New Zealand green-lipped mussel: a randomized clinical trial. In press. J Respir. 2002.
4. Miller TE, Dodd J, Ormrod DJ, Geddes R. Anti-inflammatory activity of glycogen extracted from *Perna canaliculus* (NZ green-lipped mussel). Agents Actions 1993; 38: C139–42.

Keywords: NZGLM, n-3 PUFA, triglyceride

Comparison of n-3 polyunsaturated fatty acid and total lipid content of spawning and nonspawning Australian blacklip abalone

XQ Su[1], KN Antonas[1], D Li[2]

[1]*School of Life Sciences and Technology, Victoria University, Melbourne, 3000*
[2]*Department of Food Science, Hangzhou University of Commerce, Hangzhou, China, 310035*

Seafood has been reported to have health benefits due to the high concentration of n-3 polyunsaturated fatty acids (PUFA) in the forms of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) (1). Previous studies showed that there was a variation of n-3 PUFA concentration and total lipid content between spawning and nonspawning fish and shellfish (2,3). However, no data are available on the variation of fatty acid concentration between spawning and nonspawning abalone in Australia.

This study investigated the fatty acid and total lipid contents in spawning and nonspawning blacklip abalone (*Haliotis rubra*), collected from Port Phillip Bay, Victoria, Australia. The total lipid was extracted with methanol-chloroform containing butylated hydroxytoluene. The fatty acid methyl esters were prepared by standard methods, and fatty acids were separated by capillary gas liquid chromatography. The results are given below.

	Spawning (n = 8)	Nonspawning (n = 8)
20:5n-3(mg/100g)	29 ± 5	26 ± 5
22:5n-3 (mg/100g)	51 ± 4	46 ± 6
22:6n-3 (mg/100g)	5 ± 1	5 ± 1
Total n-3 PUFA (mg/100g)	90 ± 9	83 ± 10
Total n-6 PUFA (mg/100g)	78 ± 7	84 ± 14
SFA (mg/100g)	176 ± 34	203 ± 23
MUFA (mg/100g)	114 ± 14	108 ± 7
Total lipid (g/100g)	2.2 ± 0.2	2.5 ± 0.9*

Values are means ± SD. SFA = saturated fatty acids, MUFA = monounsaturated fatty acids. *P<0.05.

There were no significant differences in the concentration of total n-3 PUFA, 20:5n-3, 22:5n-3 and 22:6n-3 between the spawning and nonspawning samples. The concentration of total n-6 PUFA and MUFA also did not show a significant variation. It is likely that spawning has a tendency to decrease the concentration of SFA although the variation was not significant (P = 0.06). Total lipid content was significantly reduced in spawning samples compared with that in nonspawning samples ($P < 0.05$), which may indicate that more energy was consumed during spawning period.

References

1. Simopoulos AP. Human requirement for n-3 polyunsaturated fatty acids. Poult Sci 2000; 79: 961–970.
2. Styrishave B, Andersen O. Seasonal variations in hepatopancreas fatty acid profiles of two colour forms of shore crabs, *Carcinus maenas*. Marine Biology 2000; 137: 415–422.
3. Tocher DR, Sargent JR. Analyses of lipids and fatty acids in ripe roes of some northwest European marine fish. Lipids 1984; 19: 492–499.

The research was funded by Victoria University. All samples were provided by Ocean Wave Seafoods, Victoria.
Key words: n-3 PUFA, spawning/nonspawning abalone

Implementation of the DASH (Dietary Approaches to Stop Hypertension) intervention program in an Australian community setting

C Margerison, CA Nowson

School of Health Sciences, Deakin University, Burwood, VIC, 3125

The DASH (Dietary Approaches to Stop Hypertension) study (1) was a large dietary intervention study conducted in the US, which provided all food to participants for 8 weeks and effectively lowered blood pressure. The feasibility and effectiveness of this dietary modification in free-living individuals is not known.

The differences in nutrient content between the published DASH diet (US food database) (2) and the DASH diet using an Australian food database were assessed and an appropriate diet for Australians (OZDASH) was devised. The composition of the DASH diet for an energy intake of 8,820 kJ was analysed using the Australian food database.

The nutrient content of the DASH diet calculated using the Australian food database was lower in iron, potassium, energy, % energy from carbohydrate and magnesium, and was higher in vitamin C and calcium (see table). The DASH diet specifies 7 serves of grains, 5 of fruit, 4 of vegetables, 2.7 of dairy products (low fat), 2.5 of fats/oils, 0.5 of red meat, 0.6 of poultry, 0.5 of fish, and 0.6 of legumes/nuts per day.

Our newly developed OZDASH diet specifies 7 serves of grains, 4 of fruit, 5 of vegetables, 3 of low fat dairy and 2 of fats/oils per day as well as 1 serve of legumes, 3.5 of nuts/seeds, 3 of red meat, 3 of white meat and 3 of fish per week. The OZDASH diet is slightly lower in vitamin C and iron, and marginally higher in calcium and phosphorous than the DASH (US food data) (see Table). The iron content of the US diet appears to be higher than Australia for the same intake of foods, probably due to extensive iron fortification in the US.

	DASH diet (US food database)	DASH diet (Aus food database)	OZDASH diet (Aus food database)
E (kJ)	8820	8011	8563
% E from CHO	58	47	52
% E from fat	27	32	27
Fe (mg)	20.2	11.4	15.8
K (mg)	4589	3938	4409
Mg (mg)	465	388	470
Vit C (mg)	266	336	236
Ca (mg)	1220	1319	1332
P (mg)	1481	1752	1945

Nutrient intakes differ between countries, and are dependent on the food supply. The OZDASH diet was matched to the DASH on the basis on fat sources and % energy from fat, whilst maintaining high intakes of fruits, vegetables and dairy products in quantities acceptable to the general population. This has resulted in some nutrient differences between the DASH and the OZDASH diets. The OZDASH diet is currently being tested in a group of free-living individuals (hypertensives and normotensives).

References

1. Appel LJ *et al.* A clinical trial of the effects of dietary patterns on blood pressure. DASH Collaborative Research Group. N Engl J Med 1997; 336: 1117–24.
2. Karanja NM *et al.* Descriptive characteristics of the dietary patterns used in the Dietary Approaches to Stop Hypertension Trial. J Am Diet Assoc 1999; 99: S19–27.

Key words: DASH, blood pressure, diet

Taste preferences and blood pressure response to stress: a pilot study

SJ Torres, CA Nowson

School of Health Sciences, Faculty of Health and Behavioural Sciences, Deakin University, VIC, 3125

The physiological response to stress involves stimulation of the hypothalamic-pituitary-adrenal axis and the sympathetic nervous system, together with increases in blood pressure and heart rate. Increases in appetite for sweet foods and higher salt intakes have been reported in laboratory animals subjected to stress (1) (2). This pilot study was undertaken to examine blood pressure response and taste preferences for salt and sucrose solutions in human volunteers subjected to a standardised arithmetic stress test.

Twenty subjects were recruited from staff and students at Deakin University. Height and weight were measured and then subjects were asked to rest in the sitting position for five minutes. An ambulatory blood pressure monitor (TM-2421, A&D, Japan) automatically measured blood pressure every two minutes. Baseline blood pressure was assessed over 13 minutes following the rest period. Subjects then underwent a standardised arithmetic stress test for six minutes, which consisted of serial subtractions of seven starting from the number 9000, with subjects aiming to complete a set number of subtractions per minute. Twelve minutes after the conclusion of the stress test, the post stress (PS) period, subjects were asked to indicate their preference for both tomato juice with a range of salt concentrations (0, 0.1, 0.2, 0.3 and 0.5%) and water with a range of sucrose concentrations (4.5, 6.0, 7.5, 9.0 and 11.0%). Solutions were presented in random order. Within one week of completing the stress test, the subjects repeated the taste test at home when they were non-stressed (NS).

Seventeen subjects, 14 females and three males, completed the stress test, and the taste test in the PS and NS states. Subjects had a mean age of 39(12) (SD) years and a mean BMI of 24(3) kg/m^2. Systolic and diastolic blood pressures, and pulse rate were all significantly greater at the end of the stress test when compared to baseline (see table). Systolic and diastolic blood pressures were still elevated ten minutes after the stress test when compared to baseline levels. The mean concentration preferred by subjects for the salt solution PS was 0.22 ± 0.03 (SEM) % and 0.19 ± 0.04% when they were NS, and tended to be lower for the sucrose solution PS when compared to the NS state, 6.03 ± 0.52% and 7.24 ± 0.69% respectively, (P = 0.056).

There was no difference in the mean concentrations preferred by subjects for both the salt and sucrose solutions in the PS and NS states.

	Baseline period (13 minutes)[1]	Stress test (6 minutes)[1]	Post stress test period (10 minutes)[1]
Systolic (mmHg)	117 ± 2	131 ± 4*	122 ± 2*
Diastolic (mmHg)	74 ± 2	82 ± 3*	77 ± 2*
Pulse (bpm)	67 ± 2	78 ± 3*	66 ± 2

[1]mean ± SEM; *P<0.05, compared to baseline.

References

1. Silvera P, Xavier M, Souza F, Manoli L, Rosat R, Ferreira M, *et al.* Interaction between repeated restraint stress and concomitant midazolam administration on sweet food ingestion in rats. Braz J Med Biol Res 2000; 33: 1343–50.
2. Bourjeili N, Turner M, Stinner J, Ely D. Sympathetic nervous system influences salt appetite in four strains of rats. Physiol Behav 1995; 58: 437–43.

Key words: taste preference, stress, blood pressure

Alcoholic beverages lower acute glucose and insulin responses in healthy subjects

J Brand-Miller, V Liu, SHA Holt, S Liu

Human Nutrition Unit, University of Sydney, NSW, 2006
Brigham and Women's Hospital, Harvard Medical School, Boston MA, 02115, USA

Moderate alcohol intake has been related to lower risk of type 2 diabetes and coronary heart disease in epidemiological studies. In intervention studies, consumption of one or two alcoholic drinks has been associated with improvements in insulin sensitivity. Little is known about the direct effect of alcoholic beverages on acute glucose and insulin responses, either when consumed alone or with a mixed meal.

We studied the impact of three types of drinks (beer, white wine and gin) with and without a carbohydrate-containing meal on responses over two or three hours in 10 (5 M, 5 F) young healthy volunteers. Blood was sampled by capillary fingerprick at 15 min intervals during the first hour and at 30 min intervals during the second and third hour. A standard breakfast was given 2 h before the test meals.

In the first study, the three types of drinks or white bread were fed alone as 1000 kJ (240 Calorie) portions. Taking the area under the glucose curve (AUC) after bread as 100, the AUC after beer was 58 ± 1, after white wine 7 ± 3 and after gin 10 ± 5 (all differences $P < 0.01$). Both glucose and insulin responses declined gradually over the second hour and insulin levels were 15–30 pM less at 2 h than at baseline.

In the second study, the same beverages or water were fed together with a 2000 kJ (480 Calorie) portion of white bread. Taking the glucose AUC to bread and water meal as 100, the beer and bread was 84 ± 11, wine and bread 63 ± 6 and gin and bread 80 ± 11 (Figure). Only the white wine produced significantly ($P < 0.01$) lower glucose responses than bread and water and the effect was more pronounced in males than in females. There was a trend toward lower insulin responses in the first hour but the differences were not statistically significant at the $P < 0.05$ level.

Figure. Relative glucose responses to 2000 kJ portion of white bread with and without 1000 kJ serving of alcoholic beverage. Only white wine was significantly different to bread and water ($P < 0.01$).

We conclude that alcoholic beverages, particularly wine, help to lower glucose and insulin responses after a carbohydrate-containing meal. Since postprandial hyperglycemia has been linked to higher risk of cardiovascular and total mortality (1), one of the mechanisms by which alcohol might exert its protective effect is via reductions in the glycemic response to high carbohydrate meals.

Reference

1. de VF, Dekker J, Ruhe H, *et al.* Hyperglycaemia is associated with all-cause and cardiovascular mortality in the Hoorn population: the Hoorn Study. Diabetologia 1999; 42: 926–31.

The glycaemic and insulin index values of a range of Australian honeys

SHA Holt[1], V de Jong[1], JC Brand Miller[1], J Arcot[2]

[1]*Human Nutrition Unit, Dept of Biochemistry GO8, University of Sydney, NSW, 2006*
[2]*Dept of Food Science and Technology, University of NSW, NSW, 2052*

Honey is often considered to be a uniform foodstuff consisting, on average, of 17% water, 82% carbohydrate and 0.5% proteins, amino acids, vitamins and minerals. However, the colour, flavour, and sugar profile of honeys vary depending on the floral nectar source visited by the bees. Australian honey has a greater range of flavours and colours than honeys from other countries and can vary markedly in the amounts of fructose and glucose they contain. Consequently, it is likely that different types of Australian honeys will produce different blood glucose and insulin responses. The aim of this study was to compare the effects of equal-carbohydrate portions of eight different types of Australian honey on postprandial blood glucose and insulin responses. A glycaemic index (GI) and insulin index (II) value was calculated for each honey, using glucose as the reference food (index value of glucose = 100). The sugar profiles and organic acid contents of the eight honeys were determined using standard HPLC methods and the osmolality of each honey was measured using an osmometer.

The study was conducted in two parts using two separate groups of 9–10 healthy, non-smoking, normal-weight subjects. The reference food and four types of honey were tested in each part. The study employed a repeated-measures design, such that in both parts of the study, each subject consumed each of the four test honeys on one occasion and the reference food on two occasions. Subjects fasted for = 10 h overnight and then reported to the test centre the next morning, where they first gave a fasting finger-prick blood sample and then consumed a fixed portion of the reference food or a honey containing 25 g of available carbohydrate. Additional finger-prick blood samples were obtained at 15, 30, 45, 60, 90 and 120 min after eating commenced. For each subject, a GI value was calculated for each honey by dividing the area under the 120-min plasma glucose response curve (AUC) for that food by the average AUC value for the reference food and then multiplying by 100. An II value for each food was calculated using the same formula with insulin instead of glucose AUC values. The mean ± SEM GI and II values for the eight honeys are shown in the table below (n = 10 except for the honeys tested in the second part of the study * where n = 9).

Honey variety	GI value (%)	II value (%)	Fructose (g in test portion)	Glucose (mg/test portion)	Osmolality	Total organic acid content
Yellow box	35 ± 4	40 ± 5	15.3	9.0	5676	0.50
Stringybark*	44 ± 4	47 ± 3	15.9	8.5	5678	0.56
Red Gum	46 ± 3	51 ± 3	11.7	11.2	4884	0.48
Iron bark	48 ± 3	42 ± 4	14.1	9.8	4624	0.72
Yapunyah*	52 ± 5	42 ± 4	15.5	8.8	4824	0.84
Commercial blend 2 (WA)*	62 ± 3	62 ± 4	13.6*	10.5	4551	0.47
Salvation Jane	64 ± 5	52 ± 3	12.9	11.2	4804	0.43
Commercial blend 1 (NSW)*	72 ± 6	67 ± 6	13.6	15.5	5708	0.57

The mean GI and II values of the honeys were significantly related ($r = 0.88$, $n = 8$, $P < 0.01$). The glucose content (grams per 25-g available carbohydrate test portion) of the honeys was significantly related to their mean GI ($r = 0.79$, $n = 8$, $P < 0.05$) and II ($r = 0.77$, $P < 0.05$) values. The honeys' fructose contents were negatively but not significantly related to the mean GI and II values ($r = -0.41$, $n = 8$, NS). Both the sucrose and maltose contents were positively but not significantly related to the honeys' mean GI and II values. Similarly, the honeys' osmolality and organic acid contents were negatively but not significantly related to their mean GI or II values. Honey is a complex substance containing as many as 180 different compounds, some of which, such as flavonoids and phenolic acids, may reduce glycaemia. The results of this study show that all types of honey should not be classified as one foodstuff, particularly for people with diabetes.

Supported by: RIRDC grant
Key words: glycaemic index, insulin, honey

The effects of chocolate-containing foods on postprandial blood glucose and insulin

SHA Holt, V de Jong, S Loyer, J Kennedy, JC Brand Miller

Human Nutrition Unit, Dept of Biochemistry GO8, University of Sydney, NSW, 2006

Claims that chocolate elicits high postprandial insulin responses, resulting in rebound hypoglycaemia and hunger have little scientific justification. While some chocolate products have been shown to generate greater insulin responses relative to their glycaemic responses, this may not be due to the chocolate content *per se* (1). The aim of this study was to compare the effects of equal-carbohydrate portions of common chocolate-containing products vs their non-chocolate counterparts on postprandial blood glucose and insulin levels.

Six pairs of foods were tested. Within each pair, one food contained chocolate and the other food was a non-chocolate version of the same food with a similar macronutrient content. The study was conducted in two parts, and three pairs of food and the reference food were tested in each part. A separate group of 10 healthy, non-smoking, normal-weight subjects was recruited to participate in each part of the study. The study employed a repeated-measures design, such that in both parts of the study, each subject consumed the six test foods on one occasion and the reference food on two occasions. Subjects fasted for = 10 h overnight and then reported to the test centre the next morning, where they first gave a fasting finger-prick blood sample and then consumed a fixed portion of the reference food or a test food containing 50 g of available carbohydrate. Additional finger-prick blood samples were obtained at 15, 30, 45, 60, 90 and 120 min after eating commenced. For each subject, a GI value was calculated for each food by dividing the area under the 120-min plasma glucose response curve (AUC) for that food by the average AUC value for the reference food and then multiplying by 100. An II value for each food was calculated using the same formula with insulin instead of glucose AUC values. The mean ± SEM GI and II values for the six pairs of foods are shown below (n = 10 except for foods labelled with * where n = 9).

Chocolate test food	GI value (%)	II value (%)	Non-chocolate test food	GI value (%)	II value (%)
Chocolate premium ice cream	37 ± 3	71 ± 3	Vanilla premium ice cream	38 ± 3	54 ± 4
Chocolate cake with icing	41 ± 4	88 ± 14	Vanilla cake with icing	41 ± 4	67 ± 12
Chocolate reduced-fat milk	41 ± 4	86 ± 11	Strawberry reduced-fat milk	35 ± 3	59 ± 5
Plain milk chocolate	42 ± 7	71 ± 13	Plain white chocolate	43 ± 6	63 ± 13
Chocolate instant pudding	47 ± 4	80 ± 5	Vanilla instant pudding	40 ± 4*	62 ± 5
Chocolate puffed rice cereal	76 ± 3*	79 ± 10	Plain puffed rice cereal	84 ± 4	64 ± 6
Food group mean value	47 ± 3	80 ± 4	Food group mean value	47 ± 3	62 ± 3

The mean GI values of the two food groups were not significantly different, but the mean II value for the chocolate food group was significantly greater than the mean II value of the non-chocolate food group (paired two-tailed t-test; P = 0.004). The individual subjects' glucose AUC values were significantly associated with their corresponding insulin values for the non-chocolate products (r = 0.37, n = 60, P < 0.01), but not for the non-chocolate products (r = 0.23, n = 60, NS). The disproportionately higher II values for the chocolate products could be due to higher cephalic phase insulin secretion, due to the greater sensory enjoyment of these foods, or specific insulinogenic compounds in chocolate. The physiological significance of the greater insulin secretion, which has also been observed for some other dairy products, remains to be determined.

Reference

1. Brand-Miller JC, Pang E, Broomhead L. The glycaemic index of foods containing sugars: comparison of foods with naturally-occuring v. added sugars. Br J Nutr 1995; 73: 613–23.

Key words: glycaemic index, insulin, chocolate

Do definitions of anaemia based on haemoglobin or haematocrit yield the same prevalence of anaemia in peri-pubertal children?

D Mackerras, G Singh

Menzies School of Health Research, Darwin, NT, Australia

Anaemia can be defined using a cutoff level for haemoglobin (Hb) or a cutoff level for haematocrit (Hct). The World Health Organisation has defined cutoffs for both parameters, and slightly different cutoffs of both parameters are used to define anaemia in the United States. In view of the international focus on micronutrient deficiency at present, we compared the prevalence of anaemia obtained when the WHO and US cutoffs are used for each of the two parameters.
Data were used from the Aboriginal Birth Cohort study. Children were seen when aged 8–14 years in 1999–2001. At this time, growth and markers of chronic disease were assessed and venous blood was collected for estimation of biochemical and haematological indices. No biochemical tests of iron deficiency were done. The blood films were also examined but no child had macrocytosis.

Children were defined as being anaemic using the WHO criteria if their Hb was below 11.5 g/dL for those aged 11 years or less or if Hb was below 12.0 g/dL for those aged 12 years and older. They were defined as being anaemic if their Hct was below 34% or 36% respectively (1). Using the US criteria, children aged 11 years or less were classed as anaemic if Hb was below 12.0 g/dL, boys aged 12 and older if Hb was below 12.6 g/dL and girls aged 12 years and over, if Hb was below 11.9 g/dL. Using Hct as the basis of the definition, children were classed as anaemic if their values were below 35.5%, 37.4% and 35.8% respectively. Six children aged eight or 14 years were excluded leaving 518 in the analysis.

The table shows the prevalence of anaemia using the different definitions. As haemoglobin and haematocrit measure slightly different things, it is not surprising that there are some differences in prevalence. The overall difference between the WHO and US criteria can be explained by the fact that the US cutoffs were set at the 5th centile of their population distribution (2) and the WHO criteria appear to be based on a lower level for this age group, although this is not true of all age groups (1,2). However the more than two-fold discrepancy in prevalence using the two WHO criteria may to be due to the use of a conventional conversion factor to derive the Hct cutoff from the Hb value (1) rather than deriving the Hct cutoff from the population distribution of Hct values.

These results suggest that the prevalence of anaemia cannot be compared between locations that have used different WHO indices, even though these are presented as being interchangeable (1).

	Prevalence of anaemia (%)	95% CI
WHO cutoffs		
Haemoglobin	13.1	10.2–16.0
Haematocrit	6.0	3.9–8.0
US cutoffs		
Haemoglobin	24.1	20.4–27.8
Haematocrit	19.5	16.7–22.9

References

1. World Health Organisation. Iron deficiency anaemia. Assessment, prevention and control. A guide for programme managers. WHO, 2001. (http://www.who.int/nut/documents/ida_assessment_prevention_control.pdf)
2. Recommendations to prevent and control iron deficiency in the United States. Morbidity Mortality Weekly Reports, 1998 Vol 47 April 3rd. (http://www.cdc.gov/mmwr/PDF/rr/rr4703.pdf)

Supported by: Colonial Foundation, Channel 7 Children's Research Foundation of South Australia and the NHMRC
Key words: anaemia, surveillance

Iron intake and iron status of rural Malaysian adolescents

LH Foo[1], GL Khor[1], ES Tee[2], D Prabakaran[3]

[1]*Department of Nutrition and Health Sciences, Faculty of Medicine and Health Sciences, Universiti Putra Malaysia, Serdang, Selangor, Malaysia*
[2]*Human Nutrition Division, Institute For Medical Research, Ministry of Health, Malaysia*
[3]*Sabah State Health Department, Ministry of Health, Kota Kinabalu, Sabah, Malaysia*

The iron content of the diet is especially important in the adolescent period because of the growth spurt and to replace losses. A convenience sample of 199 apparently healthy adolescents consisting of 94 males and 105 females aged 12 to 19 years from six fishing villages of Tuaran, Sabah was studied in order to determine iron intakes and dietary iron sources in relation to iron status. A validated food frequency questionnaire (FFQ) containing 62 usual food items of these fishing villages was used to quantify food and nutrients intake. The mean age of subjects was 15.2 years. Half of the subjects lived in medium size households and almost three-quarters of the households were below poverty line (<RM122.65/month). The mean dietary iron intakes of males and females were similar (10.7 ± 2.6 mg/d and 10.0 ± 2.9 mg/d, respectively). A high proportion of the females (90%) had dietary iron intake below the 2/3 Malaysia RDA level compared with their male counterparts (63%). The majority of the subjects derived most dietary iron from foods of plant origin (78%), whereas only 22% was contributed by animal-origin products. Males had a significantly higher intake of dietary iron from rice and rice products ($P < 0.05$), local snack foods ($P < 0.05$), cereals and tubers ($P < 0.001$), fruits ($P < 0.001$), fish and seafood ($P < 0.05$) and chicken and meat ($P < 0.01$). In contrast, female adolescents had a significantly higher iron intakes derived from nutrient fortified milk beverages ($P < 0.05$). Multiple iron status indicators indicated that 18.9% of males and 26.4% of females were iron deficient, while 5.4% of males and 26.4% of females had iron deficiency anemia. The correlation analysis showed that total dietary iron intake was significantly correlated with serum ferritin ($r = 0.457$, $P < 0.001$), serum iron ($r = 0.309$, $P < 0.001$), transferrin saturation ($r = 0.339$, $P < 0.001$), mean corpuscular volume ($r = 0.477$, $P < 0.001$) and hemoglobin ($r = 0.431$, $P < 0.001$), and there was a significant negative correlation with total iron binding capacity ($r = -0.284$, $P < 0.001$). Further, dietary iron intake derived from plant-origin was also found to be significantly correlated with the iron status indicators. The significant correlation between the intake of non-heme iron and iron stores indicates the importance of both haem and non-haem iron in the diet. The low amount of bioavailable heme iron places Malaysian adolescents at risk of iron deficiency. The identification of sources of dietary iron in these adolescents hopefully can serve to formulate food-based dietary guidelines tailored to the specific needs of adolescent population groups.

Key words: iron status, dietary iron intake, adolescents

Retinyl acetate stimulates ferritin synthesis in Caco-2 cells in vitro

IR Record, JK McInerney, PI Leppard

CSIRO Health Sciences & Nutrition, Adelaide, SA, 5000

Differentiated cultures of Caco-2 human colon cancer cells provide a widely used model system to study the accumulation and transport of nutrients. This cell line is responsive to iron in that ferritin synthesis occurs as a result of exposure to the metal (1). In this study we have examined some of the interactions between iron and retinyl acetate in the synthesis of ferritin. Concomitant addition of retinyl acetate and iron for 24h showed no interaction, however incubation of the cells for 72h in the presence of retinyl acetate prior to the addition of iron resulted in a significant increase in ferritin synthesis. In the absence of added iron, retinyl acetate increased ferritin synthesis slightly but significantly.

Retinyl acetate added (μmol/L)	Exposure time prior to addition of iron (h)	Cell Ferritin ng/mg protein No added iron	Cell Ferritin ng/mg protein Iron (20 μmol/L)
0	0	8.9	894
0	72	10.1	787
6.25	0	11.3	971
6.25	72	12.6	1070
12.5	0	11.2	983
12.5	72	15.4	1160
25	0	10.7	990
25	72	22.8	1510
Pooled SE		1.3	49

When β-carotene was added to the cultures there was a small increase in the amount of ferritin produced, although duration of exposure appeared to have no effect.

Addition of either phytic acid or black tea infusion reduced ferritin synthesis in a dose-dependent manner. Addition of vitamin A partially overcame the effect of phytic acid, but not tea, whereas β-carotene had no effect. This study shows that the model system can be useful in unravelling some of the more complex interactions between nutrients and antinutrients in foods.

Reference

1. Glahn RP, Lee OA, Yeung A, Goldman MI, Miller DD. Caco-2 cell ferritin formation predicts nonradiolabelled food iron availability in an in vitro digestion/Caco-2 cell culture model. J Nutr 1998; 128: 1555–1561.

Key words: Iron, vitamin A, ferritin

Fat deposition pattern in pork primal cuts from finisher gilts

DN D'Souza[1], FR Dunshea[2], D Suster[2], DW Pethick[3], JR Pluske[3], BP Mullan[1]

[1]*Department of Agriculture, South Perth, WA, 6151*
[2]*Victorian Institute of Animal Science, DNRE, Victoria, 3030*
[3]*Division of Veterinary and Biomedical Sciences, Murdoch University, Murdoch, WA, 6150*

Pork bellies are a highly-valued cut in Singapore but consumers will not pay premium prices for pork belly cuts that have high intermuscular and subcutaneous fat depots. The issue of excess belly fat has been the most evident and widespread problem since Australia started exporting chilled pork carcasses to Singapore. Carcasses for export to Singapore are selected based on having a backfat thickness at the P2 site (6.5 cm from the midline over the last rib) less than 12 mm. Anecdotal feedback from Singapore indicates that excess belly fat remains a major problem even in the 'lean' carcasses. However, before any strategies to decrease belly fat are undertaken, it is essential to gain a better understanding of fat deposition patterns in the different pork cuts in relation to total body fat. The aim of this study was to determine the deposition of fat in the different primal cuts relative to the total fat in the carcass of female finisher pigs.

A total of 80 Large White x Landrace x Duroc crossbred gilts of similar age were used in this experiment. The pigs were stratified on a weight basis and randomly allocated to one of 10 pens (8 pigs/pen). The pigs in each pen were allocated a slaughter age over a 10-week period starting from 16 to 25 weeks of age. At their pre-designated slaughter age all pigs within the pen were slaughtered at a commercial abattoir. Twenty-four hours post-slaughter the right side of each carcass was divided into primal cuts (shoulder, loin, belly and ham), and weighed. The subcutaneous and intermuscular fat content for the shoulder, loin, belly and ham primal cuts was determined by dissection (1). The ratio of % fat in each primal cut to the % fat of the half carcass was then determined.

	Age (weeks)											
	16	17	18	19	20	21	22	23	24	25	l.s.d.	P-value
Slaughter weight (kg)	68.7	72.3	72.5	79.1	87.7	94.9	104.6	107.2	112.0	119.7	9.60	<0.001
Carcass weight (kg)	44.4	44.0	45.4	50.4	55.0	60.2	67.2	69.4	72.8	77.2	6.42	<0.001
% Carcass Fat	15.5	14.1	15.3	14.9	17.6	17.2	18.0	17.4	21.3	19.2	3.76	0.010
% Primal fat / % carcass fat												
Shoulder/Carcass	0.88	0.84	0.91	0.84	0.89	0.91	0.93	0.86	0.80	0.77	0.127	0.267
Loin/Carcass	1.3	1.4	1.3	1.6	1.2	1.3	1.4	1.2	1.2	1.3	0.258	0.104
Belly/Carcass	1.2	1.2	1.3	1.4	1.8	1.7	1.8	2.5	2.9	2.9	0.478	<0.001
Ham/Carcass	0.84	0.86	0.81	0.82	0.80	0.83	0.83	0.81	0.76	0.80	0.109	0.895

The % carcass fat increased by 5–6% while the % fat in the shoulder, loin and ham primal cuts in relation to % carcass fat did not significantly change from 16 to 25 weeks of age. However, the proportion of belly:carcass fat significantly increased from 16 to 25 weeks of age. The increase in subcutaneous and intermuscular fat in the belly primal cut occurred from 20 weeks of age or a slaughter weight of 88 kg. This 'late' deposition of fat in the belly compared to the other primal cuts in finisher gilts is of major concern to the Australian pork industry as there is a trend in Australia to slaughter pigs at heavier weights. Increasing slaughter weights could further exacerbate the issue of excess belly fat in carcasses. These data also indicate that the late finisher phase from 20 weeks of age is the most appropriate period to initiate any management strategies to control belly fat deposition in gilts.

Reference

1. Suster D, Leury BJ, Hewitt R, Kerton DJ, Dunshea FR. Porcine somatotrophin (Reporcin) decreases carcass and belly fat in the finisher gilt. In: Cranwell PD, ed. Manipulating pig production VIII, Werribee, Aust Pig Sci Assoc 2001; 68.

Supported by: Australian Pork Limited, Canberra
Key words: Pork belly, fat, gilts

Distribution and developmental changes of transforming growth factor-β receptors in the gastrointestinal tract of pigs

J Mei[1], YQ Zhang[2], D King[1], RJ Xu[1]

[1]*Dept of Zoology, University of Hong Kong, Pokfulam Road, Hong Kong, China*
[2]*Dept of Histology and Embryology, Fourth Military Medical University, Xian, China*

Transforming growth factor-beta (TGF-β) has been detected in the milk of various species, including the pig (1). It may play a significant role in postnatal adaptation of the gut in the suckling neonate (2). This study investigated whether TGF-β receptors exist in the gut of newborn pigs, and whether there are any developmental changes in the receptor expression during the postnatal period.

Gastrointestinal tissue samples were collected from newborn unsuckled piglets and suckling piglets of different ages. The localization and quantification of TGF-β receptors along the gastrointestinal tract were performed by immunohistochemical and Western blot analyses.

In newborn unsuckled piglets, TGF-β receptors were widely distributed along the gastrointestinal tract (Figure). In the small intestine, TGF-β receptors I and II were predominantly localized on the apical membrane of the villus epithelium, while TGF-β receptor III was predominantly localized in the crypts. The mucus glands in the esophagus and the Brunner's glands in the duodenum were strongly stained with receptor III antibody. Scattered mucosal lymphocytes located in lamina propria and in Peyer's patches were also stained positively. In suckling piglets, there was a transient decline in the density of TGF-β receptors I and II in the intestinal mucosa. The positive staining of receptor I and II on the villus epithelium and the staining of receptor III in the esophageal and Brunner's glands were less intense in 1- and 3-day-old suckling piglets than in the newborns, while more intestinal mucosal lymphocytes were positively stained in the suckling piglets.

The present study established for the first time the existence of TGF-β receptors in the gastrointestinal tract of newborn pigs, and demonstrated spatial and expression level changes of receptors following the onset of sucking. The findings support the hypothesis of a regulatory role of milk-borne TGF-β in postnatal adaptation of the gastrointestinal tract in neonatal animals.

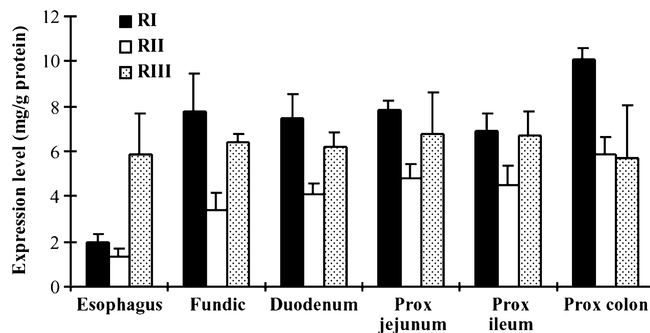


Figure. Expression levels of TGF-β receptors I (RI), II (RII) and III (RIII) in the gastrointestinal mucosa in newborn piglets determined by quantitative Western blot analysis (mean ± SEM).

References

1. Xu RJ, Doan QC, Regester GO. Detection and characterization of transforming growth factor-beta in porcine colostrum. Biol Neonate 1999; 75: 59–64.
2. Xu RJ, Wang F, Zhang SH. Postnatal adaptation of the gastrointestinal tract in neonatal pigs: a possible role of milk-borne growth factors. Livestock Prod Sci 2000; 66: 95–107.

Supported by: Reseearch Grants Council of Hong Kong (HKU 7234/98M)
Key words: TGF-β receptor, intestine, pig

Detection of betaglycan in porcine and human milk

HK Cheung, J Mei, RJ Xu

Department of Zoology, University of Hong Kong, Hong Kong

Betaglycan is a transmembrane proteoglycan with a high binding affinity to transforming growth factor beta (TGF-,). It may play a specific role in TGF-, signal transduction by presenting TGF-, to the receptors (1). Soluble form of betaglycan has been detected in various biological fluids, including cell culture medium, cellular matrix extract (2) and rat milk (3). It has been suggested that soluble betaglycan may act as a dual modulator regulating the binding of TGF-, to the surface receptors (4) and neutralizing excessive ligands (5). In the present study, betaglycan was detected in porcine and human milk by Western blot analysis.

Porcine milk samples were collected from four Large White sows at day one, day three, day seven and day fourteen of lactation. Human milk was collected from a lactating mother during the second month of lactation. Fat in the samples was removed by centrifugation and protein concentration was determined by Lowry's method. Characterization of betaglycan in porcine and human milk was performed by western blot analysis under non-reducing and reducing conditions. Polyclonal antibodies against betablycan (Santa Cruz Biotechnology) was used in the analysis.

Immunoreactive betaglycan was detected in all milk samples. In porcine milk the concentration of betaglycan was highest on the first day of lactation and the concentration declined gradually with the progress of lactation (Figure).

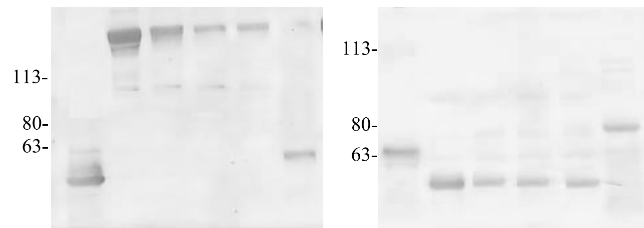


Figure 1. Western blot analysis under non-reducing condition. Lane 1, isolated human betaglycan. Lanes 2–4, porcine milk at day 1, 3, 7, 14 respectively. Lane 5, human milk.

Figure 2. Western blot analysis under reducing condition. Lane 1 isolated human betaglycan, Lanes 2–4, porcine milk at day 1, 3, 7, 14 respectively. Lane 5, human milk.

The estimated molecular size of immunoreactive betaglycan in porcine milk was about 180 kDa. Under reducing condition, the molecular size of immunoreactive betaglycan reduced to below 60 kDa, indicating the existence of disulphide bonds in the original compound. Incubation of porcine milk with deglycosylation enzymes, chondritinase and heparitinase reduced the molecular size from 180 kDa to about 110 kDa, confirming the existence of chondroitin sulphate and heparan sulphate glycosaminoglycan chains (3). In contrast, the immunoreactive betaglycan detected in human milk had a molecular size of around 75 kDa and it did not change under reducing condition. The difference between human and porcine milk in the molecular characteristics of immunoreactive betaglycan may be due to a species difference or to post-excretory degradation of the compound.

This study represents the first report of betaglycan in porcine and human milk. The physiological significances of milk-borne betaglycan remain to be elaborated.

References

1. Massagué J. TGF-â signal transduction. Annu Rev Biochem 1998; 67: 753–91.
2. Andres JL, Stanley K, Cheifetz S, Massagué J. Membrane-anchored and soluble forms of betaglycan, a polymorphic proteoglycan that binds transforming growth factor-â. J Cell Biol 1989; 109: 3137–45.
3. Zhang MF, Zola H, Read LC, Penttila IA. Identification of soluble transforming growth factor-â receptor III (sTâIII) in rat milk. Immunol Cell Biol 2001; 79: 291–97.
4. López-Casillas F, Payne HM, Andres JL, Massgué J. Betaglycan can act as a dual modulator of TGF-â access to signaling receptors: mapping of ligand binding and GAG attachment sites. J Cell Biol 1994; 124: 557–68.
5. Vilchis-Landeros MM, Montiel JL, Mendoza V, Mendoza-Hernández G, López-Casillas F. Recombinant soluble betaglycan is a potent and isoform-selective transforming growth factor-â neutralizing agent. Biochem J 2001; 355: 215–22.

Supported by: Research Grants Council of Hong Kong (HKU7234/98M)
Key words: Betaglycan, milk, TGF-β

Effect of an experimental oligosaccharide on bacterial populations in the large bowel of healthy piglets

JEAC Napo1i[1], B Wang[1], A Staples[1], A Hunter[1], JC Brand Miller[1], PL Conway[2]

[1]*School of Molecular and Microbial Biosciences, University of Sydney, NSW, 2006*
[2]*School of Biotechnology and Biomolecular Sciences, University of New South Wales, NSW, 2052*

The intestinal mucosa of pigs lacks the enzymes capable of cleaving a number of oligosaccharides. These oligosaccharides are readily fermented in the caecum and colon by the microflora and promote the growth of lactic acid bacteria such as *Bifidobacterium* and *Lactobacillus* species which inhibit colonisation by pathogens. Although there has been a growing interest in the beneficial effects of oligosaccharides there is limited information on the prebiotic properties of oligosaccharides in pigs. This study investigates the effect of an experimental oligosaccharide (OS), found in human milk, on bacterial populations in the large bowel of infant pigs.

Twelve three-day-old male piglets (1.4–2.4 kg) were randomly allocated to a standard diet (n = 6) of soy/whey/casein sow milk pig-replacer (55:9:36) or the standard diet supplemented with 215 mg/kg OS per day (n = 6) for 30 days. Animals were euthanased for collection of digesta from regions of the large bowel (caecum, asending colon, transverse colon, desending colon and sigmoid colon). Samples were homogenised and diluted 1:10 w/v in Wilkins-Chalgren (WC) anaerobe broth. Ten μL of serial dilutions (10^{-1} to 10^{-8}) were plated on WC anaerobe blood agar (total anaerobes) and supplemented WC anaerobe blood agar (*Bacteroides*), Reinforced Clostridial agar (*Clostridium*), Rogosa agar (*Lactobacillus*) and raffinose bifidobacteria agar (*Bifidobacterium*), Nutrient agar (total aerobes), and MacConkey agar (*Enterobacteriaceae*). Plates were incubated anaerobically for 72 h and aerobically for 24 h at 37°C.

	Oligosaccharide diet[1]		Standard diet[1]	
	Caecum	Colon	Caecum	Colon
Total aerobes	6.31[2]	6.32[2]	7.03	7.18
Enterobacteriaceae	6.44[2]	6.52	7.21	6.95
Total anaerobes	6.87	6.93	7.23	7.01
Bacteroides	5.83	6.93	7.15	6.43
Bifidobacteria	7.11	6.64[2]	6.35	6.31
Lactobacilli	6.70	6.83[2]	6.49	6.40
Clostridia	4.87	5.10	4.94	4.75

The bifidobacteria and lactobacilli populations were significantly higher in the colon of piglets fed the OS diet ($P < 0.05$). Total aerobes were lower in the caecum and colon of piglets fed OS compared with standard sow milk pig-replacer ($P < 0.05$). Oligosaccharide fed piglets also had lower counts of gram negative rods in the caecum ($P < 0.05$). This study showed that supplementation of diets with OS at this dose modifies bacterial populations in the large bowel of piglets, in particular increased numbers of bifidobacteria and lactobacilli.

Key words: oligosaccharides, bacterial populations, piglets

Western refined dietary pattern is associated with risk of Inflammatory Bowel Disease

AJ Bencke[1], DCK Roberts[2], RG Batey[3], K D'Este[4]

[1]*Nutrition & Dietetics, School of Health Sciences, University of Newcastle, NSW, 2308*
[2]*AFGC Canberra, formerly Nutrition & Dietetics, University of Newcastle, NSW, 2308*
[3]*Dept of Gastroenterology, John Hunter Hospital, New Lambton Heights NSW, 2305*
[4]*Centre for Epidemiology & Biostatistics, University of Newcastle, NSW, 2308*

Background: Nutrients and foods may have interactions through non-nutrient components that can not be examined by single nutrient or food group analyses. Dietary patterning may examine this aspect of diet and is particularly useful when there is no consistent dietary aetiology, as in Inflammatory Bowel Disease (IBD) and its two main diseases Crohn's disease (CD) and Ulcerative Colitis (UC). In an energy adjusted nutrient analysis which controlled for modifiers/confounders, saturated and total fat intake were the only significant predictors of IBD (1). These and a protective effect of starch intake were also significant in UC, whilst only Vitamin A was a significant predictor for CD.

Objective: To determine the pre-symptomatic dietary patterns which may be involved in the aetiology of IBD and in the CD and UC subgroups of this case control study.

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 six months of diagnosis from NSW and ACT by referral from gastroenterologists. Diet was assessed within two years of onset of symptoms by FFQ (including vitamin supplements). Food groups were formed and dietary patterns determined using Principal Components Analysis (PCA) and analysed by conditional logistic regression after controlling for confounders*.

Outcomes: Data from 107 case and 308 matched controls were used in the analyses. The 'western refined' dietary pattern in IBD correlated highly with cakes & biscuits, takeaways, sugar & confectionery, soft drinks & juices and margarines & oils. Two separate dietary patterns were associated with CD – 'teetotal, sugar & cakes' (correlated highly with sugar & confectionery; cakes & biscuits; soy products, bread & grains, and lack of alcohol) and 'fast foods' (Trend P value NS) which correlated highly with takeaways, soft drinks and lack of fruit. No significant dietary patterns were found for UC.

Risk of IBD – 107 cases matched to 308 controls, Odds ratio (OR) for quartiles of dietary score

Dietary Pattern	Lowest Quartile	2nd Quartile	3rd Quartile	Highest Quartile	95% CI lowest versus highest Quartile	Trend P value
Western refined diet*	1	1.13	1.81	3.55	1.72– 7.33	0.0002

*controlled for oral contraceptive use, breast fed less than 6 weeks & past smoking.

Risk of CD – 65 cases matched to 186 controls, Odds ratio (OR) for tertiles of dietary score

Dietary Pattern	Lowest Tertile	Middle Tertile	Highest Tertile	95% CI lowest versus highest Tertile	Trend P value
Fast foods*	1	1.29	2.51	1.06–5.94	0.10
Teetotal, sugar & cakes*	1	1.24	2.88	1.31–6.37	0.006

*controlled for oral contraceptive use, breast fed less than 6 weeks & past smoking.

Conclusions: A 'western refined' diet is related to the subsequent appearance of IBD and 'teetotal sugar and cakes' and 'fast foods' with the appearance of CD in genetically susceptible individuals. This data suggests that a diet avoiding these food patterns, such as a Mediterranean eating style may reduce the incidence of IBD in genetically susceptible people.

Reference

1. Bencke AJ, Roberts DCK, Batey R. 'Saturated fat linked to risk of inflammatory Bowel Disease' (Conference Abstract). APJCN 2001; 10: S87.

Prevalence of overweight in Hunter primary school children – a pilot study

M Burgoyne[1] and CE Collins[1]

[1]*Nutrition and Dietetics, School of Health Sciences, University of Newcastle, NSW, 2308*

Australian children and adolescents are getting fatter. When the 2000 International Obesity Task Force BMI for age cut points (2) were applied to two national data sets collected 10 years apart (1985–1995), the prevalence of overweight in Australian children aged 7–15 years had almost doubled, whilst the prevalence of obesity had tripled (1). This suggests that overweight and obesity levels are increasing in Australian children.

Students were recruited from four Hunter primary schools of varying socio-economic status (SES) (2-low SES, 1-high SES, 1-mod SES). Anthropometric data was obtained from children who had parental consent to participate in the survey. The study was approved by the Hunter Area Health Service, the University of Newcastle and the NSW Department of Education Ethics Committees. Children were categorised, using the BMI cut-points as either at a Healthy Weight (equivalent to an adult BMI $\leq$ 25), 'At Risk' of becoming overweight (equivalent to an adult BMI >25 to <30) or Overweight (equivalent to an adult BMI $\geq$ 30).

Of the 917 children who received an invitation to participate in the study, 290 children were weighed and measured. The average response rate across the schools was 31.6%. Of these 69% were identified as being in the healthy weight range, 19% in the 'at risk' group and 12% in the overweight group. The proportion of children in each category was not different across the four schools. The trends observed in this study suggest that childhood overweight and obesity in the 4 Hunter schools included in the study are similar to national statistics. However, due to the low consent rate of parents, it is difficult to determine if the children measured are truly representative of the schools' populations, therefore results cannot be generalised.

In conclusion, further studies are needed in the Hunter region to determine the true prevalence of overweight and obesity in school children. Methods of approaching parents and children need to address the low response rate for future prevalence estimates.

References

1. Margarey AM, Daniels LA, Boulton TJC. Prevalence of overweight and obesity in Australian children and adolescents: reassessment of 1985–1995 data against new standard international definitions. MJA 2001; 174: 561–564.
2. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a Standard Definition for Childhood Overweight and Obesity: International Survey. BMJ 2000; 320: 1240–1243.

Supported by a grant from the John Hunter Children's Hospital and Hunter Medical Research Institute
Key words: overweight, children, prevalence

Congruence of red meat descriptors reported by a group of elderly volunteers and those found in an Australian nutrient database

LC Tapsell, L Gillen, AT McMahon, IF Gutteridge, AJ Owen

Smart Foods Centre, University of Wollongong, NSW, 2522

The role of red meat in the Australian diet has been the subject of substantial interest in both academic and industry circles (1) with further research expanding nutrition knowledge and its applications. However, in studies of dietary habits the issue of accuracy of assessments warrants attention. Some of the problems relate to discrepancies between consumer accounts of foods consumed and the descriptors used in food databases. The aim of this paper is to describe the categories of red meat reported by elderly subjects in a trial involving red meat consumption, and to identify potential sources of error in using this data as the basis for intervention.

Thirty four elderly adults (mean age 68.1 years, range 61–80 yrs) volunteered to participate in an intervention trial with 3 dietitians undertaking a detailed open ended diet history interview where subjects described their eating patterns in everyday terms (2). A checklist was included at the end of the interview, summarizing meat types, amounts and frequency of consumption. Participants reported 5, 4 and 1 main categories of beef, lamb and veal respectively. Within the beef and lamb categories there were a further 15 and 7 sub-categories respectively. The 3 main categories reported were rump steak, mince (with 'premium' regularly used as a qualifier) and short loin lamb chops. Participants reportedly consumed meat at dinner in the evening at home, with the exception of one male who ate lunches at clubs twice a week and another who chose meat pie for lunch once a week. Five individuals and 3 couples reported consuming small amounts of cold meats (60 g) at the lunch meal 1–3 times per week and only one subject reported eating meat in a takeaway breakfast twice a week. The AUSNUT (ANZFA, 1999) database in the Foodworks nutrient analysis software program (version 3.00, 2002. Xyris software, Brisbane) indicated 12 options for rump steak: 3 raw and 9 cooked (3 fried, 3 grilled, 3 non-specified, NS), and each with fat descriptors of fat trimmed, lean & fat, and NS fat trimmed. If barbecued rump steak were itemized a potential difference of 6 g fat and 300 kJ could result from the average serve of the study sample if the fat specification were not accurately determined.

Volunteers for our dietary study generally reported meat categories congruent with descriptors in the current database. Three quarters reported consuming mixed dishes and all but 2 gave some description of fat removal. This and the relative stability of their eating patterns appear to reflect the age of the group. Nevertheless, the risk of measurement error still lies with elucidating recipes for mixed dishes, and in the accurate assessment of the fat content in consumed meats.

Beef descriptors					Lamb descriptors				Veal descriptors
Grilled	Mince	Roast	Cuts	Boiled	Chops	Roast	Fillet	Liver	Fillet
Rump	Premium	Topside	Blade	Corned beef	Shortloin	Leg	Lamb fillet	Lamb's fry	Snitzel
Fillet	Sausage	Sirloin	Chuck		Foreqtr	Joint			
T-bone	Rissoles	B Blade	Topside		Cutlets				
Sirloin	Meat pie								

References

1. Meat and Livestock Australia. The role of red meat in the Australian diet. MLA. Sydney. 2001.
2. Tapsell, LC, Brenninger, VL and Barnard, JA. Applying conversation analysis to support accurate reporting in the diet history interview. JADA. 2001; 100: 818–824.

Supported by: Meat and Livestock Australia
Key words: dietary assessment, nutrient databases, food descriptors

Comparison of fat intake in Australian and New Zealand CHD patients: the LIPID (long term intervention with Pravastatin in ischaemic disease) study

P Nestel[1], K Baghurst[2], A Kirby[3], DM Colquhoun[4], H White[5], R Stewart[5], P Glasziou[6], S Pirozzo[6]

[1]*Baker Research Institute, Melbourne*
[2]*CSIRO Health Sciences and Nutrition, Adelaide*
[3]*NH & MRC Clinical Trials Centre, University of Sydney*
[4]*Core Research Group, Brisbane*
[5]*Green Lane Hospital, Auckland*
[6]*University of Queensland, Brisbane*

Background: Patients with coronary heart disease (CHD) are advised to reduce their intake of foods rich in saturated fatty acids (SFA) and substitute foods rich in complex carbohydrates, polyunsaturated fatty acids (PUFA) and monounsaturated fatty acids (MUFA).

Methods: In the LIPID Study, 9014 patients were randomised to Pravastatin 40mg/day or placebo and followed up for a mean of 6 years. Dietary advice was given to all patients to reduce fat intake to less than 30% of energy intake, with equal amounts of SFA, PUFA and MUFA. Mortality and CHD event rates were higher in New Zealand patients. Baseline characteristics of patients were similar in both countries. However in New Zealand, median low-density lipoprotein cholesterol was higher (3.95 vs 3.81 mmol/L, P = 0.0002) and high-density lipoprotein cholesterol was lower (0.88 vs 0.94 mmol/L, P < 0.001). A validated questionnaire was used to assess adherence to the diet in three different years (1991, 1992, 1995) in 1077, 951 and 849 patients respectively.

Diet Components	Australia		New Zealand		P
	n	Mean ± SD	n	mean ± SD	
Energy 1991 (MJ/day)	684	8 ± 2	393	9 ± 2	<0.001
Energy 1995 (MJ/day)	532	7 ± 2	317	8 ± 2	0.001
SFA 1991 (g/day)	684	23 ± 11	393	28 ± 13	<0.001
SFA 1995 (g/day)	532	22 ± 9	317	24 ± 9	0.001
MUFA 1991 (g/day)	684	22 ± 10	393	25 ± 9	<0.001
MUFA 1995 (g/day)	532	21 ± 8	317	22 ± 7	0.002
PUFA 1991 (g/day)	684	13 ± 7	393	14 ± 7	0.001
PUFA 1995 (g/day)	532	12 ± 6	317	13 ± 6	0.03

Results: Total energy intake was higher in New Zealand in 1991, 1992 and 1995 and declined in both countries. Total fat and SFA and MUFA in absolute amounts and as a percentage of energy intake were higher in New Zealand. PUFA, as a percentage of energy intake, were similar in both countries.

Conclusions: The higher intake of fat in New Zealand patients may partly explain the higher CHD event rates observed in the LIPID Study.

Key words: New Zealand, CHD, energy intake