



## Plenary 3: Nutritional Epidemiology and Chronic Diseases

### Diet-related chronic diseases in Indigenous Australian populations

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#### Background

Lifestyle related chronic diseases (type 2 diabetes and associated conditions including vascular disease and kidney failure) are causing most damage in Indigenous Australian populations in what should be the most productive years of life. For example, in the 20-50 year age group diabetes is 10-15 times higher and deaths from heart disease 10 times higher than for other Australians. While in the general population 76% of acute myocardial infarctions (AMI) occur in people over 65 years, almost 80% of AMIs among Indigenous people occur before age 65.

#### Risk factors

Centralised patterns of fat distribution are common in both men and women, and are strongly linked to increased risk of metabolic syndrome and diabetes, which are both strong risk factors for vascular disease and kidney failure. These serious conditions frequently occur together in the same individuals and interact to amplify mortality risk. Major risk factors include high rates of smoking (50-70%), low birth weight, and poor quality diet. Post streptococcal glomerulonephritis is associated specifically with risk of kidney disease. The role of background burden of infectious disease in potentially amplifying the risk of chronic diseases in disadvantaged populations needs to be investigated systematically.

Recent data suggests that the prevalence of overweight and obesity is rising particularly among young women, with average weight gain of 1.5 kg per year being reported for young women in north Queensland over a 6 year period, in association with an incidence of diabetes of 29 cases/1000 p.y. This raises the possibility of diabetes in pregnancy leading to serious adverse health outcomes (early onset obesity and diabetes in the offspring) in future generations.

Extremely poor nutrition across the lifespan, particularly in remote communities, where very low incomes combine with limited availability and high cost of fresh high quality food, is a major driver of these serious health outcomes. Low income people in affluent societies tend to maximize calories/\$, and therefore choose the low cost foods that are rich in fat, sugar and salt. Over the past three decades the cost differential between healthy foods and unhealthy processed foods rich in fat and sugar has widened, in parallel with the rising prevalence of obesity (throughout the world).

#### Examples of interventions

There are a number of examples of significant improvements in diet quality in remote Indigenous

communities which have been achieved through close partnerships with the communities. Improved quality of food sold in stores, has been shown to be associated with improved biomarkers of both diet and health status at the population level.

#### Implications

However, what is clear is that improvements are limited and generally not sustained after the intervention ceases, and health status remains well below the average for Australia. Indeed the gap is widening for this population, as mortality has declined generally in the mainstream. Thus while health promotion activities in relation to healthy diets have an important role to play, they cannot succeed without also addressing fundamental systemic issues of availability, cost and sustainability. It is now abundantly clear that system-level change to improve nutrition and health trajectories is urgently needed.

#### System level change?

Many people in remote Indigenous communities rely on a local store for most of their food needs. The stores are of variable quality, often poorly managed and their products expensive, particularly perishable items such as fresh fruit and vegetables. However, a single food outlet in a small community offers unique opportunities for change not possible currently in larger towns and cities. There are two organisations now managing a growing number of remote stores: ALPA (Arnhem Land Progress Association) and Outback Stores. They are committed to quality improvement in management practices and food supply, with any profits going back to the communities. The government, through FaHCSIA, is committed to improving food security in remote Indigenous communities. Together, these developments provide an exciting opportunity (in close collaboration with the participating communities) to evaluate new approaches to enhancing the quality of the food supply, with the aim of improving health status over time. Possible approaches under discussion include price discounting on healthy foods; 'taxes' on unhealthy products; the introduction of healthy take-aways; breakfast and lunch programs for pre-school and primary school children; informative food labelling; and financial rewards for healthy purchasing.

Such system change elements offer great promise as an efficient and sustainable approach to improving food intake in Indigenous communities.

#### Sources of funding

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## Plenary 3: Nutritional Epidemiology and Chronic Diseases

### Optimal dietary approaches for prevention of chronic disease: a lifecourse perspective

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For the past three decades, health authorities have said 'eat less fat, especially saturated fat'. Australians have obediently reduced their fat intake, yet over the same timeframe, the prevalence of obesity has more than doubled, type 2 diabetes has tripled, and cardiovascular disease is still the number one cause of death. The nutritional landscape has changed.

Recent systematic reviews and meta-analyses of prospective epidemiologic studies have concluded that there is no significant evidence for concluding that dietary saturated fat is associated with an increased risk of coronary heart disease or cardiovascular disease.

A new paradigm is arising: that the processed carbohydrates which replaced the energy from fat, may increase the risk of obesity, diabetes and heart disease more so than fat or saturated fat, – a finding that has enormous implications for health professionals and the Australian food and agricultural industry.

Both quantity and quality of carbohydrate are relevant to the debate. Sugars and starches, with or without fibre, can produce adverse effects on blood glucose levels after consumption, a characteristic that reflects their rate of digestion and absorption, and is assessed as their 'glycaemic index' or 'glycaemic load' per serving.

Alternate dietary approaches, including high protein, Mediterranean-style and low glycaemic load diets, have been shown to improve weight control and cardiovascular outcomes more so than conventional low-fat, high-carbohydrate diets. These alternate diets share an under-recognised unifying mechanism: the reduction of postprandial glycaemia and insulinaemia.

Similarly, epidemiological evidence shows that food patterns associated with increased risk of adiposity and diabetes include specific carbohydrate choices (potatoes, refined grains, white bread, sugar-sweetened beverages, sweets, fast foods) that provoke higher fluctuations in blood glucose and insulin.

The insulin resistance and compensatory hyperinsulinaemia, exacerbated by overweight, that occurs naturally during critical life stages such as pregnancy and puberty, increase the burden on the beta-cells that produce insulin.

The recent rise in gestational diabetes and type 2 diabetes in young people may therefore be traced to food patterns that are currently encouraged but exaggerate postprandial glycaemia and insulinaemia.

Clearly, diets high in either saturated fats or refined carbohydrates are not suitable for cardiovascular disease prevention. However, high glycaemic index carbohydrates in particular are likely to cause even greater metabolic damage than saturated fat in the typical sedentary and overweight Australian.

While intake of saturated and trans fats should remain at low, a singular focus on reduction of total and saturated fat is counterproductive because dietary fat is typically replaced by high glycaemic index carbohydrate.

According to Frank Hu, Harvard Professor of Public Health 'in this era of widespread obesity and insulin resistance, the time has come to shift the focus of the diet-heart paradigm away from restricted fat intake and toward reduced consumption of refined carbohydrates'.

Our view is that the most evidence-based dietary strategy for prevention of obesity, type 2 diabetes and cardiovascular disease is not the accepted low-fat, high carbohydrate diet, but 'alternate' dietary approaches (low glycaemic index, Mediterranean-style and high protein, moderate carbohydrate diets) that reduce postprandial glycaemia and insulinaemia, without adverse effects on other risk factors.



## Concurrent Session 10: Vascular Disorders

### Endothelial dysfunction in patients with insulin resistance

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There is growing recognition that the vascular endothelium plays a central role in the regulation of vascular homeostasis. Abnormalities of endothelium-dependent vasodilation are detectable at all stages of atherosclerosis in experimental models and in human subjects. In addition to loss of the bioactivity of endothelium-derived nitric oxide, atherosclerosis and cardiovascular disease risk factors are also associated with abnormalities of many other aspects of "endothelial function" including control of thrombosis, inflammation, and intimal growth. Patients with syndromes of insulin resistance, including Type 2 diabetes mellitus, obesity, and the metabolic syndrome, have increased risk for cardiovascular disease, and such patients also display endothelial dysfunction. For example, cross-sectional studies have demonstrated loss of brachial artery flow-mediated dilation in patients with diabetes mellitus, the metabolic syndrome, and elevated HOMA-IR index. We recently observed the rapid onset of endothelial dysfunction in the setting of acute insulin resistance induced by a short period of bed rest. Insulin resistance and endothelial dysfunction also develop during infusion of Intralipid and heparin, which increases circulating free fatty acids. Insulin sensitizing drugs improve endothelial function in patients with diabetes mellitus, but not prevent the development of endothelial dysfunction produced by bed rest or Intra-lipid infusion. Ongoing studies are investigating the contribution of endothelial activation to endothelial dysfunction in these settings. We also have recently obtained evidence linking dysfunction of the mitochondria to endothelial dysfunction in the setting of insulin resistance and Type 2 diabetes mellitus. These studies provide insight the mechanistic links between syndromes of insulin resistance and cardiovascular disease and may suggest new approaches for patient management.

### Effects of a high salt meal on flow-mediated dilatation

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#### Background

Dietary salt is related to blood pressure and cardiovascular disease and has been shown to impair vascular function short-term. The effect of salt on endothelial function post-prandially is unknown but is of potential importance because of the high levels of salt consumed in commonly eaten foods.

#### Objective

To examine whether a high salt meal has a detrimental effect on post-prandial endothelial function in healthy subjects.

#### Design

Sixteen healthy normotensive subjects received a high salt meal (HSM) (65mmol Na) and a control low salt meal (LSM) (5mmol Na) on two separate occasions in a randomised order. Endothelial function was measured fasting and post-prandially at 30, 60, 90 and 120 minutes using flow-mediated dilatation (FMD) and reactive hyperaemia peripheral arterial tonometry. Blood pressure (BP) was also measured.

#### Outcomes

Overall FMD was reduced 2 hours post-prandially. At 30 and 60 minutes FMD was significantly impaired after the HSM compared with the LSM (6.05±3.21% LSM vs. 3.39±2.44% HSM, P=0.006 at 30min and 4.64±2.48% LSM vs. 2.20±2.77% HSM, P=0.003 at 60min). No differences in BP or reactive hyperaemia index were observed between meals.

#### Conclusion

A high salt meal, typical of the amount of salt consumed in a commonly eaten meal, can significantly suppress brachial artery FMD within 30 minutes. This suggests high salt intakes may have detrimental effects on endothelial function in the short-term and which may increase the risk of vascular disease.

#### Source of funding

CSIRO Preventative Health Flagship



## Concurrent Session 10: Vascular Disorders

### Flavonoid-rich apples and nitrate-rich spinach augment nitric oxide status and improve endothelial function in healthy men and women

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#### Background

Dietary flavonoids and nitrates have independently been shown to augment nitric oxide (NO) status and improve endothelial function, factors related to cardiovascular health. Available data suggest that flavonoids and nitrates augment NO status via different mechanisms. Apples are an important dietary source of flavonoids and spinach is rich in nitrate. The acute effects of these foods on NO status and endothelial function have yet to be assessed.

#### Objective

To investigate the independent and additive effects of flavonoid-rich apples and nitrate-rich spinach on NO status and endothelial function.

#### Design

Healthy participants (n=30) were recruited to a randomised, controlled, cross-over (latin square) trial. The acute effects of apple and spinach, independently and in combination were compared to energy-matched control. Primary outcome measures were on plasma NO status (NO<sub>x</sub>) and endothelium function by measuring flow mediated dilatation (FMD) of the brachial artery.

#### Outcomes

A significant increase in NO<sub>x</sub> ( $P < 0.01$ ) was observed with apple (126.74  $\mu$ M, 95% CI: 100.41, 159.97), spinach (193.66  $\mu$ M, 95% CI: 153.09, 244.96), and apple+spinach (157.35  $\mu$ M, 95% CI: 124.75, 198.50) treatments when compared to the control (71.42  $\mu$ M, 95% CI: 56.66, 90.04) treatment. Similarly a significant increase in FMD ( $P < 0.05$ ) was observed with apple (5.48%, 95%CI: 4.42, 6.54), spinach (4.90%, 95%CI: 3.84, 5.96) and apple+spinach (5.30%, 95%CI: 4.25, 6.35) treatments when compared with the control (4.36%, 95%CI: 3.30, 5.41) treatment.

#### Conclusion

Flavonoid-rich apples and nitrate-rich spinach acutely improve nitric oxide status and endothelium function, factors which may benefit cardiovascular health. These effects were not additive.

#### Source of funding

National Health and Medical Research Council (NHMRC) and the Australian Research Council (ARC).

### The effect of canola oil intake on oxidative stress in the vasculature

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#### Background

Canola oil (CO) intake as the only dietary fat source shortens the life-span of stroke-prone spontaneously hypertensive rats (SHRSP). Oxidative stress (OS) as a possible mechanism leading to the acceleration of hypertension-related deterioration of organs in the SHRSP rat has not been investigated previously. Oxidative stress leads to vascular damage and plays a critical role in the pathogenesis of cardiovascular diseases such as hypertension. In vascular cells, NADPH oxidase (NOX) is a major source of reactive oxygen species (ROS) and is therefore implicated in endothelial dysfunction.

#### Objective

To investigate the effect of CO intake on the gene expression of NOX subunits and superoxide dismutase (SOD) isoforms in the aorta of SHRSP rats.

#### Design

Male SHRSP rats were fed: 10 wt/wt% CO with water; 10 wt/wt% soybean oil (SO) with water; 10 wt/wt% CO with 1% NaCl in water; or 10 wt/wt% SO with 1% NaCl in water for 25 days. Blood pressure was measured weekly. The aorta was removed and gene expression analysed for: NOX2 and p22phox (NOX subunits), mitochondrial SOD (MnSOD), cytosolic SOD (CuZn-SOD) and extracellular SOD (eSOD).

#### Outcomes

Blood pressure increased over time in all diet groups. CO intake with salt significantly reduced ( $P < 0.05$ ) MnSOD, CuZn-SOD and eSOD gene expression compared to the SO with salt and the CO group. CO intake with salt significantly reduced ( $P < 0.05$ ) NOX2 gene expression compared to the SO with salt and CO group. CO intake with salt significantly reduced ( $P < 0.05$ ) p22phox gene expression compared to the CO group. The CO and SO groups with salt had significantly increased ( $P < 0.05$ ) water consumption compared to the SO and CO groups.

#### Conclusion

CO along with salt modulates the gene expression of NOX and SOD in the aorta. In contrast to the hypothesised outcomes, the combination of the reduced pro/anti-oxidant proteins may reflect a decrease in OS in the vasculature in this salt loaded hypertensive model due to CO.

#### Source of funding

Not applicable.



## Concurrent Session 10: Vascular Disorders

### A whole animal model for the rapid assessment of potential bioactives for blood pressure lowering

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#### Background

Diet based strategies to lower blood pressure (BP) are becoming popular and bioactives with potential therapeutic roles are increasingly being sought by the food and pharmaceutical industry. For example, several natural compounds that modulate the renin-angiotensin-aldosterone system and/or peripheral vascular resistance have been isolated, and products formulated.

#### Objective

To develop a rapid whole animal model for the evaluation of anti-hypertensive effects of potential bioactive compounds following oral administration and validation of the model using purified polyphenols.

#### Design

BP of the conscious adult spontaneously hypertensive rats (SHR) were monitored over a 48 h period following single bolus dose administered orally. Changes in BP were monitored (tail-cuff method) at 3, 6, 9, 24 and 48 h post administration. The therapeutic agent enalapril (4 mg/kg) was used as the positive control, saline as the negative control and three dose levels of chlorogenic acid (CGA), a polyphenol found in coffee, as the test compound. Potential of CGA to lower vascular resistance was also assessed using *in vitro* vascular preparations (aortic rings and perfused mesenteric vascular bed).

#### Outcomes

Enalapril (4mg/kg) reduced BP in the 3-9 h time window (8-12% vs saline control), with BP values returning to baseline by the 24 h point. Similarly, CGA (75 and 125 mg/kg lowered BP ( $P < 0.05$  or better) at 3, 6, 9 and 24 h post administration whilst at 250mg/kg a lower BP was evident even at the 48 h time point ( $P < 0.01$ ). The greatest reduction in BP after CGA was noted at the 3-6 h time window (18-21%,  $P < 0.05$ ). CGA caused different actions on the isolated vascular preparations pre-contracted with phenylephrine suggesting that the changes in peripheral vascular resistance may not fully account for the BP lowering actions of CGA. Preliminary screening studies of several candidate polyphenolic bioactives showed the usefulness of this approach where the limited sample availability at the discovery stage may not allow conduct of long-term feeding trials.

#### Conclusion

The acute BP model is an appropriate experimental tool in bioactive discovery and substantiation. The positive outcomes with CGA and test bioactives also demonstrate that certain polyphenolic compounds are rapidly absorbed and bioavailable following dietary intake.

**Source of funding:** Not applicable

### Screening for *in vitro* ACE inhibition and angiotensin II receptor blocking activity across edible plant families

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#### Background

Over many years there have been reports from traditional and complementary medicine that individual foods may influence blood pressure via the Renin-Angiotensin-System (RAS). However, there has not been a systematic approach to investigating the capacity of edible plants in modulating key aspects of RAS particularly in foods found in the typical Australian market place.

#### Objective

To investigate the effects of extracts from a range of edible plants families for their ability to target 2 key sites in *in vitro* of the RAS that regulate BP. The 2 sites were the angiotensin converting enzyme (ACE) and the angiotensin II receptor (AT<sub>1</sub>R) systems.

#### Design

Up to 138 plant, fungi or yeast species or varieties from 44 families were sourced from Australian markets, processed according to in-house methodology and tested for their ability in relatively high throughput assays to: 1) inhibit porcine kidney ACE activity using a Cobas<sup>®</sup> based system and 2) compete for [<sup>125</sup>I-Sar<sup>1</sup>-Ile<sup>8</sup>]-angiotensin II binding at the AT<sub>1</sub>R from rat liver tissue using membrane filtration apparatus.

#### Outcomes

ACE inhibitory activity of over 50% was found in 13 families from a variety of species with IC<sub>50</sub> values as mg/mL (family) of 0.18 for cinnamon (Lauraceae), 0.19 for tea leaf (Theaceae), 0.33 for grape 0.04 for grape seed extract (Vitaceae), 0.58 for Illawarra plum (Podocarpaceae) and 0.68 for coconut (Sterculiaceae). Five families produced plant extracts with over 50% inhibitory values, IC<sub>50</sub> as mg/mL, for blocking the AT<sub>1</sub>R. These were 1.5 for rosemary (Lamiaceae), 2.1 for Madura green tea (Theaceae), 2.8 for spring onion (Alliaceae), 4.0 for coriander leaf (Apiaceae), and 4.2 for watercress (Brassicaceae). There was no correlation between plant families or species exhibiting ACE inhibition or AT<sub>1</sub>R blockade. However, in plant subsamples there was a correlation between total phenolic content and ACE inhibitory activity ( $r^2 = 0.493$ ,  $P = 0.002$ ).

#### Conclusion

These findings suggest that a variety of plant based foods display the ability *in vitro* to modulate key components of RAS. It remains to be determined what the physiological significance of this activity plays in the control of blood pressure in animal models of hypertension.

#### Source of funding

Not applicable



## Concurrent Session 10: Vascular Disorders

### The effect of fructose and glucose on cerebrovascular integrity: implications for Alzheimer's disease risk

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#### Background

Alzheimer's disease (AD) is a neurodegenerative disease characterised by the formation of cerebral amyloid plaque and neuro-inflammation. Previously, we showed that a diet high in saturated fats compromise cerebrovascular integrity, possibly as a consequence of exaggerated exposure of plasma amyloid- $\beta$  ( $A\beta$ ) complexed to triglyceride-rich-lipoproteins (TRL's). Cerebral extravasation of plasma proteins was evident, including the influx and the extracellular retention of TRL- $A\beta$ . Hypertriglyceridemia is commonly reported in subjects with insulin-resistance and diabetes is a risk factor for AD. The mechanisms for this association are presently unknown, but we hypothesise that TRL- $A\beta$  may induce disturbances in cerebrovascular function.

#### Objective

To explore if insulin resistance induces disturbances in cerebrovascular function.

#### Design

Wild-type model rats were randomised to either a control low-fat diet, a high saturated fat diet (positive control), an insulin-resistant diet (fructose supplemented), or control carbohydrate diet (glucose supplemented). At 2 and 4 weeks following commencement of diets, parenchymal extravasation of plasma proteins was determined by 3-D immunofluorescent microscopy.

#### Outcomes

Blood-to-brain leakage of the plasma protein IgG and the lipoprotein marker apolipoprotein B was found in rats fed either the SFA > fructose and glucose fed rats, but not in low-fat fed controls. Fructose fed rats were hypertriglyceridemic, however extravasation of plasma proteins in glucose fed rats appeared to be independent of changes in plasma lipids.

#### Conclusion

Disturbances in cerebrovascular function in fructose fed rats was associated with plasma TRL, however this may not be causally related.

#### Source of funding

NHMRC project grant 533531.



## Concurrent Session 11: Nutrition Policy and Behaviour

### How important is neighbourhood access and availability of foods in influencing intakes?

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#### Background

While a considerable body of research has examined the influence of personal factors (eg knowledge, attitudes, beliefs, preferences) and social factors (eg social support by family and peers) on food intake, less work has focused on factors in the local neighbourhood environment (eg access, availability, cost) that might impact on consumption. This is despite the fact that the environment is a potentially strong determinant of eating behaviours and structural change, such as increasing access and availability, or reducing the cost of healthy foods, has been proposed as means of promoting healthy eating.

#### Objective

The aim of this paper is to examine Australian evidence of the relation between access, availability and cost with fruit and vegetable consumption among women and children.

#### Design

We have conducted a number of population studies in Victoria, Australia that have examined the association of structural level factors with fruit and vegetable consumption among women and children. This paper will present findings from the HEAPS study (800 children), the SESAW study, (1500 women; in HEAPS and SESAW participants were selected from neighbourhoods across the socioeconomic spectrum), and the READI study (4300 women living in disadvantaged neighbourhoods). Each gathered sociodemographic, behavioural and other data on individual and social variables, and used Geographic Information Systems to objectively assess each individual's neighbourhood food environment. In the SESAW study we also gathered information on the availability and price of fruits and vegetables in stores.

#### Outcomes

Analyses of the HEAPS data on children showed that the more fast food outlets and convenience stores there were close to home, the lower was the likelihood of consuming fruit two or more times/day. There was also an inverse relation between density of convenience stores and the likelihood of consuming vegetables three or more times/day. The likelihood of consuming vegetables three or more times/day was greater the further away children lived from a supermarket or a fast food outlet.

In READI we examined whether poorer access to major supermarkets, smaller supermarkets and fruit and vegetable stores in local neighbourhoods was associated

with lower intakes of fruits and vegetables. Six variables were used to assess access. None of our measures of access were associated with vegetable intake, and only one (greater distance to the nearest fruit and vegetable store) was associated with lower fruit consumption.

In the SESAW study we examined the role of individual, social and neighbourhood factors as mediators of the relation between socioeconomic status (SES) and fruit and vegetable consumption. We found that while a number of the individual and social variables partly explained SES differences in consumption, store density did not mediate the relation between SES and fruit or vegetable intakes.

In the SESAW study we also considered whether differences in intake across socioeconomically diverse neighbourhoods could be explained by the availability and price of fruits and vegetables in those neighbourhoods or by store opening hours. Fruit intake did not vary by neighbourhood, while vegetable intake was lower among women living in disadvantages neighbourhoods. However the availability and price of vegetables and store opening hours did not explain neighbourhood differences in vegetable intake.

#### Conclusion

These findings, along with those from other studies conducted internationally, highlight the complexity of understanding the role of neighbourhood food environments as a determinant of fruit and vegetable consumption. Future research, and policies and programs aimed at understanding and influencing the food environment, need to consider a broader range of contextual factors that impact on food choice and to better understand the ways in which individuals interact with their local neighbourhood environments.

#### Source of funding

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## Concurrent Session 11: Nutrition Policy and Behaviour

### Habitual fat intake modulates fat taste sensitivity in lean and overweight subjects

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#### Background

Individual sensitivity to certain tastants influences food consumption and preferences. Dietary fats are 'tasted' within the mouth, and differences in fat taste sensitivity may be associated with certain dietary behaviours such as fat consumption. The determinants of oral sensitivity, and whether they emerge via genetic predisposition, or environmental factors, such as habituation following excess consumption of fat remain unclear.

#### Objective

To assess the influence of exposure to high- and low-fat diets on taste sensitivity to oleic acid (C18:1) in lean and overweight/obese subjects.

#### Design

Subjects (n=32), lean; (n=20; BMI: 22.6±0.2 kg/m<sup>2</sup>, and overweight/obese (n=12; BMI: 28±0.2 kg/m<sup>2</sup>), participated in a randomised, 10 week crossover dietary intervention during which a high-fat (HF) (>45% fat) and low-fat (LF) (<20% fat) diet were consumed over a 4 week period, with a 2 week washout between. Weight, height and taste sensitivity to C18:1 were determined at baseline (BL), and the end of each intervention. Repeated measures ANOVA was used to determine effects of the intervention on taste threshold.

#### Outcomes

There was a significant time\*diet interaction [ $F(1,29)=8.7$ ,  $P=0.006$ ] indicating that taste thresholds were differentially affected by the HF and LF diets, which increased following the HF diet (BL: 3.5±0.1, END: 5.2±0.14 mM) and decreased following the LF diet (BL: 4.9±0.14, END: 2.6±0.09 mM). A significant diet\*BMI interaction [ $F(1,29)=5.1$ ,  $P=0.03$ ] was detected indicating lean and obese/overweight subjects responded differently to the diets, with effects being more robust in lean (HF,BL: 2.8±0.16, END: 6±0.22 mM; LF,BL: 4.5±0.2, END: 2.4±0.1 mM) compared to obese/overweight subjects (HF, BL: 4.6 ± 0.35, END: 3.9±0.08, LF, BL: 5.5±0.4, END: 2.9±0.3 mM).

#### Conclusion

This study reveals a level of plasticity for C18:1 taste thresholds in lean and obese subjects that can be modulated by diet. Of interest was that the effects were more robust in lean, in comparison to obese/overweight subjects indicating differences in the oral processing of fat, which may be associated with development of overweight and obesity.

#### Funding source

C-PAN, Deakin University and Food Futures Flagship, CSIRO.

### Encouraging children to eat vegetables: the effects of taste exposure and reward

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#### Background

A recent Australian national survey found that only 25% of children aged 2 - 8 years meet Australian vegetable intake recommendations. European survey data on children aged 11 years found that whilst there was variation across countries, all intakes were below population goals and food-based dietary guidelines.

Innate dislike of bitter taste is a barrier to children's intakes of vegetables.

#### Objective

The present study sought to overcome taste aversion through exposure. It was unknown if an immediate non-food reward (stickers) would have positive or negative effects on short and medium-term ratings of liking.

#### Design

A randomised controlled trial was undertaken with 3 groups: control, exposure (EO), and exposure + sticker reward (E+R). Taste assessments were undertaken in the home and parents implemented the interventions. Based on taste assessment of 6 parent-selected vegetables, the 4th ranked vegetable was offered everyday for 14 days (target vegetable). Ratings were assessed at baseline, 2 weeks (post-intervention), 4 weeks and 3 months (follow-up).

#### Outcomes

At baseline there were 185 children (aged 4 - 7 years) and parents and 164 (87%) at 3 month follow-up. Data analysis was undertaken on children who complied with a minimum of 9 tastings (70%).

From baseline to post-intervention liking increased for both the EO and E+R groups ( $P<0.001$ ) but not for Controls. At post-intervention liking ratings for EO and E+R groups were both significantly higher than Controls but not different from each other ( $P>0.05$ ). There was no further change in liking. Despite having similar effects on liking, the E+R group was able to offer the target vegetable more often, achieve more days of taste exposure, and experienced fewer refusals from the child, compared to the EO group.

#### Conclusion

An immediate non-food reward was effective in assisting parents to carry out a taste exposure strategy and does not negatively impact in the short or medium term

#### Source of funding

This project was funded by Horticulture Australia Limited using the vegetable levy and matched funding from the Australian Government.





## Concurrent Session 11: Nutrition Policy and Behaviour

### Do antioxidants in dark chocolate, red wine and coffee improve cardiovascular health? Introducing a Heart Foundation position statement

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#### Background

There are many foods, drinks and supplements that claim to have cardiovascular benefits due to their antioxidant content. The Heart Foundation identified widespread consumer and health professional confusion around the mechanisms and health benefits of antioxidants; in particular vitamin C, vitamin E, beta-carotene and polyphenols. Some of the discussion has stemmed from the hypothesis that antioxidants can protect against various processes that lead to atherosclerosis.

#### Objective

In 2009 the Heart Foundation began our review of the international scientific evidence around antioxidants in supplements and commonly consumed food and drinks: fruit and vegetables, tea, cocoa and chocolate, coffee and red wine. Our position statement *Antioxidants in food, drinks and supplements for cardiovascular health* hoped to clarify the role of these foods and drinks in providing antioxidants, as well as their effect on cardiovascular risk factors.

#### Outcomes

Synthesis of the evidence showed some mixed outcomes for cardiovascular risk factors and the prevention of heart disease. The review found that a balanced diet with a wide variety of plant-based foods will provide the antioxidants beneficial for cardiovascular health.

#### Conclusion

This review uncovers the misconceptions and facts around the relationship between antioxidants, food, beverages and cardiovascular health. From our learning the Heart Foundation will offer guidance for practicing health professionals and explain the difficulties in recommending dark chocolate, red wine and coffee for cardiovascular health.

#### Source of funding

Not applicable



## Concurrent Session 11: Nutrition Policy and Behaviour

### Infant Feeding Guidelines for Australia

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#### Background

Breastfeeding is the foundation of lifetime nutrition to optimise health, wellbeing and life expectancy. The WHO's International Code of Marketing of Breast-Milk Substitutes (WHO Code 1981), was developed in response to concerns over the effects on infant health of the unregulated promotion of infant formula, particularly in developing countries. Australia supported the WHO Code and our response included the Marketing in Australia of Infant Formula (MAIF) agreement, the National Breastfeeding Strategy and the development of the NHMRC Infant Feeding Guidelines. It is planned to release a new edition of the Infant Feeding Guidelines in 2011. Dietary Guidelines, including the Infant Feeding Guidelines are based on systematic literature reviews, umbrella reviews (consolidation of major international reviews of breastfeeding) and expert opinion. This paper will review some of the major issues that may be considered in the forthcoming Infant Feeding Guidelines.

#### Objective

Current issues in nutrition include factors associated with the initiation and duration of breastfeeding, definitions of breastfeeding and the collection of data, early nutrition and later disease (epigenetics), prelacteal feeds, the new WHO growth reference and its potential impact on breastfeeding, allergy and breastfeeding and the continuing implementation of the WHO "International Code of Marketing of Breast-milk Substitutes". Infant feeding is closely linked to culture and one of the important factors is whether the infant's parents were breastfed. For this reason it is important to also consider infant feeding practices in the countries of origin of mothers giving birth in Australia. The overall aim of this paper is to review studies related to the issues central to developing the new guidelines.

#### Design

Infant feeding is integral to the culture of the country of birth of the mother (and other family members) and the country in which the birth occurs. In Australia the birthplaces of mothers delivering infants in 2008 were Australia NZ 77.8%, United Kingdom 3.3%, India 1.6%, Vietnam 1.6% and China 1.5%. But Australia has citizens from many countries and mothers from other countries contributed 14.2% of births. This makes developing guidelines that are inclusive difficult, but in seeking to promote breastfeeding and the appropriate introduction of complementary foods

One of the most difficult features of discussion of infant are the definitions used for breastfeeding. The definitions used will be taken from the NHMRC Infant Feeding Guidelines, which in turn are consistent with the WHO Code. More specifically breastfeeding is defined as:

**Exclusive Breastfeeding** "the infant has received only breast milk from his/her mother or a wet nurse, or expressed breast milk, and no other liquids or solids with the exception of drops or syrups consisting of vitamins, mineral supplements or medicines"

**Full Breastfeeding** "infants who are receiving almost all of their nutrients from breastmilk but take some other liquids such as water, water-based drinks, oral rehydration solutions, ritual fluids, and drops or syrups.

**Any Breastfeeding.** The infant is receiving some breastmilk.

Unfortunately many potentially useful studies use methods (such as feeding method in the past 24 hours) that do not permit their inclusion in systematic reviews.

#### Outcomes

There have been many recent reviews documenting the importance of breastfeeding for optimal growth and development. It is now known that impact of early development persists into influencing chronic disease rates in adult life. In the shorter term breastfeeding minimises morbidity and mortality, reduces obesity optimising cognitive development.

Infant feeding studies using the same methodology have been undertaken in Australia (several locations, including an indigenous sample, and in several time periods), China (several locations), Vietnam, Japan and Malaysia. Breastfeeding initiation rates remain above 90% in all of these countries. However the UK initiation rate is currently 73%. There is a high, and probably, increasing rate of prelacteal and complementary feeding while in hospital in all of these countries, which contributes to low rates of exclusive breastfeeding.

In all of the countries to be included in the review support by the father and other relatives and making an early decision to breastfeed had a positive influence. Smoking by either parent and the use of prelacteal feeds had a negative impact. Factors that had a varying impact depending on the country included education, c-section, rural or city location and birthweight.

Australia will also have to consider the impact on breastfeeding if the new WHO growth reference is adopted in this country. The new reference is heavier than the old in the first year of life and may misclassify infants as having growth deficits leading to declines in breastfeeding rates.

#### Conclusion

The revision of the Infant Feeding Guidelines will require the careful compilation of systematic literature reviews that will address all of these issues. Draft copies will be released in early 2011 with the final version finished in mid year.

#### Source of funding

NHMRC. Dept of Health and Ageing, Curtin University.



## Concurrent Session 12: Asthma and Gastrointestinal Disorders

### Nutritional approaches to managing inflammation in asthma

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#### Background

The dramatic increase in asthma prevalence in westernised countries in recent decades suggests that environmental factors, such as dietary intake, must play a role in the onset and development of the disease. Key features of a westernised diet are low antioxidant intake, high fat intake and chronic metabolic surplus, resulting in obesity. Each of these factors may contribute to increased asthma prevalence, due to their ability to modulate innate immune responses. A low antioxidant intake increases susceptibility to damage by reactive oxygen species, thus enhancing NF $\kappa$ B-mediated innate immune responses. A high saturated fat intake can heighten innate immune responses, by mechanisms such as activation of toll-like receptor 4, which also leads to an NF $\kappa$ B-driven inflammatory cascade. Furthermore, chronic metabolic surplus, which leads to production of an excess of adipose tissue, leads to the release of proinflammatory mediators such as IL-6, TNF $\alpha$  and CRP, which are central to innate immune pathways. Thus, a western dietary pattern may be highly relevant to activation of the innate immune response, which drives a neutrophilic pattern of inflammation.

Asthma is characterised by airway inflammation, resulting from an exaggerated immune response to triggers such as allergens and viruses. This leads to a worsening of lung function and clinical symptoms. Airway inflammation in asthma is heterogeneous. While allergen-induced T-helper type 2 responses lead to airway eosinophilia, asthma can also involve neutrophilic inflammation, associated with innate immune dysfunction. Neutrophilic asthma is characterised by increased sputum IL-8 and neutrophil influx. Airway neutrophils appear to be clinically important, as most severe forms of asthma involve elevated neutrophils and sputum neutrophils negatively correlate with lung function and airflow obstruction. Thus stimuli that activate innate immune responses and increase airway neutrophilia, are important in asthma.

We have undertaken a series of studies aimed at examining how various dietary factors, including antioxidant intake, fat intake and obesity, modify inflammation and clinical outcomes in asthma.

#### Key Findings

We have shown that antioxidant status is important in asthma. Despite normal dietary intake, circulating levels of antioxidants, including carotenoids and tocopherols, are significantly lower in asthma than healthy controls. We have also shown that this effect is exaggerated in subjects with AHR, uncontrolled asthma or a severe persistent asthma pattern. We have investigated the effect of antioxidant supplementation in various models of airway

inflammation. Using cultured epithelial cells, we examined the effect of lycopene on the inflammatory response to rhinovirus infection. We demonstrated that lycopene supplementation resulted in a 24% reduction in IL-6 release after rhinovirus-1B infection, 31% reduction in IP-10 release after rhinovirus-43 infection, and 85% reduction in rhinovirus-1B replication. In an allergic mouse model, we demonstrated that lycopene supplementation reduced eosinophilic infiltrates in BAL fluid, lung tissue and blood, and mucus secreting cell numbers in the airways. The OVA-specific release of Th2-associated cytokines IL-4 and IL-5 was also reduced. We have also investigated how manipulating antioxidant levels affects asthma outcomes in a clinical setting. Withdrawal of antioxidants from the diet of asthmatics led to a worsening of asthma symptoms, lung function and neutrophilic airway inflammation. Gene expression profiles in these subjects were examined using microarray analysis. Following dietary antioxidant depletion, upregulated genes included the innate immune receptors TLR2, IL1R2, CD93, the signalling molecules IRAK2, IRAK3 and neutrophil proteases MMP25 and CPD. Downregulated genes included those involved in endogenous antioxidant defence (GSTA1, GSTA2) and protease inhibition (SLPI, SERPINB3). Conversely, supplementation of asthmatics with 45 mg/day lycopene, in the form of tomato extract or tomato juice, reduced airway inflammation by suppressing neutrophil influx and reducing neutrophil elastase activity. Our investigations have also revealed an important role for dietary fat in the airways in asthma. Non-obese subjects with asthma were randomised to consume a high fat (48%) or low fat (15%) meal. We observed that a high fat intake was associated with activation of innate immune responses in the airways, including increased airway neutrophilia and TLR4 gene expression. Post-bronchodilator improvement in lung function was also suppressed. The type of fat was important to the effects observed, with a high-trans fatty acid meal (5.2g trans fat) inducing a heightened level of airway neutrophilia compared to a non-trans (<0.3g) fatty acid meal.

We have also been investigating the relationship between obesity and airway inflammation in asthma. We observed that obesity is related to neutrophilic airway inflammation in asthma in women, with sputum %neutrophils being positively associated with BMI. In obese asthmatic males, saturated and monounsaturated fatty acids were important predictors of neutrophilic airway inflammation in asthma.

#### Conclusion

We conclude that various aspects of western diets may contribute to asthma development and progression. Increased antioxidant intake, reduced saturated fat intake and maintenance of a healthy weight may attenuate airway inflammation in asthma, thereby providing a useful addition to current asthma management strategies.

#### Source of funding

NHMRC, Hunter Medical Research Institute, John Hunter Hospital Charitable Trust and Asthma NSW project grants.



## Concurrent Session 12: Asthma and Gastrointestinal Disorders

### Clinical asthma outcomes are improved by caloric restriction and exercise in overweight and obese asthma

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#### Background

An association between obesity and asthma has been described, however interventions examining the reversibility of this association are lacking.

#### Objective

We hypothesise that improvements in body composition, via caloric restriction or physical activity, will improve clinical asthma outcomes in subjects with asthma. The objective of this study was to compare the effect of caloric restriction and/or physical activity in overweight and obese subjects with asthma.

#### Design

Overweight and obese (BMI 28-40kg/m<sup>2</sup>) adults with asthma were randomised to either a calorie restricted diet (n=10), physical activity intervention (n=5), or combined caloric restriction and physical activity intervention (n=9) for 10 weeks. Subjects underwent pre- and post-intervention full body dual-energy x-ray absorptiometry (DXA) and spirometry to assess lung function. Asthma control was measured using the Asthma Control Questionnaire (ACQ).

#### Outcomes

Median (IQR) weight loss was 9.4 (7.4, 10.9)kg in the calorie-restricted group, 1.1 (0.8, 1.5)kg in the physical activity group and 8.1 (6.8, 14.0)kg for the combined intervention. This included a fat mass reduction of 8.5 (6.1, 8.9)kg in the calorie-restricted group, 2.8 (0.9, 3.0)kg in the physical activity group and 8.8 (5.8, 11.0)kg in the combined intervention group. Lung function, measured by FEV<sub>1</sub>, was significantly improved after the physical activity intervention [0.2 (0.1, 0.2)L, *p*=0.04] and FVC after the combined intervention [0.2 (0.0, 0.4)L, *p*=0.004]. ACQ score improved significantly after caloric restriction [-0.6 (-0.3, -0.9), *p*=0.006].

#### Conclusion

Caloric restriction and physical activity have different beneficial effects on clinical asthma measures and body composition in overweight and obese subjects with asthma. Independently and in combination, dietary modification and exercise are useful strategies for providing clinically significant improvements in asthma symptoms.

#### Source of funding

NHMRC Centre for Clinical Research Excellence postgraduate scholarship; Hunter Medical Research Institute Postgraduate student support package, sponsored by the Greaves family.

### Discordant patterns of sorbitol and mannitol malabsorption in patients with Irritable Bowel Syndrome

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#### Background

Malabsorption of dietary polyols can induce gastrointestinal symptoms in patients with Irritable Bowel Syndrome (IBS). Sorbitol and mannitol, hexahydroxy polyol isomers, are found to be widely distributed in food such as stone fruits, cauliflower and mushrooms etc. In the healthy gut, they are partly absorbed by passive diffusion, but their absorption in patients with IBS is unknown.

#### Objective

To compare absorption patterns of sorbitol and mannitol in patients with IBS with those in healthy controls.

#### Design

A randomised, double-blinded, placebo-controlled, crossover study was conducted in 15 healthy and 15 IBS subjects (Rome III criteria). Subjects were randomly challenged with 100 ml solutions of 10 g sorbitol, mannitol and glucose (as control). Breath hydrogen was measured at baseline and at 15-minute intervals for 4 hours. Grams of test polyol malabsorbed was calculated semi-quantitatively via area-under-the-hydrogen-curve of the test polyol relative to that after 15 g lactulose (100% malabsorbed).

#### Outcomes

A similar proportion of healthy (60%) and IBS (53%) subjects had sorbitol malabsorption. In contrast, fewer IBS patients malabsorbed mannitol compared to healthy controls (13% and 53% respectively, *p*=0.02). The amount of mannitol malabsorbed was significantly lower in the IBS group (median [IQR] 0[0-2.0] g) compared to healthy controls (3.4[0.5-9.9] g, *p*=0.008) whilst there was no significant difference in the amount of sorbitol malabsorbed between the two subject groups (IBS 1.1 [0.7-5.3] vs healthy 2.9[0.0-6.3], *p*=0.87). The degree of malabsorption of sorbitol and mannitol correlated significantly in the healthy subjects (Spearman's *r* = 0.70; *p*=0.005), but, in the IBS patients, no correlation was evident (*r* = -0.18; *p*=0.52).

#### Conclusion

Discordant absorption of mannitol and sorbitol occurs in patients with IBS compared with that in healthy controls. This selective absorption of mannitol is not readily explained on the basis of known absorption pathways. It may reflect abnormalities in epithelial function in IBS or selective increase in paracellular permeability due to the difference in their isomeric chemical configuration. Mechanisms warrant further investigation.

#### Source of funding

Not applicable.



## Concurrent Session 12: Asthma and Gastrointestinal Disorders

### Quantification of poorly absorbed short chain carbohydrates in processed grains and cereals: substrates for colonic fermentation and implications for gastrointestinal health

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#### Background

Grains and cereals contain a wide range of potentially protective factors relevant to gastrointestinal health. The best studied prebiotics are fructans (fructooligosaccharides (FOS), inulin) and galactooligosaccharides (GOS). These and other short-chain carbohydrates (SCC) can also be poorly absorbed in the small intestine (named FODMAPs – Fermentable Oligo-, Di-, and Monosaccharides And Polyols) and may have important implications for the health of the gut. They are fermented by colonic microflora to produce by-products including gases and short chain fatty acids and are associated with a wide range of health benefits. Nevertheless, there is a significant proportion of the general population who are intolerant to these FODMAPs, resulting in undesirable gastrointestinal symptoms. There is a paucity of comprehensive data that list the content of FODMAPs including prebiotics - FOS, GOS - in processed grain and cereal products.

#### Objective

To quantify the major SCC using established methodology present in a wide range of processed grain and cereal products commonly consumed in Australia.

#### Design

The FODMAPs; fructose in excess of glucose, FOS (nystose, kestose), GOS (raffinose, stachyose) and sugar polyols (sorbitol, mannitol) were quantified using HPLC with ELSD. Total fructan was quantified using an enzymic hydrolysis method.

#### Outcomes

55 commonly consumed grains, breakfast cereals, breads, pulses and biscuits were analysed. Total fructan were the most common SCC present in cereal grain products and ranged (g/serve as eaten) from 1.12 g in couscous to 0 g in rice; 0.6 g in dark rye bread to 0.07 g in spelt bread; 0.96 g in wheat-free muesli to 0.11 g in oats and 0.81 g in muesli fruit bar to 0.05 g in potato chips.

#### Conclusion

The quantification of FODMAPs including prebiotics - FOS, GOS - show clearly that they are naturally present in a wide range of processed grains and cereals and are important contributors of SCC in the Australian diet. This data will assist research into understanding their physiological properties in the gut.

#### Source of funding

This work was supported by the National Health and Medical Research Council (NHMRC) of Australia

### Low levels of body fat and obstructive airway disease in ex-smokers

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#### Background

Smoking is the most common cause of obstructive airway disease. Differences in body composition may be important in the development of obstructive airway disease in some ex-smokers.

#### Objective

To investigate whether body composition is associated with airflow obstruction in adult ex-smokers.

#### Design

Adult ex-smokers (n=53) with or without airflow obstruction (AFO), defined by post bronchodilator forced expiratory volume in one second/forced vital capacity <0.70, underwent a dual energy x-ray absorptiometry (DEXA) scan to assess total and regional body fat, android to gynoid ratio (And:Gyn) and fat free mass (FFM).

#### Outcomes

Thirteen participants were defined as having AFO with 40 defined as having no AFO. The mean age of participants with and without AFO was 65.8 ± 6.5 (12 male) and 52.3 ± 7.1 (11 male) years respectively. There was no statistically significant difference in smoking history between groups. Ex-smokers with AFO had a lower BMI (P=0.0070), lower total body (P=0.0004) and thoracic region (P=0.0023) percentage fat than those without AFO. The android to gynoid fat ratio was lower in ex-smokers with AFO than those without AFO (P=0.0195). Fat free mass index (FFMI) was not statistically different between the two groups (P=0.1663).

In regression models adjusted for age, sex and smoking history both a low BMI and low android to gynoid fat ratio were significantly associated with more severe AFO.

#### Conclusion

Low levels of body fat, particularly in the android region, may be important in the development of obstructive airway disease in ex-smokers. Mechanisms linking low levels of body fat in ex-smokers who develop AFO require further investigation.

#### Source of funding

Not applicable



## Concurrent Session 12: Asthma and Gastrointestinal Disorders

### Endogenous probiotics are differentially altered in the faecal microbiome of paediatric patients with gastrointestinal viral infections

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#### Background

Rotavirus (RV) is a major cause of acute gastroenteritis (AGE) worldwide, resulting in both morbidity and mortality. Little has been published on the effect of AGE caused by RV and human bocavirus (HBoV2) on the levels of endogenous probiotics within the faecal microbiome.

#### Objective

To investigate changes in selected probiotic species, *Bifidobacterium* spp., *Lactobacillus* spp., and *Bacteroides* spp., in patients with confirmed RV and HBoV2 infection compared with healthy controls.

#### Design

Samples from a study investigating the aetiology of AGE in paediatric patients were retrospectively analysed. Quantitative Real Time PCR was carried out on faecal samples obtained from children with confirmed RV and HBoV2 and healthy controls. Three groups were analysed: RV (n = 32, mean age = 2.1 yr), HBoV2 (n = 32, mean age = 2.3 yr) and controls (n = 32, mean age = 3 yr). Levels of bacteria are expressed in ng of DNA.

#### Outcome

A significant difference (P<0.03) was seen in levels of the probiotic bacteria *Bifidobacterium* spp., between the RV and HBoV2 samples, and no significant differences (P=0.07) were seen between RV, HBoV2 and controls. Significant differences were seen in the probiotic bacteria *Lactobacillus* spp., in RV and controls (P<0.03) and HBoV2 and controls (P<0.02). RV and HBoV2 did not significantly alter (P=0.7) *Bacteroides* spp., compared with controls.

#### Conclusion

The faecal microbiome is fundamental for the health of humans and changes to the microbiome may detrimentally affect the host. This data shows the faecal microbiome was altered in two different gastrointestinal viral infections in children, and this may imply an adaptive response in the faecal microbiome to different viral infections. The implications of these preliminary observations for prevention and targeted treatment of childhood diarrhoea are promising, with the use of selected probiotics (*Bifidobacterium* spp., *Lactobacillus* spp.) likely to be a successful therapeutic approach. Larger studies are now required to confirm this data, and allow optimal design of probiotic intervention trials in the future.

#### Source of funding

PERI centre, MRG, IMVS

### Diarrhoea during enteral nutrition is predicted by the poorly absorbed short-chain carbohydrate (FODMAP) content of the formula

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#### Background

Although it is recognised that diarrhoea commonly complicates enteral nutrition (EN), the causes remain unknown.

#### Objective

To identify factors associated with diarrhoea in patients receiving EN with specific attention to formula composition.

#### Design

Medical histories of inpatients receiving EN were identified by ICD-10-AM coding and randomly selected from the year 2003 to 2008. Clinical and demographic data were extracted. Formulas were classified according to osmolality, fibre and FODMAP (Fermentable Oligo-, Di-, and Mono-saccharides And Polyols) content.

#### Outcomes

Formula FODMAP levels ranged from 10.6 to 36.5 g/day. Of 160 patients receiving EN, 61% had diarrhoea. Univariate analysis showed diarrhoea was associated with length of stay >21 days (OR 4.2), EN duration >11 days (OR 4.0) and antibiotic use (OR 2.1). After adjusting for influencing variables through a logistic regression model, a greater than fivefold reduction in risk of developing diarrhoea was seen in patients initiated on *Isosource*® 1.5 (P=0.029; estimated OR 0.18). The only characteristic unique to this formula was its FODMAP content, being 47-71% lower than any other formula.

#### Conclusion

Length of stay and EN duration independently predicted diarrhoea development, while being initiated on a lower FODMAP formula reduced the likelihood of diarrhoea. As retrospective evaluation does not support a cause-effect relationship, an interventional study investigating FODMAPs in enteral formula is indicated.

#### Source of funding

This study was funded in full by Eastern Health Clinical School, Monash University.



## Concurrent Session 13: Cognition and the Brain

### Effects of omega-3 fatty acids EPA versus DHA on depressive symptoms in elderly people with mild cognitive impairment

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#### Background

Depression is a risk factor for progressing from mild cognitive impairment (MCI) to dementia in elderly people. Omega-3 polyunsaturated fatty acid (n-3 PUFA) supplementation may assist with depressive symptoms although it is unclear whether eicosapentaenoic acid (EPA) or docosahexaenoic acid (DHA) is more effective.

#### Objective

To investigate the impact of daily supplementation with EPA- versus DHA-rich fish oil capsules on depressive symptoms in elderly people with MCI over 6 months.

#### Design

Elderly people > 65 years of age were screened and recruited if they had MCI and had not consumed fish oil supplements during the previous 3 months (N=50). They were block-randomised on age, gender and depressive symptoms using the Geriatric Depression Scale (GDS) to one of the following conditions: 1.6g EPA (+0.15g DHA); 1.55g DHA (+0.40g EPA) or 2.2g linoleic acid (LA placebo) per day. They provided blood samples and completed cognitive assessments and questionnaires at baseline and 6 months (completers: N=36). GDS outcomes are reported here.

#### Outcomes

GDS scores improved in the EPA (0mo  $M=3.86\pm 3.80$ ; 6mo  $M=3.43\pm 3.36$ ) and DHA groups (0mo  $M=3.15\pm 3.08$ ; 6mo  $M=2.54\pm 2.79$ ) and worsened in the LA group (0mo  $M=2.25\pm 1.91$ ; 6mo  $M=3.38\pm 1.85$ ); however these were not statistically significant. After removing one statistical outlier (>3SD above  $M$ ) from the placebo group these differences became significant (EPA  $P=.03$ , DHA  $P=.01$ ).

#### Conclusion

These preliminary data suggest that depressive symptoms might be reduced in elderly people with MCI following supplementation with DHA-rich or EPA-rich fish oil. However the effect size for the DHA-rich oil was stronger and the oil was better tolerated. Supplementation with fish oil containing DHA and EPA may assist in ameliorating elderly people's risk for progressing from MCI to dementia. Future studies with elderly people who have MCI should assess depressive symptoms and provide follow-up of treatment with n-3 PUFA and dementia risk.

#### Source of funding

This research was funded by ARC-Linkage Grant LP776922, 'Cognitive and behaviour benefits of omega-3s across the lifespan', in partnership with Novasel Australia. CM is supported by a Department of Health Scholarship.

### Acute cognitive effects of an oat extract

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#### Background

Oat extracts are traditionally thought to deliver health benefits. Neuravena®, a wild green oat extract, has been shown to increase mental alertness in humans and may influence memory.

#### Objective

A randomised double-blind placebo-controlled crossover trial was undertaken to see whether Neuravena® could acutely improve measures of attention and concentration and the ability to cope with stressful cognitive tasks.

#### Design

Thirty-six volunteers aged  $67 \pm 9$  years (mean  $\pm$  SEM) with BMI  $26.9 \pm 3.8$  kg/m<sup>2</sup> and below average cognitive performance (DemTect score  $14 \pm 2.0$ ) consumed single doses (0, 1600 and 2400 mg) of Neuravena® at weekly intervals. Two cognitive tests were performed 75 mins after consumption of the supplement. The first was the Paced Auditory Serial Addition Task (PASAT) which was used as a mental stress test; heart rate (HR) and blood pressure (BP) were recorded continuously during this test using a Finapres™ instrument. Following this, the Stroop colour word test was performed to measure attention and the ability to suppress task-irrelevant, habitual responses.

#### Outcomes

Both BP and HR increased during the PASAT ( $P<0.001$ ), reflecting mental stress. However there was no significant effect of Neuravena® on these responses. During the Stroop test fewer colour naming errors were made after consuming the 1600 mg dose than after either the 0 mg or 2400 mg doses ( $1.2 \pm 0.5$  compared to  $3.4 \pm 0.1$  and  $3.8 \pm 0.8$  errors respectively,  $P<0.001$ ). In participants with suspected cognitive impairment (DemTect score below 12,  $n=7$ ), the Stroop interference score was also improved by the 1600 mg dose compared with 0 and 2400 mg doses ( $2.3 \pm 0.4$ , compared to  $3.1 \pm 0.5$  and  $2.9 \pm 0.3$ ,  $P<0.01$ ).

#### Conclusion

In conclusion, a 1600 mg dose of Neuravena® acutely improved attention, concentration and the ability to maintain task focus in older adults with differing levels of cognitive status. Further research is required to determine the mechanism of this effect.

#### Source of funding

Frutarom Switzerland Ltd provided supplements and financial support for this study



## Concurrent Session 13: Cognition and the Brain

### Dietary patterns are associated with cognition among older people with mild cognitive impairment

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#### Background

Increasing age is associated with reduced cognition. There is some evidence that consumption of specific nutrients may be related to cognitive decline. More recently, measures of dietary patterns have been used to investigate the role of whole diets to markers of health.

#### Objective

To determine the association between dietary patterns and cognition in a population of older men and women with mild cognitive impairment.

#### Design

Cross sectional analysis of 249 men and women aged 65-90 yr with mild cognitive impairment, who were recruited to a dietary supplement randomised trial. Dietary patterns were identified using factor analysis from a 119-item, self-completed FFQ. Cognition was assessed with the Cambridge Cognitive Examination (CAMCOG). The relationship between tertiles of dietary factors and cognition was investigated using logistic regression.

#### Outcomes

Ninety-four (38%) women and 155 (62%) men completed the baseline assessment with a mean (SD) age of 73.4 (5.6) yr and CAMCOG total score of 89.8 (5.9). Two main dietary patterns were identified: healthy and unhealthy. There was no significant difference in the CAMCOG total score between women and men so data were combined. There was no association between the healthy dietary pattern and cognition, but there was a significant association between the unhealthy dietary pattern and CAMCOG (adjusting for age, sex, education, smoking, use of antidepressants), such that participants with the highest intake of unhealthy foods were more likely to have reduced cognitive function on the CAMCOG executive function subscale OR=2.6 (95% CI 1.11-6.17). However, there was no association when physical activity was included in the model OR=1.91 (95% CI 0.69-5.27).

#### Conclusion

In a group of older people with impaired cognition, a high intake of unhealthy foods was associated with reduced executive function. Additionally, it appears that increased physical activity could ameliorate the adverse effects of an unhealthy diet on cognition in this group.

**Source of funding:** NHMRC

### Dietary intake of essential nutrients in children with ADHD and associations with behaviour and cognition

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#### Background

Poor nutritional status of several essential nutrients has been associated with attention deficit hyperactivity disorder (ADHD).

#### Objective

The aim of this study was to compare the intake of dietary zinc, magnesium, iron and folate in a sample of Australian children with ADHD against the nutrient reference values (NRVs) and to identify possible relationships between these nutrients and measures of behaviour and cognition.

#### Design

Ninety children with ADHD underwent cognitive assessments and their parents or guardians filled out food frequency questionnaires and Conners' Parent Rating Scales. To compare intake data to the NRVs, data was split into age groups of 7-8 and 9-12 yrs.

#### Outcomes

The majority (74-100%) of children aged 7-8 yr met the recommended dietary intake (RDI) for the nutrients of interest. However, only 30% and 60% of children aged 9-12 yr met the RDIs for dietary folate and magnesium respectively. No correlations were found between the nutrients of interest (zinc, magnesium, folate, iron) and Conners Parent Ratings, DSM-VI criteria or literacy and numeracy. However, intakes of folate ( $r = -0.23$ ,  $P < 0.05$ ) and iron ( $r = -0.2$ ,  $P < 0.05$ ) were negatively associated with measures of attention.

#### Conclusion

Improving diet quality to ensure that RDIs are achieved may increase attentiveness in children with ADHD.

#### Source of funding

Supported by an Australian Research Council Linkage Project grant (LP0776922) in partnership with Novasel Australia and a Department of Health Scholarship for CM.





## Concurrent Session 14: Musculo-skeletal Health

### The role of protein on bone health in older women

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#### Introduction

In the past, excessive protein intake has been thought to be detrimental to bone health by inducing chronic metabolic acidosis which could lead to hypercalciuria and accelerated mineral dissolution. However, recent studies with reliable assessments of whole body calcium retention and biomarkers of bone metabolism do not support this claim. Although epidemiological studies with younger subjects have shown variable results on the effect of protein on bone mass and fracture risk, the majority of studies with elderly subjects have shown that a relatively high protein intake was associated with reduced bone loss and reduced risk of hip fracture. Two protein intervention studies in hip fracture patients have shown positive effects in reducing bone loss. However, there have been no well-designed randomised trials of sufficient duration or power to examine the effects of increased dietary protein intake on bone health in older community dwelling women.

#### Protein intervention study with older Australian women

We conducted a two-year randomised, double blind, placebo controlled trial to evaluate the effect of protein supplementation on bone mineral density (BMD) of older postmenopausal women who were still active in the community. The study subjects were recruited in 2007 using a population-based approach in which a random selection of women (n=6,065) aged 70-80 years on the electoral roll in Western Australia received a letter inviting them to join the study. The exclusion criteria were: previous osteoporotic fracture; currently or within last year taking medication for osteoporosis apart from calcium or vitamin D; metabolic bone disease apart from osteoporosis; high protein intake; difficulty absorbing food. Two hundred and nineteen eligible participants were randomised to either the Protein group or the Placebo group. Participants in the protein group received a 250 ml milk based high protein supplement drink reconstituted with cold water from the protein drink powder, which provided 30 g of protein, 600 mg of calcium and 3.3 kJ/ml. Participants in the placebo group received an isocaloric placebo drink reconstituted with cold water from the placebo drink powder which had the same energy and calcium content but only contained 1.7 g protein per 250 ml. Assessments made at baseline, one and two years include DXA total hip and whole body BMD, anthropometry, 24h urinary calcium excretion and serum insulin-like growth factor I (IGF-I). The study was

registered with the Australian Clinical Trials Registry (Registration number: ACTRN012607000163404).

There were no significant differences between the protein and placebo groups in baseline characteristics. At study entry, the mean age of participants was  $74.3 \pm 2.7$  years and the mean protein intake was  $76.5 \pm 17.8$  g/day (1.15 g/kg body weight/day). Eighteen (9.2%) participants had protein intake below the Australian recommendation of Estimated Average Requirement (EAR, 0.75 g/kg body weight/day) and 56 (28.6%) had protein intake below the Recommended Dietary Intake (RDI, 0.94 g/kg body weight/day) for women aged over 70 years. The average calcium intake at baseline was  $914 \pm 386$  mg/day.

In both the protein and the placebo groups, there was a significant decrease in total hip BMD (Protein  $-11 \pm 3$  mg/cm<sup>2</sup>, Placebo  $-8 \pm 2$  mg/cm<sup>2</sup>) and total body BMD (Protein  $-11 \pm 2$  mg/cm<sup>2</sup>, Placebo  $-5 \pm 2$  mg/cm<sup>2</sup>) over two years. However, there was no between group difference. The 24-hour urinary calcium excretion increased significantly from baseline in both groups at one year but only in the protein group at two years. There was a non-significant trend of the protein group had greater increase in urinary calcium excretion compared to the placebo group at two years ( $13.4 \pm 7.3\%$ ,  $P=0.069$ ). Compared to the placebo group, the protein group has significantly higher serum IGF-I at one ( $7.3 \pm 2.5\%$ ,  $P=0.004$ ) and two years ( $8.0 \pm 3.3\%$ ,  $P=0.016$ ).

#### Conclusion

In these healthy ambulant women with baseline protein intake above the current Estimated Average Requirement of 0.75 g/kg body weight/day, extra protein was not a critical beneficial or deleterious regulator of their bone mass. The negative impact of increased urinary calcium loss could have been offset by the increased serum IGF-I level or increased intestinal calcium absorption as has been shown by others. The effect of increased protein intake on bone health in older people deserves further study in other populations, especially those with low dietary protein intake.

#### Source of funding

Supported by Australian Government National Health and Medical Research Council (Project Grant: 458625). Fonterra Brands Limited provided the whey protein free of charge.



## Concurrent Session 14: Musculo-skeletal Health

### The skeletal response to vitamin D supplementation during sunlight deprivation: a randomised trial in Antarctic expeditioners

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#### Background

Antarctic expeditioners experience prolonged sunlight deprivation that results in vitamin D insufficiency (25(OH)D <50 nmol/L). The deficiency occurs within 4 months unless baseline values are greater than 100 nmol/L.

#### Objective

To test the hypothesis that the maintenance of vitamin D sufficiency (>50 nmol/L) required repeat dosing.

#### Design

We randomly assigned 90 expeditioners (mean age 44 yrs) to vitamin D<sub>3</sub>; 50 000IU at departure, monthly or 2 monthly, for 12 months. Participants provided serum samples at departure, 6 and 12 months for assay of vitamin D (25(OH)D), parathyroid hormone (PTH) and bone markers (Osteocalcin). Differences were assessed using repeated measures ANOVA.

#### Outcomes

At baseline, mean 25(OH)D was 64 ± 2 nmol/L. After 12 months, serum 25(OH)D had decreased by 20 ± 6% (P<0.01) in those receiving a single dose, increased by 26 ± 6%, (P<0.01) in the monthly regimen, and was maintained in the two monthly regimen (2 ± 7%, NS). Bone turnover markers were elevated in all groups at 6 months (21-27%, P<0.05). PTH tended to be higher at 6 months in the single dose group (26 ± 11%, P<0.08).

#### Conclusion

During an expedition, 50 000IU vitamin D every 2 months was sufficient to maintain serum 25(OH)D levels, but a monthly dose is required to improve vitamin D status so should be considered for those with vitamin D insufficiency prior to departure.

#### Source of funding

Austin Hospital Medical Research Foundation, Trans-Antarctic Association.

### Australian trends in milk intake

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#### Background

Milk is consumed by more than 90% of Australians, and is available in a wide range of compositions. There has been a strong trend towards consuming fat reduced milk in the United States and the United Kingdom accompanied by decreasing intake of plain white milk.

#### Objective

To investigate the trend of milk intake in Australia since the availability of fat modified milks became widespread.

#### Design

Australian supermarket sales volume data was used to examine the trend in white milk sales for the period 1985 to 2009. This data is available by individual product allowing categorisation according to type of milk. Australian total population estimates were used to calculate per capita white milk supermarket sales.

#### Outcomes

Per capita volume sales of reduced fat milk have increased by 80% since 1985 and skim milk by 22%, while per capita volume of regular milk has fallen by 40%. Total white milk per capita volume sales have remained steady. Fat modified milk now provides 45% by volume of white milk supermarket sales. Regular milk is mainly (51.7%) sold in 3 litre packaging while fat modified milk is mainly sold in 2 litre packaging (52.6%). Private label milk makes up most of the volume sold of regular milk (70%), while only making up a minority of the volume of fat modified milks (40%). A substantial proportion of the volume of fat modified milk is from 'value added' milks – the fat content is lower than regular milk, but there has been addition of other milk solids or other bioactive ingredients.

#### Conclusion

The volume of milk sold by Australian supermarkets per capita over the past 25 years has remained steady at about 102 litres per year. However, the type of milk has changed over time towards more purchase of fat modified and 'value added' milk.

#### Source of funding

Not applicable.



## Concurrent Session 14: Musculo-skeletal Health

### Dietary patterns and bone health in women

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#### Background

Dietary patterns have been shown to be associated with health but few studies have investigated the impact of specific dietary patterns on bone health.

#### Objective

The aim of this study was to examine the association between dietary patterns and measures of bone health in a sample of Australian women aged 18-65 years (n=527) recruited through the Twin and Sister Bone Research Program at the Royal Melbourne Hospital.

#### Design

Bone mineral density (BMD; g/cm<sup>2</sup>) at the lumbar spine (L2-L4) and total hip was measured from site-specific scans and total body bone mineral content (BMC; g) was measured from a total body scan using dual energy X-ray absorptiometry. Diet was assessed using a 4-day food diary. Dietary patterns were identified using factor analysis. Scores were calculated based on the amount of each food consumed in the pattern and the weightings determined by factor analysis. Participants were categorised into quintiles according to the scores. Analysis was conducted using generalised estimating equation methods and adjusted for clustering associated with twin and sister pairs and covariates.

#### Outcomes

Factor analysis revealed 5 dietary patterns. Pattern 1 (high consumption of refined cereals, soft drinks, hot chips, sausages and processed meat, vegetable oils, beer and takeaway foods and low consumption of other vegetables, vegetable dishes, tea, coffee, fruit, wholegrain breads and breakfast cereals) was significantly inversely associated with total body BMC (adjusted for age, height, energy intake, physical activity, smoking, education and calcium intake). Pattern 4 (high consumption of legumes, seafood, seeds, nuts, wine, rice and rice dishes, other vegetables and vegetable dishes and low consumption of bacon and ham) was directly associated with BMD at both sites and total BMC in adjusted models. The remaining dietary patterns were not consistently associated with BMD or BMC.

#### Conclusion

This study identified specific dietary patterns associated with BMD and total body BMC among women and potential food-based strategies for improving bone health.

#### Source of funding

National Health and Medical Research Council, Victorian Health Promotion Foundation, Helen M Schutt Trust.

### Analysis of vitamin D and 25-hydroxyvitamin D in freshly dissected and stored chicken meat

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#### Background

Published analyses of vitamin D and 25-hydroxyvitamin D [25(OH)D] in meat report concentrations of 0.1 to 0.3 µg/100 g. It is possible that degradation of the vitamin D molecular structure may occur with post-mortem chemical changes in meat and the low values reported may not represent the concentrations in living muscle. It is postulated that because vitamin D is readily isomerised to iso-tachysterol under mild acid conditions, the post-mortem production of lactic acid in meat by anaerobic glycolysis will destroy much of the vitamin D and 25(OH)D and that concentrations in fresh muscle are higher than those in retail meat.

#### Objective

To determine, as a pilot trial, the vitamin D concentration of freshly dissected meat compared to meat treated in a manner similar to that of commercial processing

#### Design

Legs were dissected from broiler chickens immediately after death from barbiturate euthanasia. The muscle from one leg of each chicken was immediately processed for analysis by HPLC while that of the other leg was kept under conventional meat storage conditions for 3 days before analysis.

#### Outcomes

Preliminary trials show that when muscle tissue is immediately saponified or chilled, the concentrations of vitamin D<sub>3</sub> and 25OHD<sub>3</sub> are higher than values present in the literature. Mean concentration for D<sub>3</sub> was found to be 0.87 (0.03 SD) µg/100 g muscle, while mean 25OHD<sub>3</sub> was 0.35 (0.07 SD) µg/100 g muscle. Vitamin D<sub>3</sub>, in particular, was of interest as this is up to three times higher than reported literature values for chicken. These results suggest that if anaerobic glycolysis is inhibited, or slowed, vitamin D may escape acid degradation, since both saponification and chilling appeared to have a protective effect on vitamin D. Published vitamin D values obtained from retail meats may not reflect physiological concentrations present in muscle tissue.

#### Conclusion

Vitamin D concentrations of freshly dissected chicken meat are higher than published values when post mortem anaerobic glycolysis is slowed or inhibited.

#### Source of funding

Not applicable.



## Concurrent Session 14: Musculo-skeletal Health

### Errors in self reported myocardial infarction in calcium intervention studies

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#### Background

Concern has been expressed that calcium supplementation, may increase the risk of myocardial infarction (MI).

#### Objective

To evaluate the risk further an examination of self reported and verified myocardial infarction hospitalisation and mortality data from a 5-year RCT of calcium carbonate was undertaken.

#### Design

The participants were 1,460 postmenopausal women, recruited from the general population and randomised to receive 1,200 mg of calcium carbonate daily or an identical placebo for 5 years. Self reported adverse events were recorded by the patient and entered into a database devised to classify self reported data. These data were then compared to verified hospital admission and mortality data from the Western Australian Data Linkage System (WADLS).

#### Outcomes

During the 5-year RCT 38 individuals self reported MI (21 calcium vs. 17 placebo groups) of which only 68% were verified. Of patients misreporting MI (10) 70% was in the calcium group compared to 30% in the placebo group. In the calcium group 16% of the self reported MI had intestinal disorders as the adjudicated discharge diagnosis versus none in the placebo group. Furthermore, in the calcium group there were approximately twice the hospital admissions for acute abdominal pain compared to the placebo group (29 and 16),  $P = 0.049$ .

#### Conclusion

These data show that calcium supplementation increases the risk of verified acute intestinal disorders. Furthermore it identifies misclassification of myocardial infarction events by patients as the basis for the apparent increase in the risks of myocardial infarction in some studies.

#### Source of funding

This study was supported by research grants from the Healthway Health Promotion Foundation of Western Australia, Sir Charles Gairdner Hospital Research Advisory Committee, and the National Health and Medical Research Council of Australia (Project grants 254627, 303169 and 572604).



## Concurrent Session 15: Food Factors 2

### Sorghum – a sustainable grain crop for sustainable health?

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#### Background

Climate change threatens to reduce yields of some Australian grain crops, leading to increased planting of alternative high temperature and drought resistant crops such as sorghum to guarantee a harvest.

#### Objective

To present the rationale for researching sorghum's potential as a grain food to assist with dietary prevention of chronic disease.

#### Design

A review of the potential benefits of sorghum to human health set in the context of the Australian food system.

#### Outcomes

Although a well established animal feed grain, very little of the Australian sorghum crop is used for food, in contrast to parts of Africa and Asia where sorghum is a traditional staple food. Health authorities recommend around four serves of grain foods per day, including wholegrains, as part of a balanced diet for adults to help reduce chronic disease risk. In times of climate change, sorghum has the potential to fulfill future requirements for wholegrain foods in Australia, where the development of mainstream commercial sorghum foods is only in its infancy.

As well as having a similar nutrient composition to more conventional wholegrains, some varieties of sorghum are also known for having slowly digestible starch and high levels of polyphenolic phytochemicals. These characteristics may reduce energy availability and are therefore considered disadvantageous in animal feed and in human nutrition when food is in short supply. However under conditions of nutritional excess, slow starch digestibility may assist with blood glucose control and lower appetite and polyphenolic phytochemicals may provide antioxidants to reduce oxidative stress. Sorghum therefore has potential, as yet little investigated, for the development of a new generation of wholegrain foods for chronic disease risk reduction.

#### Conclusion

Recently commenced studies by the authors aim to identify optimal sorghum varieties, formulations and processes for manufacture of consumer acceptable sorghum foods with slowly digestible starch and high antioxidant properties. The effects of these foods on chronic disease risk factors will then be evaluated in human dietary studies.

#### Source of funding

Australian Research Council, Sanitarium Health Foods Ltd, George Weston Foods Ltd, Curtin University of Technology, University of Wollongong.

### The effects of lupin-enriched foods on cardiovascular disease risk factors in overweight individuals

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#### Background

Increased protein or fibre intakes may benefit cardiovascular disease risk factors. Lupin flour is a novel food ingredient high in protein and fibre.

#### Objective

To investigate the effects of a lupin-enriched diet, during and following energy restriction, on cardiovascular disease risk factors in overweight individuals.

#### Design

Participants ( $n=131$ ) were recruited to a 12 month parallel-design trial. They were randomly assigned to consume lupin-enriched foods or matching high-carbohydrate control foods. All participants underwent 3 months of weight loss, 1 month weight stabilisation, and 8 months weight maintenance. Cardiovascular disease risk factors including: blood pressure, serum lipids and insulin and glucose concentrations as well as body weight were assessed at baseline, 4 and 12 months.

#### Outcomes

Lupin, relative to control, did not significantly influence (mean difference (95%CI)) weight loss to 4 months (0.1 kg (-1.2,1.4)) and 12 months (-0.6 kg (-2.0,0.8)). Relative to control, the lupin group's twenty-four-hour ambulatory systolic (-1.3 mmHg (-2.4,-0.3),  $P=0.016$ ) and diastolic (-1.0 mmHg (-1.9,-0.2),  $P=0.021$ ) blood pressures were lower at 12 months; and fasting insulin concentrations and HOMA scores were significantly lower at 4 months (-1.2 mU/L (-1.3,-1.1),  $P=0.004$  and -0.6 units (-1.0,-0.19),  $P=0.004$ ) and 12 months (-1.3 mU/L (-1.4,-1.1),  $P<0.001$  and -0.7 units (-1.1,-0.24),  $P=0.002$ ). Total and LDL cholesterol concentrations did not differ between the groups at any time point.

#### Conclusion

A diet higher in protein and fibre derived from lupin-enriched foods appears to provide cardiovascular health benefits independent of weight loss.

#### Source of funding

This study was funded by The Western Australian Government, Department of Industry and Resources.



## Concurrent Session 15: Food Factors 2

### Acute post-prandial effects of a high-fat meal, and cocoa on inflammatory and oxidative markers in healthy subjects

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#### Background

Very few studies have observed both inflammation and oxidative stress concurrently, in humans, in the first few post-prandial hours.

#### Objectives

To concurrently measure changes that occur to oxidative/anti-oxidative and inflammatory markers, in the acute post-prandial phase following 1) a high fat meal, 2) an antioxidant-containing cocoa drink, 3) combination of these meals, or 4) water.

#### Design

A repeated measures crossover design was used. Ten subjects completed all four meals. Blood samples were collected at baseline and at 1, 2 and 3 h post-consumption. Plasma was analysed for oxidative/anti-oxidative markers (MDA, SOD and CAT), inflammatory cytokines (GM-CSF, IL-1 $\beta$ , IL-6, IL-8 and TNF- $\alpha$ ), lipid levels, and insulin. Blood glucose levels (BGL) were determined at each time point.

#### Outcomes

MDA gradually decreased over time following the water, such that after 3 h it was significantly lower than after the cocoa + fatty meal ( $P < 0.05$ ). SOD activity gradually decreased over time following the cocoa drink ( $P < 0.05$ ). At 3 h, SOD was decreased in the fatty meal group compared to the water ( $P < 0.05$ ). After 1 h CAT activity gradually decreased following the water, whereas CAT activity was higher following the cocoa + fatty meal when compared to water at 2 and 3 h post-consumption ( $P < 0.05$ ). The inflammatory cytokines GM-CSF, IL-6 and IL-8 all gradually increased from baseline to 3 h after the cocoa ( $P < 0.05$ ). IL-8 transiently increased after cocoa + fatty meal ( $P < 0.05$ ). Both insulin and BGL increased transiently after cocoa + fatty meal, and the fatty meal, and at the 2 and 3 h collections, BGL and insulin levels were lower than at 1 h, following the cocoa ( $P < 0.05$ ).

#### Conclusion

The inflammatory cytokines GM-CSF, IL-6 and IL-8 gradually increased following cocoa consumption, but this was within normal physiological ranges. Neither the fatty meal nor cocoa had an effect on MDA suggesting there were no acute oxidative effects. Both a fatty meal, cocoa and the combination showed differing modulation of endogenous antioxidants.

**Source of Funding:** Deakin University

### Theobroma cacao supplementation in obese Zucker rats with NASH

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#### Background

Methionine/choline deficiency is a model of non-alcoholic steatohepatitis (NASH) in rats due to liver steatosis and oxidative stress.

#### Objective

To investigate the antioxidant effect of a *T. cacao* (cocoa) supplement to treat or prevent NASH in obese rats fed a high fat methionine choline deficient (MCD) diet.

#### Design

Obese Zucker rats were fed one of 5 diet regimes: 52 days of MCD and 28 days of MCD with cocoa (C1); 52 days of MCD and 56 days of MCD with cocoa (C2); 80 days of MCD with cocoa (C3); 108 days of MCD with cocoa (C4); or MCD diet alone for 52 days (D). Lean (L) and obese (F) Zucker rats on standard chow were used as controls. Histological examination was performed on livers; red blood cell (RBC) and liver GSH was determined; liver mRNA and protein levels of NADPH oxidase 1 (NOX1) were determined.

#### Outcomes

Steatosis was present to a greater extent in obese rats fed the MCD diet (D) compared to obese rats on a standard chow (F), which in turn showed greater steatosis than lean rats on standard chow (L). Animals on the C2 diet displayed less steatosis compared to animals on the D regime. A higher RBC GSH was observed in the D, C1-C4 regimes compared to the F animals. A lower liver GSH was observed in animals on the D, C1 and C4 regimes compared to L animals. mRNA levels of NOX1 was not different between groups, however NOX1 protein levels were lower in cocoa supplementation groups (C1-C4) compared to animals on the L, F and D regimes.

#### Conclusion

The MCD diet is shown to induce steatosis to a greater extent in obese Zucker rats compared to obese Zucker rats on standard chow. Cocoa supplementation for 52 days (C2) produced less steatosis in these animals, which may be due to the antioxidant content of cocoa. The demonstrated post-transcriptional downregulation of NOX1 in the cocoa supplementation groups may reduce NOX1 generated superoxide and subsequent oxidative damage.

#### Source of funding

Not applicable.



## Concurrent Session 15: Food Factors 2

### Cholesterol-lowering effects of B-glucan containing breakfast cereals and snack bars

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#### Background

There is consistent scientific evidence demonstrating a cholesterol-lowering effect of oat  $\beta$ -glucan. Authorities (Europe & USA) recommend 3.0 g/d of  $\beta$ -glucan to achieve this effect. This study therefore aimed to test (1) if a lower intake level (1.5 g/d) can be as effective as 3.0 g/d; and (2) the influence of the food format (oat flakes versus rolled oats) on  $\beta$ -glucan's cholesterol lowering effects.

#### Objective

To investigate the cholesterol-lowering effect of a diet for heart health combined with minimal  $\beta$ -glucan (control), 1.5 g or 3.0 g  $\beta$ -glucan/day.

#### Design

A 6-week randomised controlled trial in 87 mild to moderately hypercholesterolaemic ( $\geq 5$  mmol/l and  $< 7.5$  mmol/l) men and women. Subjects were randomised to one of three diet arms: A fat-modified diet plus (1) Minimal  $\beta$ -glucan (Control - C): 45 g non-oats breakfast cereal + Milo snack bar; (2) Low dose oat  $\beta$ -glucan (Oats Low - OL): 60 g oatmeal crisp flakes (plus raisins) + Milo snack bar (1.5 g  $\beta$ -glucan) or (3) Higher dose oat  $\beta$ -glucan (Oats High - OH): 60 g hot porridge oats + muesli bar (3.0 g  $\beta$ -glucan). All three diets were matched for macronutrient profile (25% E protein; 45% E CHO; 30% fat), according to individual energy requirements for weight maintenance. Changes in total and LDL-cholesterol from baseline were assessed using a linear mixed model and repeated measures ANOVA, adjusted for weight change over time

#### Outcomes

Total cholesterol reduced significantly in all groups (-7.8 (SD=13.8)%, -7.2 (12.4)% and -5.5 (9.3)% in OH, OL and C groups, respectively;  $P=0.728$ ). Similar reductions were found for LDL-C (-8.4 (18.5)%, -8.5 (18.5)% and -5.5 (12.4)% in OH, OL and C groups, respectively;  $P=0.712$ ). In responders only ( $n = 60$ ), post-hoc analyses showed a significantly higher reduction in LDL-C in the  $\beta$ -glucan groups (-18.3 (11.1)%, -18.1 (9.2)% and -11.7 (7.9) %, in OH, OL and C groups, respectively;  $P=0.044$ ).

#### Conclusion

The favourable trend towards higher reductions in total and LDL-cholesterol with the two  $\beta$ -glucan groups is supported by strong mechanistic evidence of oat  $\beta$  glucan on cholesterol levels in the literature. An intake of 1.5 g/d was as effective as an intake of 3 g/d and food format may play a role in this.

#### Source of funding

Cereal Partners Worldwide funded this research.

### Mechanisms of endothelial protection by dietary flavonoids

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#### Background

Epidemiological data suggests that regular dietary intake of plant-derived foods and beverages benefits cardiovascular health by reducing the risk of coronary heart disease and stroke and decreasing the risk of cardiovascular disease. A recent meta-analysis of randomised controlled trials of plant-derived foods on cardiovascular health suggest that flavonoids, a large subgroup of polyphenols derived from tea and cocoa or dark chocolate, may have more cardiovascular protection than other substances.

#### Objective

Our research aims to investigate the activation of AMP-activated protein kinase (AMPK), a key regulator of cellular energy status, in endothelial cells by quercetin and its metabolites, and the subsequent protective effect to oxidative stress or uncoupling of eNOS.

#### Design

To cultured Human Aortic Endothelial Cells (HAECs), we will determine the effect of increasing doses of quercetin and its metabolites on eNOS activation, AMPK phosphorylation, ACC phosphorylation and Hsp90-eNOS coupling via western blot and production of S-nitrosothiols via gas-phase chemiluminescence technique. Induction of HO-1 will be assessed at the mRNA by Quantitative RT-PCR and protein level by western blot.

#### Outcomes

In our preliminary short-term studies, we have found that methyl-quercetin, one of the main metabolites produced in humans, resulted in a significantly higher activation of AMPK, ACC as well as a higher production of S-nitrosothiols by HAECs, when subject to oxidative stress. HO-1 activation increased significantly after 10mins of pre-treatment with methyl-quercetin.

#### Conclusion

It is known that flavonoids contribute to cardiovascular protection in humans. *In vitro* studies have shown that this is due in part to increasing bioavailability of NO and activity of eNOS. However, there is little to no data available on the effects of flavonoids on the endothelium through induction of HO-1 and activation of AMPK pathway, which are important components in the response to vascular injury and endothelial dysfunction. This project will provide insight into the specific effects flavonoids have on the signalling pathways in endothelial cells, and the *in vivo* effects of HO-1 through HO-1<sup>-/-</sup> mice.

#### Source of funding

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## Concurrent Session 16: Lupin: A Sustainable Crop and Healthy Food

Sponsored by Centre for Food and Genomic Medicine

### Harvesting the benefits of lupins

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#### Background

The Australian Sweet Lupin (ASL) was only domesticated as a crop plant for modern agriculture in the later half of the 20<sup>th</sup> century. ASL is a grain legume well suited to dry-land farming systems. By 1999, Western Australia produced to 1.6 million tonnes making up 80% of the world's supply. Since then production has declined as farmers have found the crop less profitable to grow.

The lupin kernel has a unique combination of high protein, high dietary fibre, low oil and virtually no starch. To date the lupin grain has been mostly utilised as an animal feed.

#### Objective

To breed more profitable lupin varieties for farmers and with grain qualities to enable widespread adoption as a healthy food ingredient.

#### Design

The paper describes the development of lupin as a crop plant and a multidisciplinary approach to characterise the nutritional and potential health benefits of the grain.

#### Outcomes

Higher yielding varieties with improved disease resistance have been bred for farmers. Lupin protein is higher in lysine than cereals and is a very high source of arginine. The lipid fraction is high in oleic acid and is a rich source of lecithin, carotenoids and tocopherols. It is low in protease inhibitors, phytates, tannins, saponins and has no detectable lectins.

Lupin kernel flour lowers the glycaemic load in wheat based products such as biscuits, breads, pasta and noodles. Small scale niche products are now commercially available. Protein isolates, fibre fractions have excellent functional properties and specific biological activities.

#### Conclusion

Issues to be resolved include: (i) managing the risk of allergy as lupin can trigger anaphylaxis in a small proportion of the population; (ii) the continuity of supply of lupin ingredients of consistency quality and flavour stability; (iii) promoting the benefits of lupins to the food industry and consumers.

#### Source of funding

The Western Australian government and the Grains Research & Development Corporation

### Effect of lupin supplemented diets on diabetes traits

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#### Background

Type 2 Diabetes is increasing in incidence, driven by increasing obesity in the community. Diets supplemented with lupin flour have been reported to have a beneficial effect on short-term weight loss in humans, and increase feelings of satiety. It is not known how lupins achieve this effect.

#### Objective

Our aim was to validate the effects of lupin diets on weight loss in a mouse model, and to characterise the molecular basis by which lupins may mediate this outcome.

#### Design

Mice of three inbred strains were maintained on lupin or lupin-free diets that were matched for content of protein, energy, fat, carbohydrate and fibre. Body weights of individual mice were determined weekly. Insulin tolerance tests were performed at the end of the experiment. Blood samples were collected for determining levels of leptin. Tissue samples were collected for microarray analyses to determine gene expression differences between mice on different diets.

#### Outcomes

Mice maintained on lupin diets had lower body weights than those on control/normal diets. Lupin diet mice had lower basal insulin and leptin levels, and had lower insulin resistance. No difference was found in hypothalamus gene expression, but there were significant changes in liver gene expression.

#### Conclusion

Lupin supplemented diets lower body weight and improve diabetes-related traits.

#### Source of funding

This work was supported by the Centre for Food and Genomic Medicine, and by a Project Grant from the National Health and Medical Research Council of Australia.





## Concurrent Session 16: Lupin: A Sustainable Crop and Healthy Food

Sponsored by Centre for Food and Genomic Medicine

### Lupin based foods with unique nutritional value

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#### Background and Objectives

Some of the major health challenges in the world, especially in the developed countries, are obesity, diabetes, cardiovascular disease and associated health issues. One of the main causes is the increased consumption of unhealthy foods (junk food) with high levels of sugar, starch and saturated fats along with low levels of dietary fibre and proteins. Despite continuing public programs promoting healthy eating, junk food consumption is increasing due to strong sensory appeal. Development of healthy foods with similar attractions to popular junk foods could be the most effective solution.

On the other side, prevalence of malnutrition is common in many developing countries which is associated with stunted growth and high mortality among children. Development of low cost high protein foods is vital to address the malnutrition problem.

#### Design and Outcomes

Lupin (*Lupinus angustifolius*), a grain legume similar to soybean, is one of the cheapest sources of good quality proteins and dietary fibre. Lupin flour is high in protein (40%) and dietary fibre (28%) and contains medium level of fat (6%). Lupin is also a good source of micro nutrients such as Ca, P, K, Fe and Zn and contains bioactive compounds such as carotenoids, antioxidants and phytoestrogens.

Considering the unique nutritional value of lupin, a range of foods including bread, pasta, instant noodles, biscuits and crisps were developed by incorporating 10-50% lupin. Addition of lupin flour substantially increased the nutritional value by increasing protein and dietary fibre contents while reducing the energy content. Incorporation of 20% lupin in pasta, instant noodles and biscuits resulted in 20-25% increase in protein and 140-150% increase in dietary fibre contents without deteriorating the consumer acceptance. The crisps prepared by using lupin flour had similar attractiveness to potato and other type of chips but contained three times more protein, six times more dietary fibre, six times less starch and half of the fat than traditional potato chips. By consuming 100g of lupin crisps, a person can get around 50% of their Recommended Dietary Intake (RDI) of protein and fibre.

#### Conclusion

A range of lupin incorporated high dietary fibre, high protein food products with good consumer acceptability have been developed. These foods can help reduce the problems of obesity, diabetes and cardiovascular disease in developed nations and protein malnutrition in developing countries.

**Source of funding:** The Western Australian government

### Randomised controlled trials investigating effects of lupin on obesity, diabetes and cardiovascular disease related outcomes

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#### Background

Lupin flour is a novel food ingredient rich in protein (~40%) and dietary fibre (~30%).

#### Objective

To investigate the acute and chronic health effects of consumption of lupin-four-enriched foods.

#### Design

Five randomized controlled trials have recently been performed to investigate effects on obesity, type 2 diabetes and cardiovascular disease related outcomes.

#### Outcomes

**Appetite / obesity:** Adding lupin-four to bread increased fullness, reduced energy intake acutely, and suppressed postprandial blood ghrelin (an appetite-regulating hormone associated with hunger) levels. Longer-term consumption of lupin-enriched foods did not significantly reduce body weight or body fatness in overweight men and women in the settings of an *ad libitum* diet over 4 months, a weight loss diet over 4 months, and a weight-maintenance diet (following weight loss) over 8 months.

**Diabetes / glucose / insulin:** Adding lupin-four to bread attenuated the acute post-meal blood glucose and insulin responses. Addition of lupin to carbohydrate (glucose) resulted in reduced glycaemic response in type 2 diabetic individuals. Long-term regular consumption of lupin-enriched foods during a weight loss diet over 4 months, and a weight-maintenance diet (following weight loss) over 8 months reduced fasting blood insulin levels and HOMA score (a marker of insulin sensitivity) in overweight men and women. **Cardiovascular disease risk factors:** Regular consumption of lupin-enriched foods lowered blood pressure in overweight men and women consuming an *ad libitum* diet over 4 months, and during an 8 month period of weight maintenance (following weight loss). Regular consumption of lupin-enriched foods did not lower blood cholesterol levels in overweight men and women.

#### Conclusion

Adding lupin to food has significant effects on satiety and appetite acutely. In the longer-term, these effects may be largely overridden by environmental / dietary / lifestyle factors that influence energy balance. Our results are consistent with benefits of lupin-enriched foods on glycaemic control, and on insulin sensitivity in the longer-term. Lupin may provide a simple dietary approach to reduce blood pressure and risk of cardiovascular disease.

#### Source of funding

Trials were supported by the State Government of Western Australia, and the Department of Agriculture and Food, WA



## Concurrent Session 16: Lupin: A Sustainable Crop and Healthy Food

Sponsored by Centre for Food and Genomic Medicine

### Using genomic approaches to characterise the lupin seed storage proteins

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#### Background

Lupin kernel flour is high in protein and fibre and has potential health benefits for obesity and diabetes. The major class of seed storage proteins (SSP) in lupins are conglutins.

#### Objective

Our research has been focusing on seed storage protein gene families and underpinning genomic resources for characterising the lupin grain. The focus on SSPs is because these are most likely to be the major constituents of the lupin grain responsible for related dietary benefits as well as potentially contributing to problems associated with allergenicity.

#### Design

We have undertaken genomic approaches to identify all conglutin genes in *Lupinus angustifolius*, or narrow-leaf lupin (NLL).

#### Outcomes

we have constructed two lupin cDNA libraries from an early stage and a late stage of lupin seed development. EST sequencing of the two cDNA libraries have allowed us to uncover a large number of conglutin genes existing in *L. angustifolius*. The conglutins consist of four families (alpha, beta, gamma and delta) and we have identified 11 new family members. EST sequencing and transcriptome analysis during seed development have been conducted and shown specific patterns of expression for the different NLL conglutin gene families. We have helped to identify the major component of lupin-specific allergenicity and through the construction and screening of a lupin BAC library, have identified genomic clones for important NLL conglutins.

#### Conclusion

The information obtained has substantially advanced our molecular understanding of the composition of the lupin grain and may permit us in the longer term to manipulate the SSP protein profile to increase desirable proteins and reduce undesirable proteins in the lupin grain.

#### Source of funding

Supported by the Western Australian State Government through CFGM.



## Concurrent Session 17: There's Something About Pork

Sponsored by Australian Pork Limited and Pork CRC

### Cardiometabolic health benefits of eating fresh lean pork

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#### Background

High protein meat-based diets are commonly promoted for weight loss, supposedly by increasing both satiety and energy expenditure. Pork is the most widely eaten meat in the world and is a substantial source of dietary protein but, despite its frequency of consumption, there is little evidence of weight loss or other potential health benefits associated with its consumption.

#### Objective

To investigate the impact of regularly eating lean fresh Australian pork on cardiometabolic health

#### Design

One hundred and forty three overweight/obese individuals who normally ate less than one fresh pork meal per week (age, 48±12 yrs; body mass index, 31.7±4.7 kg/m<sup>2</sup>) were randomised to a pork diet (PD; n=73) to eat 5 serves/wk for females (750 g/wk) and 7 serves/wk for males (1050 g/wk) or remain on their customary diet (CD; n=72) for 6 months. Body composition and risk factors for cardiovascular disease (fasting blood lipids, glucose and insulin, blood pressure, heart rate, large/small artery elasticity index) were measured at baseline, 3 and 6 months.

#### Outcomes

Volunteers assigned to PD increased their pork intake 10 fold (P<0.001) by substituting pork for other meats, mainly beef and chicken such that there were no differences in the total energy or protein intake. Total energy and macronutrient intakes did not differ between groups. There were no adverse effects on cardiovascular disease risk factors from pork consumption (no significant difference in fasting blood lipids, glucose or insulin, blood pressure, heart rate or large or small artery elasticity index). Importantly, compared with CD, PD improved body composition including reductions in body weight (PD v CD; -0.8±0.3 kg v 0.3±0.4 kg; P=0.001), BMI (-0.3±0.1 v 0.1±0.1; P=0.001), waist circumference (-0.6±0.4 cm v 0.7±0.4 cm; P=0.01), % body fat (-0.4±0.2 v 0.3±0.2%; P=0.002), fat mass (-0.5±0.2 kg v 0.4±0.3 kg; P<0.001) and abdominal fat (-68.7±24.7 g v 18.9±25.4 g; P=0.002); significant improvements were seen after only 3 months.

#### Conclusion

High rates of pork consumption resulted in improvements in body composition without energy restriction or changes in total meat or protein intake and did not adversely affect risk factors for cardiovascular disease.

#### Source of funding

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### Pork, beef and chicken have similar effects on acute satiety and appetite regulatory hormone responses

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#### Background

Evidence regarding the effect of different types of protein on satiety is limited to only a few studies and the results are inconsistent.

#### Objective

The effects of a meat-containing meal of either pork, beef or chicken on acute satiety and appetite regulatory hormones were compared using a within-subjects study design.

#### Design

Thirty fasting non-smoking pre-menopausal women aged 19 - 45 years, BMI range 19.2 - 38.3 kg/m<sup>2</sup> attended a research centre on three test days to consume, in random order, a meat-containing meal matched in energy (kJ) and protein content, palatability and appearance. Outcome measures were: (1) amount of food consumed at a subsequent *ad libitum* buffet lunch meal; (2) amount of food consumed and macronutrient selection for the rest of the day; (3) Visual Analogue Scale (VAS) ratings for hunger and satiety; and (4) hormonal appetite and satiety signals.

#### Outcomes

No difference was found between meat groups for either energy intake or macronutrient profile of food consumed at a buffet lunch or over the rest of the day, indicating no meal effect on later food choice. VAS scores did not differ between test meals. With the exception of a difference in PYY response between pork and chicken meals (higher Area Under Curve for pork; P = 0.027), no significant differences were found for any of the appetite hormone levels investigated (CCK, ghrelin, insulin) after consumption of test meals.

#### Conclusion

In an obesogenic environment where high protein diets are seen to provide opportunity for better satiety and potential weight loss, this study positions pork, beef and chicken as being equal in their acute effect on satiety, insulin secretion and release of appetite-related intestinal hormones. There may be some physiological advantage of choosing pork over chicken, with regard to a higher PYY response, however this requires further investigation over the longer term to expose any subsequent clinical advantage.

#### Source of funding

Source of funding: Pork CRC and Australian Pork Ltd.



## Concurrent Session 17: There's Something About Pork

Sponsored by Australian Pork Limited and Pork CRC

### A randomised control trial in young women on the effect of consuming pork meat or iron supplements on nutritional status and feeling of wellbeing

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#### Background

Limited information is available on the role of pork meat in influencing iron status.

#### Objective

To determine the effect of consuming pork meat as compared to iron supplementation on nutritional status and feeling of wellbeing in young women.

#### Design

Healthy young women were randomly assigned to a control diet (CG), a pork containing diet (PG) or a control diet with iron supplementation (SG) for 12 weeks. Monthly blood collections checked iron status biomarkers. Pre and post study food frequency questionnaires were completed.

#### Outcomes

Sixty-five women aged  $24.6 \pm 4.4$  y (mean  $\pm$  SD) completed the trial. At week 12, haemoglobin concentrations were significantly ( $P < 0.05$ ) higher in PG and SG as compared to individuals in CG. Serum ferritin concentrations were increased significantly ( $P < 0.05$ ) in participants assigned to SG as compared to the other groups. Plasma zinc concentrations at the end of the intervention were similar to baseline concentrations for individuals in the CG and PG but were decreased significantly ( $P < 0.05$ ) in SG. Plasma folate, erythrocyte folate, serum vitamin B6 and serum vitamin B12 concentrations were unaffected by the intervention, although the concentrations of vitamins B6 and B12 tended to increase in PG. Responses to the Health Survey (SF36) showed a significant improvement in the scores for "vitality" (1 of the 8 concepts) in SG as compared to participants assigned to CG or PG. In those consuming pork, the score for "bodily pain" was more favourable than scores in CG and SG. PG showed some improvement in the remaining health concept scores. However, no significant relationships were observed between health concept scores and biomarkers of nutritional status.

#### Conclusion

Consumption of pork meat by young women maintains haemoglobin levels to the same extent as low dose iron supplementation and enhances the feeling of wellbeing.

#### Source of funding

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The authors have no conflict of interest for this research.

### Does hypoxia alter iron storage in pork?

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#### Background

Selection for lean growth of pigs is considered to have resulted in an increase in white, fast twitch type IIb muscle fibres and a decline in muscle iron content. Dietary supplementation of pigs with organic iron to increase muscle iron content has not been successful. High altitude hypoxia (reduced oxygen, O<sub>2</sub>) stimulates erythropoiesis and may affect iron storage in the muscle.

#### Objective

To determine the effect of intermittent hypoxia (simulated high altitude) and dietary iron content on haematopoiesis and iron storage in pork.

#### Design

The effect of diet and hypoxia on iron in pork was examined in a 2 x 2 x 2 factorial design consisting of two O<sub>2</sub> levels (20.9% vs. 13%), two diets (plus vs. minus iron (Fe) supplement) and two genders (boar vs. gilt). Hypoxia was generated in purpose-built chambers using 2 simulated altitude generators per chamber with identical living area as those pens housing normoxic pigs (NP). Hypoxic pigs (HP) were exposed to maximum hypoxia ( $13 \pm 1.4\%$  O<sub>2</sub>) for 9 hours every night for 56 days. Blood was collected at -13 d (baseline), 28 d (midpoint) and 56 d (end point) of the experiment for serum iron and full blood evaluation. Pigs were humanely slaughtered at a commercial abattoir where tissue samples were collected from heart, spleen, liver and 5 skeletal muscles (representing the fore, hind and loin regions of the carcass) for Fe assay using atomic absorption spectrometry.

**Outcomes** Pigs exposed to intermittent hypoxia had significantly ( $P < 0.05$ ) greater (~10%) haematocrit, haemoglobin concentration and erythrocyte count compared to normoxic controls at 28d, however, by 56d, only a trend ( $P = 0.07$ ) was found. This suggests some adaptation to hypoxia had occurred, with all values increasing independently with time. Dietary iron content did not influence these variables. Hypoxia did not affect growth performance or P2 fat depth but there was a tendency ( $P = 0.056$ ) for the carcasses of hypoxic pigs to be ~4 kg lighter. No effect on muscle Fe concentration was found due to hypoxia or dietary Fe content although Fe-supplemented pigs had 2-fold higher ( $P < 0.01$ ) Fe levels in the liver and spleen than those pigs not supplemented.

**Conclusion** Despite experimental perturbation of other components of iron homeostasis such as liver, spleen and blood chemistry, these data confirm that dietary Fe-supplementation and/or hypoxia do not lead to an increase in Fe storage in pork after 56d of exposure.

#### Source of funding

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## Concurrent Session 17: There's Something About Pork

Sponsored by Australian Pork Limited and Pork CRC

### Dietary lecithin may improve meat quality and the health attributes of pork

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#### Background

Tenderness is considered to be the most important quality characteristic of pork. Both chewiness and shear force can contribute to tenderness. Collagen is a major component of intramuscular connective tissue that impacts on tenderness. Lecithin has the potential to inhibit collagen cross-linking and preliminary studies suggest that dietary lecithin supplementation to pigs may reduce chewiness and hardness but not shear force (D'Souza et al. 1995).

#### Objective

To determine if lecithin could reduce collagen synthesis and improves meat tenderness and other pork attributes.

#### Design

A total of 36 female pigs were fed one of four diets containing either 0, 0.4, 2 or 8 % lecithin during the finisher phase. After 6 weeks, the pigs were harvested and muscle samples obtained to determine meat quality and gene expression. Meat colour was determined on fresh loin pork while shear force was determined on cooked loin samples. Pork chewiness and hardness will be determined on cooked pork.

#### Outcomes

Dietary lecithin had no effect on shear force (3.1 vs 3.8, 3.4 and 3.3 kg for 0, 0.4, 2 or 8 % lecithin) but increased dressing percentage (73.6 vs 75.5, 75.5 and 75.5 %,  $P=0.009$ ). Dietary lecithin also made the pork more red and darker as indicated by a linear increase in  $a^*$  (5.1 vs 5.3, 6.0 and 6.2,  $P=0.004$ ) and decrease in  $L^*$  (51.1 vs 49.9, 49.4 and 49.1,  $P=0.048$ ) values respectively. Samples are still being analyzed for compression, fatty acid profile and expression of genes involved in collagen synthesis and degradation.

#### Conclusion

The present data indicate that dietary lecithin can alter meat quality and increase dressing percentage. The cause of the increased dressing percentage is unknown.

#### Reference

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