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## MURIEL BELL MEMORIAL LECTURE 2011

### The politics of nutrition

J REID

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Dr Muriel Bell was a prominent figure in the Health Department in the 1930s and the first appointed “nutrition officer”. She became a household name with her column in the *Listener* and placed key nutritional issues on the agenda of the politicians of the day. It is noteworthy that the issues that she was dealing with are not that different from what is important today: iodine deficiency, rising rates of obesity and poverty. Her lectures to nurses reinforced the importance of eating more fruit and vegetables and less sugar and fat.

Strong advocates for nutrition who can champion the scientific elements of the issues with a lens that recognises the political environment are a rare breed but are critical to the advancement of nutritional issues.

The last 20 years have been a political roller coaster for nutrition. A significant development was the establishment of the National Nutrition Taskforce (1989 – 1991) that was tasked with developing nutrition policy goals and objectives and exploring the potential of a comprehensive food and nutrition policy. It is noteworthy that the terms of reference of the Taskforce recognised that nutrition and food were integrally connected and this was further supported by the choice of members of the body. Membership included expertise from academia, food industry, education, economics, clinical nutrition and policy. The multi-faceted nature of nutrition and the recognition that to make progress in nutrition, all of these key sectors need to be working together, combined with political will, were key to the progress that was made. The political cartoons of the day provide good insight into the debate of the times and nutrition related issues feature – obesity, child poverty and food insecurity and folic acid fortification were all in the press.

Key to the debates were (and are) scientific rigour of research and credible spokespeople to champion the science, interested politicians to take the issues forward, engagement (not necessarily in agreement) across sectors – including food industry, public health and consumers and robust advocate groups. One connection that is also important to the debate is the media. Muriel Bell recognised this and maximised exposure of nutrition through the *Listener*.

As scientists and nutritionists it is essential to form the connections with those that can help move nutrition issues – including the food sector, public health and the media. Evidence based nutrition must take into account the political environment; this is integral to the total evidence based picture. Evidence based nutrition is more than science it is an art of combination.

## **An evaluation of a healthy eating on a budget programme**

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### **ABSTRACT**

**Background:** Based on a “Train the Trainer” model, Cook’n Kiwi was designed to increase the nutrition knowledge and awareness of staff from organisations working with vulnerable and disadvantaged families. The programme, which is offered across the Auckland Metropolitan region, provides healthy eating information that staff can give their clients on both a formal and informal basis. The community focused Cook’n Kiwi programme works alongside the Gardens4Health programme to provide healthy low cost options. Ongoing support is given to staff for up to 6 months after the initial programme to facilitate lifestyle change.

**Objective:** To evaluate nutritional knowledge of and attitude towards a healthy lifestyle

**Design:** A post programme questionnaire was used to evaluate nutritional knowledge and attitudes of participants attending programme presentations during 2011. In total, 20 groups (n = 184) completed the questionnaire.

**Outcomes:** A mean score of 68% was achieved for nutritional knowledge. Scores achieved ranged from 24-90% between groups. Concepts that participants continue to struggle with are the number of recommended servings and serving sizes from the four food groups. Attitudinal trends showed more participants had addressed an increase in exercise (37%) as well as fruit and vegetables (39%) than decreasing fat (21%), sugar (26%) and portion size (17%). Requests are often made for healthy recipes and cooking classes. Participants frequently used the resources provided when visiting clients. However, lack of motivation, skills and financial constraints remained barriers to initiating dietary change. As a result of this, a training package was developed and launched in the April 2011 to increase access to accurate healthy eating information for both staff and clients.

**Conclusion:** The resources provided in the Train the Trainer package allow for more confident and accurate use of healthy eating information that meets the specific needs of the staff and clients of community focused organisations.

### **INTRODUCTION**

Obesity is an ever increasing presence in New Zealand (UoO and MoH, 2011). The increasing cost of food, identified lack of food literacy and lack of education in schools mean that the population are unlikely to be improving in terms of nutritional status. Eating well at home has both health and financial advantages. Accessing the people most in need of the support and motivation to develop a better relationship with the concept of ‘quality home cooking’ can be a problem. One approach is to work through those who are in contact with disadvantaged and vulnerable families every day as part of their work.

Based on a “Train the Trainer” model, Cook’n Kiwi was designed to increase the nutrition knowledge and awareness of staff and volunteers from organisations working with vulnerable and disadvantaged individuals and families. The programme is offered across the Auckland Metropolitan area as a package and provides information on healthy eating on a budget which staff can present to their clients or pass on informally. Staff can also pass on practical tips about healthy eating to their clients, who they might be seeing about entirely different issues. This allows access to families that would otherwise be hard to reach with health promotion activities. The community focused Cook’n Kiwi programme works alongside the Gardens4Health programme to provide healthy low cost options.

The successful ‘Healthy Eating on a Budget’ pilot programme was designed and delivered as part of the Counties Manukau District Health Board’s “Let’s Beat Diabetes” initiative. A full scoping exercise was carried out, and consultation held with Healthy Eating, Healthy Action (HEHA) managers in the different District Health Board’s and other stakeholders. The original pilot programme was modified for Auckland wide use incorporating

the experiences of other similar programmes, local and overseas, into the 'Cook'n Kiwi - Healthy Eating on a Budget' programme. It was then tested in a variety of situations and resources sourced or designed to support it.

The Cook'n Kiwi programme includes food choice, health consequences of poor nutrition, energy balance, food groups, shopping on a budget, cost comparisons, meal planning, food labelling, food and nutrition policy, special nutritional needs, recipes, getting positive messages across effectively, practical demonstrations and opportunities to practice skills with a component of 'train the trainer' included. The programme is designed to be interactive, and encourages hands on activity such as preparation of lunch by participants using simple, cost effective recipes. Barriers are reduced through a no cost, proactive approach, with flexible timing and content dependent on participants' needs. Venues are at or near the organization's premises.

To facilitate lifestyle change, mentoring style support is given to staff for at least 6 months after the initial programme to facilitate lifestyle change. This incorporates further training on motivational interviewing and on the extensive resources available such as; a quarterly newsletter, the distribution of recipe cards and provision of further information or training. A certificate is provided on completion.

The objective of this study is to evaluate nutritional knowledge and attitude towards a healthy lifestyle as well as risk of Diabetes for participants on the Cook'n Kiwi programme.

## METHODS

### Study 1: Nutritional Knowledge

In 2009 a pre and post programme 20 minute questionnaire was used to evaluate nutritional knowledge of participants. In total, 19 groups completed the questionnaire. Participants included mostly staff and occasionally their clients as requested. Booking were made through the website, word of mouth and poster distribution. Results of staff and clients were compared. Knowledge questions remained the same in both questionnaires to measure change once the programme had been completed. Questions included portion size and number of serving, the healthy eating plate; and fat and sugar content in food.

### Study 2: Perceived Diabetes risk, Attitude towards Healthy Eating

A 30 minute post programme questionnaire was used in 2010 and 2011 to assess the perception of participants for their risk for diabetes as well as their attitude towards a healthy lifestyle. In total, 20 groups (n = 184) completed the questionnaire. A four point scale was used to assess perception of Diabetes risk. The Likert scale was used to elicit information on consumer attitudes.

Responses to both questionnaires were converted to percentages and the mean calculated for nutritional knowledge scores.



Figure 1: Programme poster and resources

## RESULTS

The typical respondents were women between the ages of 18 and 65 years old who lived in the Auckland CBD and were responsible for the planning and purchase of household groceries and working for a community organization.

**Study 1: Nutritional Knowledge**

On average there was an increase in the level of nutritional knowledge for participants, pre and post the programme, by 23%. Average knowledge scores for staff and client groups were similar at 25% and 21% respectively. This showed that there was not much difference in the knowledge of the two groups.

Feedback from the questionnaires show that participants find information most useful regarding the following areas: reading food labels, healthy food choices, cooking healthy food, and the following resources: DVD, fat and sugar display and recipe book. The resources have proved to be popular, making the information simple and visual is key to the success of the programme.

**Study 2: Perceived risk for Diabetes**

Before the programme 62% didn't think they were at risk for Diabetes while 38% thought they were. After the programme 35% still thought they were not at risk and 27% had changed their response to thinking they were at risk. Out of the participants who initially thought they were definitely at risk, 16% had that confirmed and for 21% the risk was lower. A total of 43% therefore claimed they were at risk and 56% felt they were not at risk for Diabetes.

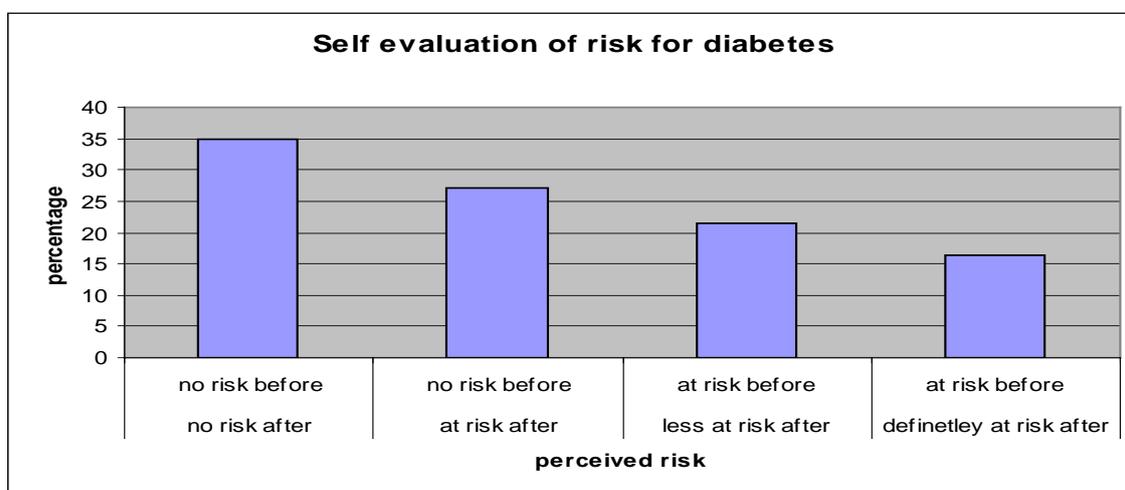


Figure 2: Perceived risk for diabetes pre and post programme

**Study 2: Attitude towards Healthy Eating and Exercise**

More people had addressed increasing exercise (37%), as well as fruit and veg (39%) than decreasing fat (21%) sugar, (26%) and portion size (17%).



Figure 3: Lifestyle change planned post programme

The challenges that participants noted were lack of motivation, the word that they used was they were too “lazy to” change. Lack of economic means was also a challenge affecting vegetable intake. Portion control was something people often remark about, especially men when they see the plastic food examples used to show correct portion size. Portion size is also important as far as budgeting is concerned. The more food eaten, the more it costs. Cookery skills were limited with many people wanting cookery lessons. Many people cannot identify what a healthy recipe is and therefore we are often asked for healthy, low cost recipes. We have cook books on low fat cooking and budget cookbooks but few examples of both concepts in one book.

Because of the amount of information participants found it difficult to remember it all after the presentation. This led to the development of the manual and resource kit made available in April 2011 which includes material staff can use in presenting part or the whole programme.

## **DISCUSSION**

Individuals who provide services to at risk or vulnerable members of the community who may be expected to give advice and support to others do not necessarily have a good basic understanding of nutrition and battle with concepts such as number of servings and portion size. They and the clients they provide advice to may therefore be unaware of what and how much they should be eating (EUFIC, 2011). The Cook'n Kiwi programme has had a positive impact on both nutritional knowledge of and perception towards healthy eating of participants.

Although participants may not have addressed portion size or reduced fat and sugar in the diet prior to attending the Cook'n Kiwi programme, they appear to be more motivated to do so as a result of education in these concepts through the programme. Participants may have been more motivated to increase fruit and vegetables in the diet and increase activity rather than reduce fat and sugar intake. One possibility is that this is due to change in taste. Another is the potential influence of educational drives at the time such as 5+ a day and Push Play. According to Rush et al. (2011a) using simple messages is effective in changing dietary behaviour.

Motivation is also affected by economic means. Feedback received from participants pointed to the significant influence of cost on food choice. Resources which provide alternative options such as reducing meat and increasing vegetables, reducing portion size or recipe modification of home recipes can help to make healthy eating a more cost effective, healthier option. Recipe modification also allows for healthier versions of cultural recipes that have good flavour.

Participants report having made changes at the follow up session, two to three months post programme. However, the extent of the changes made and therefore the long term success of the programme needs to be evaluated. A philosophy of involvement is one people can aspire to. Using motivational interviewing teaches participants to set attainable goals over time. This creates a sense of achievement and progress which is more likely to result in permanent behaviour change as reported by Rush et al. (2011b) and would appear to be supported by our findings.

## **CONCLUSIONS**

The Cook'n Kiwi Train the Trainer programme increased nutritional knowledge and provided for a more positive perception towards a healthy lifestyle. Increased nutritional knowledge supported by practical resource and on-going support makes for more realistic self-assessment and confident delivery of the Cook'n Kiwi Train the Trainer programme to staff and clients of community focused organizations.

## **ACKNOWLEDGEMENTS**

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## **Psychosocial determinants of fruit and vegetable consumption among New Zealand university students: results of focus group interviews**

DP WADSWORTH<sup>1</sup>, H HARTMAN<sup>1,2</sup>, P VAN ASSEMA<sup>2</sup>, SJ PENNY<sup>1</sup>, RA PAGE<sup>1</sup>

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### **ABSTRACT**

**Background:** University students living away from home for the first time are particularly susceptible to making poor dietary choices. This includes low fruit and vegetable consumption, and may have long-term health implications.

**Objectives:** To utilise focus groups in order to identify psychosocial determinants associated with fruit and vegetable consumption among university students in New Zealand.

**Design:** Twenty-nine university students, aged 18-24 years old, studying health-related and non-health-related subjects volunteered for the study. Participants took part in one of six focus group interviews, all of which were transcribed verbatim and analysed by thematic analysis. Participants also completed a 24-hour recall in order to quantify fruit and vegetable intake in the previous day.

**Outcomes:** On average 74% of participants met NZ fruit or vegetable intake guidelines. Factors such as taste and health awareness/knowledge were deemed important for consumption of both fruit and vegetables. Flatmates had the greatest social influence, with sharing groceries and cooking, and providing support being central to intake. Cost and availability were major barriers to consumption, with the main factor for intent to change habit being having the money to do so. Students' suggested interventions included: cooking sessions providing quick, easy and cheap recipes; new and more-varied nutritional information about fruit and vegetables; developing 'made-to-measure' interventions; making students more aware of cheap sources of fruit and vegetables; and increasing campus availability of appealing, inexpensive, single pieces of fruit.

**Conclusion:** University students have a different lifestyle to other young adults and thus have specific determinants for fruit and vegetable consumption. Attitude, self-efficacy and an (un)awareness/misconception of dietary guidelines and health consequences should be considered in interventions using a variety of different media for implementation. Additional research is suggested among other groups of young adults, so that promotional strategies can be specifically targeted.

### **INTRODUCTION**

An intake of at least 400 g of fruit and vegetables per day (or 5+ a day) is proposed to assist in weight control and help prevent non-communicable diseases such as cancer, cardiovascular disease and type 2 diabetes mellitus (WHO, 2005). However, poor dietary choices, including low fruit and vegetable consumption, are common in young adults throughout the world, including New Zealand (UoO and MoH, 2011) and may have long term health implications (Papadaki et al., 2007). University students living away from home for the first time are particularly susceptible to developing unhealthy lifestyle habits, including low fruit and vegetable consumption (Dodd et al., 2010; Keller et al., 2008; Papadaki et al., 2007; Ünüsan, 2004). Consequently, promoting adequate intake of fruit and vegetables in young adults is likely to have maximum preventative potential of developing diseases at a later stage of life (Raitakari et al., 1994). Developing successful strategies to implement such long-term behavioural changes requires recognition of variance in motivation and barriers between specific population groups (Uglen et al., 2008). The present study used focus groups to explore the psychosocial determinants associated with fruit and vegetable consumption among university students in New Zealand, where access to fresh fruit and vegetables should not be a problem.

## METHODS

Six focus groups were conducted for this study, each consisting of 4-5 male and female volunteers aged 18-24 years old. Participants were recruited from Massey University, New Zealand, on the premise of participating in 'food-based focus groups', with no specific link made to fruit and vegetable consumption. Participants were asked to quantify their regular physical activity level, and completed a 24-hour recall of fruit and vegetable consumption. The methods and procedures of this study were approved by the Massey University Human Ethics Committee Wellington (Ethics approval number 05/23).

### *Description of the focus groups and topics*

Four focus groups consisted of a combination of students from health- and non-health related study programs; two further focus groups consisted of only students enrolled in health-related and non-health-related studies, respectively. Focus group interviews took between one-and-a-half and two hours each. All interviews were audio taped and conducted with two researchers; one facilitated and moderated the group discussion, the other made notes and monitored the interview process. Focus groups were structured as per the Attitude-Social influence-self-Efficacy (ASE) model (de Vries et al., 1988). Discussion of influences in eating fruit and vegetables were addressed separately during the interviews, as motivations for consumption could vary among these two different groups of food products (Brug et al., 1995).

### *Data analysis*

Results of the focus group discussions were analysed with QSR NVivo®1.3 (QSR International Pty Ltd, 2001), with fruit and vegetables coded separately. Data was analysed according to ASE model (de Vries et al., 1988).

## RESULTS

Twenty-nine participants were recruited, aged 18-24 years old (mean = 22 years); demographics are shown in Table 1. Most participants were flatting and responsible for their own meals (72%), and a small number still resided with parents (7%). Approximately one-third reported walking for at least 20 minutes a day as their only activity, whilst the remainder of participants reported additional physical activity beyond this. The proportion of participants meeting the recommended consumption levels of fruit or vegetables in the previous 24 hours were similar (Table 2).

*Table 1: Participant Demographics*

	Percentage (%)
<b>Gender</b>	
Males	41
Females	59
<b>Degree Topic</b>	
Health-related study	59
Non-health-related study	41
<b>Ethnicity</b>	
European New Zealander	80
European-Maori	10
Asian	10

Influences most frequently identified with regards to attitude were taste, perceived health benefits and satiety. Flatmates were reported to have a greater influence on vegetable consumption than that of fruit, whilst parents and friends had a similar influence. Feelings of self-efficacy were reported as important determinants, particularly having the time and ability to prepare vegetables. The perception of fruit as a snack was a positive determinant, whereas the feeling of vegetables belonging in a meal, and not as a snack, was viewed almost equally positively and negatively. The importance of convenience was highlighted for both fruit and vegetables.

Factors identified as barriers to fruit and vegetable consumption were cost, availability and seasonal influences. An item being too expensive was recognised as a barrier to

consumption by the majority of participants. Some participants acknowledged that low cost can also encourage consumption especially if they were buying a fruit that they considered a treat. The lack of readily-available fruit and vegetables when flatting was a particular barrier, though this was also highlighted as a barrier outside the home. The perishability of fruit and vegetables was recognised as influencing how readily available they were, and thus their consumption. Seasonal availability, and on-campus availability, cost and quality were also identified by participants as being barriers towards fruit and vegetable consumption.

Participants stated that fruit and vegetable consumption was normal to them as during their childhood such foods were always available, so it had become habit for them to eat fruit and vegetables. Leading on from this, some participants recognised that being responsible for children of their own was an important factor in their intentions to consume more fruit and vegetables in 5 years' time. Other factors behind such intentions were money, health concerns and the element of routine.

The majority of participants were aware of the 5+ a day recommendations (79%, Table 2) but only 29% correctly identified that it was 2 servings of fruit and 3 of vegetables. When asked to quantify their fruit and vegetable intake, most students met the recommendations (Table 2). Although some participants said they did not (always) eat enough fruit and vegetables, others incorrectly thought they were eating sufficient amounts. There appeared to be a correlation between knowledge of the recommended intake and actual consumption.

In providing suggestions about how to promote fruit and vegetable consumption among their age group, participants suggested a targeted campaign using billboards and the internet. They also suggested providing information about sources of cheap fruit and vegetables, cheap/quick/easy recipes, and sufficient reasons why it is good to eat fruit and vegetables, especially in regard to short-term health implications.

*Table 2: Participant awareness of fruit and vegetable recommendations and reported intake.*

<b>Category</b>	<b>Frequency</b>
Knowledge of current recommendations	23
Misconceptions	12
Met recommended fruit intake (2+ a day)	21
Met recommended vegetable intake (3+ a day)	22

## **DISCUSSION**

Similar to other reported research (Brug et al., 1995; Neumark-Sztainer et al., 1999; Ünüsan, 2004), taste was found to be the most important attitudinal determinant of fruit and vegetable consumption by university students in New Zealand. Low self-efficacy was a second important determinant, and suggested factors likely to lead to this were convenience, lack of time, not preparing meals yourself, and lack of skills needed to prepare vegetables; all of which are also supported by other studies (Brug et al. 1995; Ünüsan 2004). Exposure can help people to appreciate taste (Guidetti and Cavazza, 2008), so offering tasting and cooking sessions at the university could provide students with the opportunity to experience different kinds of fruit and vegetables and develop their skills.

Perceived health consequences were important outcome beliefs found in both the current and previous studies (Brug et al., 1995; Ünüsan, 2004; Keller et al., 2008). In the current study, females mostly mentioned their general well-being and males spoke about healthy eating because of their sport-participation. The possible association between fruit and vegetable consumption and prevention of chronic diseases was never mentioned spontaneously, and it is doubtful whether future health benefits are motivating enough for young people to include fruit and vegetables in their diet (Neumark-Sztainer et al. 1999; Uglem et al., 2008). Some students thought that providing information about the cancer-protective effects of fruit and vegetables might have an impact, because of the increased awareness of cancer. Students of health-related studies knew about the beneficial effects of fruit and vegetables, whereas some non-health related students could only mention the general healthiness of fruit and vegetables. Though most participants in the present study were aware of the recommended intakes because of the 5+ a day campaign, there was misconception among some about the exact recommended servings of fruit and vegetables, also reported in previous research (Brug et al., 1995; Uglem et al., 2008; Ünüsan, 2004). It is interesting to note that in this study the proportion of participants consuming the

recommended amounts of fruit or vegetables was substantially higher than has been previously reported in students across the world (Dodd et al., 2010; Keller et al., 2008; Papadaki et al., 2007; Ünüsan, 2004) or in young adults in New Zealand (UoO and MoH, 2011). In the current study, two-thirds of participants were studying health-related topics and perhaps this, coupled with levels of awareness/knowledge, provides an explanation behind the higher consumption reported in this study.

Cost, lack of variety, seasonal influences, and availability were found to be important barriers for eating the right amount of fruit and vegetables in this study, in line with the findings of earlier studies (Brug et al., 1995; Neumark-Sztainer et al., 1999; Ünüsan, 2004). Previous research (Stewart and Tinsley 1995) has shown that cost was not most important for young adults in employment making their food choices, but the current study suggests this is unsurprisingly an important consideration/barrier for young adults studying at university. In line with previous research on Turkish students (Ünüsan, 2004), the poor availability and quality of fruit and vegetables at campus, coupled with high prices, were reasons for low consumption. Alongside attitudinal interventions, it seems to be important to make university students aware of places where cheaper fruit and vegetables are available. Students spend a lot of their time at the university, and colleges/universities have previously been recognised as important settings for dietary intervention in adolescents (Huang et al., 2004).

Influences of others were strongly associated with the intention to eat fruit and vegetables. People of a similar age, particularly flatmates and partners, seemed to have the biggest influence on students, exerting both positive and negative influences by acting as models and providing both social support and pressure. Consistent with other research (Brug et al., 1995; Papadaki et al., 2007), living circumstances were important influences on student's eating patterns, because for many participants the availability of fruit and vegetables in the flat depended on their flatmates (also reported by Uglem et al., 2008), whereas students still living with their parents did not complain about availability. Student's recognition of parents as important influences on fruit and vegetable consumption reflects the findings of other populations, where parents have passed on their norms and given social support; both directly through parental role-modelling and indirectly through the transmission of attitudes or values (Uglem et al., 2008).

In a mass-media society, choosing the appropriate media tool in which to deliver nutritional interventions can greatly enhance its effectiveness and efficiency. Comprehensive studies within America and Europe demonstrated online nutritional interventions and nutritional information booklets to be well received, convenient, easy to disseminate, and associated with sustained dietary change (Alexander et al. 2010; Freisling et al., 2009). Participants in these studies were not solely of university age, yet the findings highlight the potential of media for enhancing fruit and vegetable consumption. In order to make dietary changes, nutritional messages must be seen as personally relevant (Uglem et al., 2008). Therefore as well as mass media promotion, we suggest the development of 'made-to-measure' interventions that are tailored to the individual.

## **CONCLUSIONS**

For successful implementation, determinants including attitude, low self-efficacy, and an (un)awareness of dietary guidelines and health consequences should be considered when developing future interventions aimed at increasing students' fruit and vegetable intake, whilst a variety of media should be used to deliver these. University students have a different lifestyle to many other young adults and thus have factors and individuals influencing their fruit and vegetable intake specific to their population. Additional research is suggested among other groups of young adults, so that future promotional strategies can be specifically targeted. New Zealand is an ever-growing multicultural society, and consequently cultural considerations should be included in any nutrition promotion project based here.

## **ACKNOWLEDGEMENTS**

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## The release of peptides by *in-vitro* digestion of fermented red deer (*Cervus elphus*) and cow (*Bos taurus*) milk.

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### ABSTRACT

Fermentation and digestion release peptides from milk as a source of amino acid and energy, as well as a source of biofunctional peptides that may have health benefits. The aim of this study was to ferment deer and cow milk using three strains of lactic acid bacteria (LAB) and to compare the release of peptides following *in-vitro* digestion of the fermented milks. The three strains were *Lactobacillus belburueckii* subsp *bulguricus*, *Streptococcus salivarius* subs *thermophilus* and *Lactobacillus casi* strain Shirota and the fermentations were carried out at 37 °C for 24 hours. The *in-vitro* digestion was performed in two steps; imitating both the human stomach (Pepsin, pH 2.5) and the duodenum (Corolase PP, pH 7.5). Release of peptides during milk fermentation and subsequent digestion was quantified using OPA (*o-phthaldialdehyde*) assay. Changes in protein and peptide profiles were evaluated using sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) and quantified using ImageJ software. After 24 hours of fermentation, the pH of cow milk dropped below 4.6 and precipitated caseins while pH of deer milk remained above pH 4.6 for all three strains. Deer milk fermentations gave higher peptide production than cow milk fermentations. Following *in-vitro* digestion peptide production was significantly greater in deer milk than cow milk ( $p \leq 0.05$ ). *Lactobacillus casi* strain Shirota showed the highest release of peptides. The main milk proteins were degraded during fermentation of both milks. Lactoferrin (LF) in cow milk was more resistant to fermentation than that in deer milk for all 3 strains. After 24 hours of fermentation by *Lactobacillus casi* strain Shirota, cow LF was still intact while 47% of the deer LF had degraded. Digestibility of  $\beta$ -lactoglobulin,  $\alpha$ -lactalbumin of deer milk and immunoglobulin of cow milk was improved by fermentation. This study showed LAB fermentation prior to *in-vitro* digestion increased the digestibility and release of peptides from both milks. This effect was greater in deer milk than cow milk.

### INTRODUCTION

Fermented milk products, in addition to providing a source of energy and nutrients, are also a source of peptides with biological functions that may improve health when ingested. The health benefits of fermented milk have been recognised and documented (Gobbetti et al., 2006). Over the last decade a great number of peptide sequences with different bioactivities (e.g. immunomodulatory, antihypertensive, antithrombotic, antibacterial, antioxidative and opioid activity) have been identified in various milk proteins (Korhonen, 2009). Many commercial lactic acid bacteria (LAB) based starter cultures are highly proteolytic (Korhonen, 2009). The *in vitro* study by Pihlanto-Leppala et al. (1998) demonstrated that fermentation of milk with starter cultures or enzymes derived from such cultures prior to treatment with digestive enzymes can enhance the release and alter the profile of bioactive peptides produced. Further, Matar et al. (1996) showed that fermentation of milk by LAB has a major impact on the release of novel low molecular mass peptides during *in vitro* digestion. It is likely that peptides are also released during digestion in the gastrointestinal tract. Deer milk has 8.8 % protein which is twice as concentrated as cow milk (Opatha Vithana et al., 2010). This could be a good substrate for proteolytic activity of LAB strains and may produce more peptides which might have biological functions beneficial to health.

The aim of this study was to compare deer and cow milk during fermentation using three LAB cultures in terms of peptide production before and after *in vitro* digestion and compare deer and cow milk protein digestibility. This is the first report studying the fermentation of deer milk followed by *in vitro* digestion and the information from the present work is potentially useful in understanding the release of peptides in animal nutrition and to evaluate the potential use of deer milk products for human use.

## METHODS

Skim deer and cow milk were inoculated with *Lactobacillus delburueckii subsp bulguricus*, *Streptococcus salivarius subs thermophilus* and *Lactobacillus casi* strain Shirota with 2% (vol/vol) inoculums separately and incubated at 37°C for 24 hr (Matar et al., 1996).

*In vitro* digestion was performed for milk and fermented milk in two steps using Pepsin and Corolase PP (mixture of trypsin, chymotrypsin and several amino and carboxypeptidases) according to Eriksen et al. (2008). The release of peptides during milk fermentation and digestion was quantified using OPA (*o-phthaldialdehyde*) assay (Church et al., 1983). The changes in protein and peptide profiles were compared by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) as per Laemmli (1970) and quantified using ImageJ software (rsbweb.nih.gov/ij/).

All experiments were carried out in triplicate and the reported values are mean  $\pm$  standard deviation. The data were subjected to one way analysis of variance (ANOVA), followed by Tukey's test to determine the significant difference between means at  $p < 0.05$  level using Minitab (16 version).

## RESULTS

The pH of deer milk was 6.1 prior to fermentation whereas it was 6.5 for cow milk. After 24 hr of fermentation; the pH of cow milk was significantly lower than deer milk ferments for all three stains of LAB; *Lactobacillus casi* strain Shirota, *Lactobacillus delburueckii subsp bulguricus* and *Streptococcus salivarius subsp thermophilus*. The pH fell below 4.6 for cow milk at which pH the casein is precipitated. Deer milk pH did not fall below 4.6 for any of the three LAB fermentations over 24 hr (Figure 1).

The proteolytic activities of the studied strains (Figure 2) were significantly higher in deer milk than cow milk ( $p < 0.05$ ). The highest production of peptides was generated by *Lactobacillus casi* strain Shirota (Figure 2) after 24 hours of fermentation at 37°C. Once ferments were digested, significantly ( $p < 0.05$ ) higher peptide production was observed in deer milk compared to cow milk for all three strains and the highest production was found in digest of *Lactobacillus casi* strain Shirota fermented deer milk. Fermented milk digests always had slightly higher peptide concentration than raw milk digest. When comparing the three stains in terms of peptide production there was no significant difference after *in vitro* digestion of both deer and cow milk ferment.

The degradation of milk proteins was visualized using SDS gel electrophoresis. The main milk proteins started to degrade during fermentation of both milks (Figure 3). After *in vitro* digestion, most of the milk protein bands completely disappeared and appearance of new lower molecular mass bands on the gel could be seen. Quantification of protein bands using ImageJ software is shown in Table 1.

Lactoferrin (LF) of cow milk was more resistant to degradation during fermentation by all three LAB strains than deer milk (Table 1). After 24 hours of fermentation by *Lactobacillus casi* strain Shirota, LF was still intact (100%) in cow milk while 47% LF remained intact after deer milk fermentation. Digestibility of  $\alpha$ -lactalbumin ( $\alpha$ -la) of deer milk was significantly ( $p < 0.05$ ) improved by *Lactobacillus delburueckii subsp bulguricus* fermentation. About 11% intact  $\alpha$ -la was found after *in vitro* digestion of raw deer milk while *Lactobacillus delburueckii subsp bulguricus* ferment digest only contained 1% intact  $\alpha$ -la (Table 1). The digestibility of  $\beta$ -lactoglobulin in *Streptococcus salivarius subs thermophilus* fermented deer milk was also significantly ( $p < 0.05$ ) greater than  $\beta$ -lactoglobulin in cow milk ferment by the same culture. Deer milk fermented by the other two stains and digested showed slightly higher digestibility than cow ferment digest for  $\beta$ -lactoglobulin ( $\beta$ -lg). There was a significant difference observed in immunoglobulin (IG) of cow milk. Intact protein level was 21% after *in vitro* digestion of raw milk and this decreased to 0 (no intact protein) after *in vitro* digestion of milk fermented by all three strains. These differences in digestibility of milk protein may lead to formation of novel peptides.

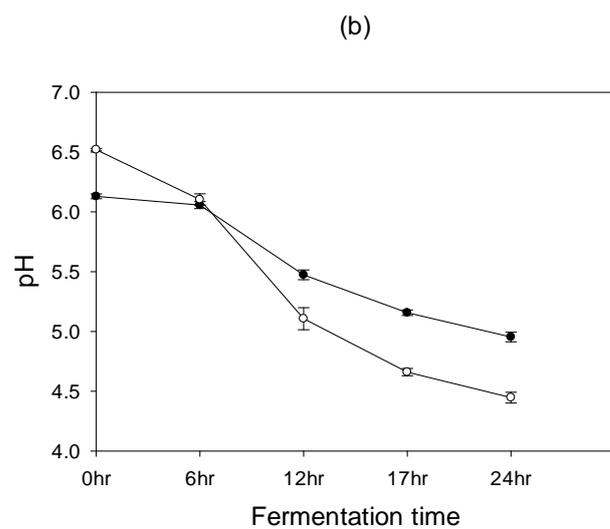
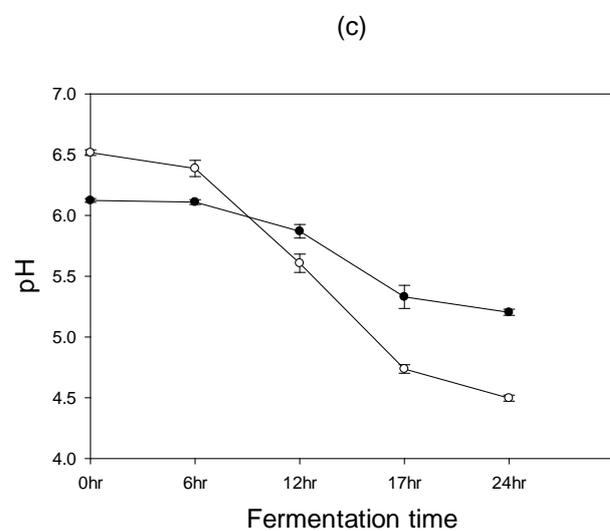
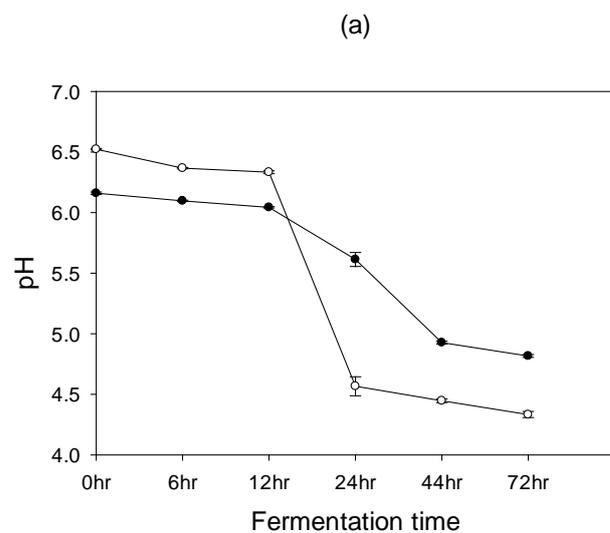


Figure 1: pH during fermentation of deer (—○—) and cow milk (—●—) at 37°C. (a) *Lactobacillus casei* strain Shirota (Lcs), (b) *Lactobacillus delburueckii* subsp *bulguricus* (Ldb), (c) *Streptococcus salivarius* subsp *thermophilus* (Sst). Data are mean of three independent fermentations ± SD

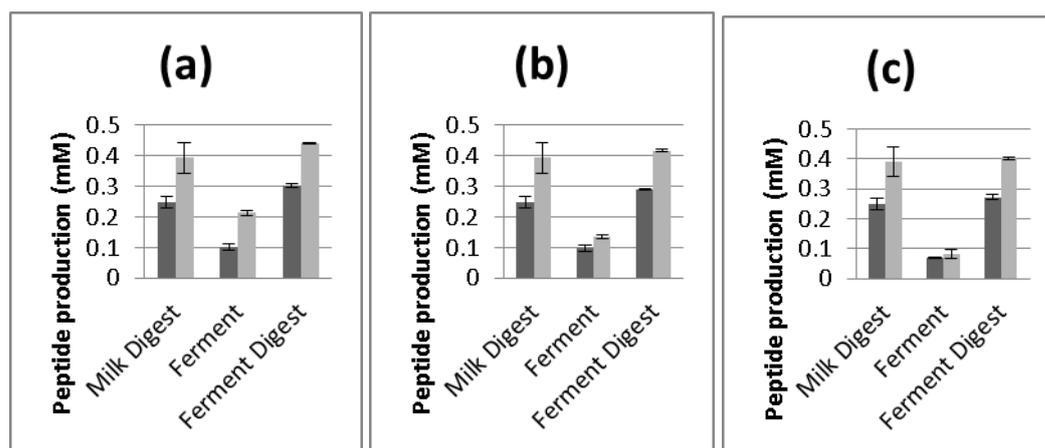


Figure 2: Peptide production following *in vitro* digest of raw milk and milk ferment for 24 hr as well as undigested ferment. Cow (■) and deer (□) milk fermented with: (a) *Lactobacillus casei* strain Shirota; (b) *Lactobacillus delburueckii* subsp bulguricus; and (c) *Streptococcus salivarius* subsp thermophilus. Data are mean ± SD (n = 3).

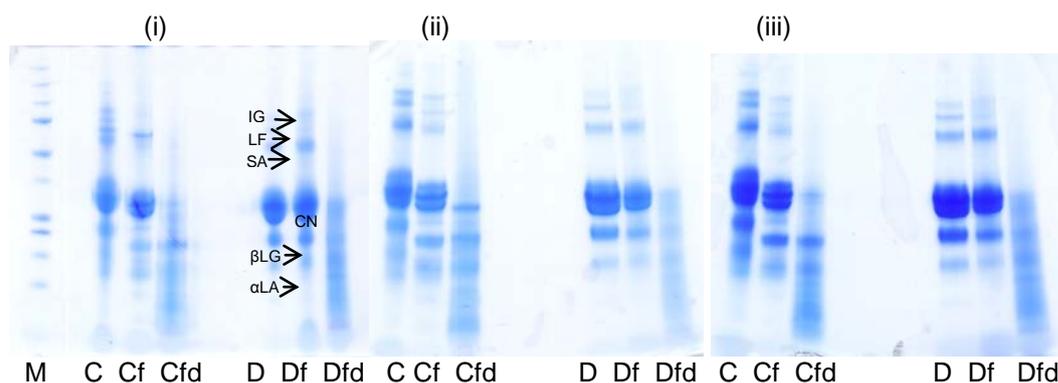


Figure 3: SDS-PAGE (4-12%) of deer and cow milk, ferment and digested ferment using: (i) *Lactobacillus casei* strain Shirota; (ii) *Lactobacillus delburueckii* subsp bulguricus; and (iii) *Streptococcus salivarius* subsp thermophilus. The wells contain: (C) cow milk; (Cf) cow milk ferment; (Cfd) cow milk ferment digest; (D) deer milk; (Df) deer milk ferment; (Dfd) deer milk ferment digest; and (M) molecular weight marker.

*Table 1: Remaining protein content (%) in digest of milk, Lactobacillus casei strain Shirota, Lactobacillus delburueckii subsp bulguricus and Streptococcus salivarius subs thermophilus fermented (24 hr at 37°C) milk before and after in vitro digestion (30 min simulated stomach + 30 min simulated duodenum). Values are obtained by Image J.*

	Lactobacillus casei strain Shirota						Lactobacillus delburueckii subsp bulguricus				Streptococcus salivarius subs thermophilus			
	Milk Digest		Ferment		Ferment Digest		Ferment		Ferment Digest		Ferments		Ferment Digest	
	Deer	Cow	Deer	Cow	Deer	Cow	Deer	Cow	Deer	Cow	Deer	Cow	Deer	Cow
IG	1±2	21±5	62±1	64±2	0±0	0±0	71±4	74±3	0±0	8±1	73±3	77±3	0±0	0±0
LF	0±1	1±1	47±3	101±1	1±1	2±1	42±5	63±4	0±0	2±1	48±2	69±5	0±0	0±0
SA	0±0	1±1	77±1	68±1	0±1	1±0	62±7	63±6	0±0	1±1	70±1	67±6	0±0	0±0
CN	0±0	1±1	83±2	85±1	0±0	0±1	76±8	82±4	1±0	1±1	85±2	84±1	2±0	2±0
β-Ig	54±5	55±8	87±1	86±1	52±1	57±1	85±2	91±7	51±1	57±4	83±4	78±2	51±1	68±1
α-la	11±2	27±5	92±2	90±1	8±4	17±1	87±2	90±3	1±1	13±4	85±3	91±2	15±3	23±4

*Data are mean ± SD (n=3)*

## DISCUSSION

Deer milk was slightly more acidic than cow milk. Differences in pH and buffering capacity of fresh milk reflect compositional variation (McCarthy, 2002). After 24 hour of fermentation, the deer milk ferment showed significantly higher pH ( $p < 0.05$ ) than cow milk which went below the casein iso electric point resulting in precipitation of casein for all three strains (Figure 1). The smaller pH drop of deer milk may be due to several factors such as low bacterial growth resulting in low lactic acid formation or a higher buffering capacity of deer milk than cow milk. The ultimate pH of cheese results from the lactic acid produced (from starter culture lactose metabolism) and moderated by the buffering capacity of the milk (Pandey et al., 2003). Principal buffer components in milk are soluble phosphate, colloidal calcium phosphate, citrate, bicarbonate, casein, salt and a number of minor constituents (McCarthy, 2002). Goat milk has high buffering capacity due to its high content of major buffering components such as minerals and protein (Park, 1992). The mineral and casein content of red deer milk is about twice that of cow milk (Opatha Vithana et al., 2010). This high casein and minerals can lead to high buffering capacity of deer milk and could reduce the pH drop during fermentation when compared to cow milk. Further studies should be done to confirm the nature and the components contributing to the buffering capacity of deer milk.

*Lactobacillus delburueckii subsp bulguricus* and *Lactobacillus casi* strain Shirota fermentation of deer milk produced significantly ( $p < 0.05$ ) higher peptide concentration than the fermentation of cow milk (Figure 2). When all three strains ferments were digested, deer milk ferment produced significantly higher peptide concentration than cow milk ferment. Gobbetti et al. (2000) showed that during milk fermentation, probiotic strains may produce several oligopeptides which generate bioactive peptides only after subsequent digestion by pepsin and trypsin. Peptide bonds that are protected within the native protein structure might be exposed by the action of LAB enzymes, thereby allowing the release of new peptides. According to Korhonen and Pilanto (2006), the use of both fermentation and digestion has been more effective in generating biofunctional peptides than fermentation or digestion alone. Matar et al. (1996) reported that proteolysis during fermentation enhanced the release of peptides and may lead to the formation of novel peptides during gastrointestinal digestion. Overall deer milk digest produce more peptides than cow milk and this was further increased by fermentation prior to digestion.

The ability to produce extracellular proteinases is a very important feature of LAB. They catalyse proteolysis of milk proteins, providing the amino acids essential for growth of LAB (Fira et al., 2001). LAB fermentation in yogurt is a desirable process improving milk digestibility and enhancing nutritional quality by protein degradation and hence, changes the texture, the taste and the aroma of fermented products (El-Ghaish et al., 2011). In this study LAB fermentation improved the digestibility of  $\beta$ -lg of deer milk more than for cow milk (Table 1).  $\beta$ -lg is the main cause of 80% of milk allergies in children and infants.  $\beta$ -lg is the major whey protein in milk and in dairy products and it is of particular interest since it is the only whey protein of cow milk absent in human milk (El-Ghaish et al., 2011). Kleber et al., 2006 studied the ability of some LAB strains to reduce the antigenicity of  $\beta$ -lg using skim milk and sweet whey by indirect competitive ELISA, using polyclonal antibodies. They observed the reduction of antigenicity of skim milk by 90% and sweet whey by 70% compared to the initial value. Our results suggest that less  $\beta$ -lg remained in the ferment digest of deer milk compared to cow ferment digest which may lead to less allergenicity of LAB fermented digest in deer milk than cow milk.  $\alpha$ -Lactalbumin is another whey protein which is less allergenic than  $\beta$ -lg (Wal, 1998). Eleven percent intact  $\alpha$ -la of deer milk was reduced to 1% by digestion with *Lactobacillus delburueckii subsp bulguricus* and to 8% by digestion with *Lactobacillus casi* strain Shirota ferments (Table 1). There was significantly lower concentration of intact  $\alpha$ -la in digest of deer ferment than cow ferment for all three strains. Bu et al. (2010) found that LAB fermentation reduced the antigenicity of cow  $\alpha$ -la and  $\beta$ -lg by hydrolysis. Since our results showed less  $\alpha$ -la and  $\beta$ -lg in deer ferment digest than cow ferment digest, fermented deer milk digest may be less allergenic than fermented cow milk digest. Digestibility of cow IG was significantly improved after fermentation by all tested LAB strains (Table 1). Cow milk IGs are relatively resistant to proteolytic digestive enzymes in the gastrointestinal tract, but can be sensitive to microbial enzymes (Marnila and Korhonen, 2011). In this study lactoferrin (LF) in cow milk was significantly more resistant ( $p < 0.05$ ) to fermentation than that in deer milk for all three strains. Lactoferrin is an 80 kDa iron-binding glycoprotein of the transferrin family that is expressed in most biological fluids and is a major

component of the mammalian innate immune system (Gonzalez-Chavez et al., 2009). In characterising the various peptides generated by LF hydrolysis, it was found that minimal variations in the amino acid sequence change the antimicrobial activity of the peptide. For example, LFampin 268–284 and LFampin 265–284, chemically synthesised fragments from the N-terminal sequence of bLF, differ in only three amino acids (265Asp-Leu-267Ile) but exhibit different antibacterial activities (Kraan van der et al., 2006). Therefore different levels of hydrolysis in deer and cow milk during fermentation may form different peptides which may have different strength of biological activity. Similar to our cow milk fermentation results, results of Franco et al. (2010) indicate that LF structure does not seem to be altered much by the activity of commercial LAB bacteria (yogurt). In contrast digestibility of deer milk is higher than cow milk and was further improved by LAB fermentation prior to digestion.

## CONCLUSION

Fermentation prior to digestion increased the production of peptides and the digestibility of major milk proteins from both deer and cow. Deer ferment digest produced more peptides than cow ferment digest. There was no significant difference in terms of peptide level produced after digestion among three LAB stains used in this experiment. Digestibility of major milk proteins was higher in deer ferment than cow ferment. This study is a novel approach which might have a great impact in diversifying income stream for deer farmers and generate new information which will help in understanding fawn nutrition. In future work deer and cow milk peptides will be fractionated using FPLC and immunomodulatory activity will be studied using human peripheral blood mononuclear cells (PBMC).

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## Oxalate availability in raw and cooked rhubarb

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### ABSTRACT

Total and soluble oxalate content levels, along with gastric and intestinal available oxalates, were measured in raw and cooked rhubarb petioles. Boiling the rhubarb petioles in water or cooking with trim or standard milk significantly ( $p < 0.001$ ) reduced the total oxalate content of the mixture by dilution (raw 902.7 mg/100 g fresh weight (FW) to a mean cooked 454.3 mg/100 g FW). Soluble oxalate levels were also reduced by dilution but cooking with standard and trim milk reduced the soluble oxalate content a further 65.9% and 74.5%, respectively when compared to the soluble oxalate content of the raw petioles. The gastric available and intestinal available oxalates in the raw rhubarb petioles were 739.1 and 420.5 mg/100 g FW, respectively. The total intestinal available oxalate in the raw petioles was reduced by 63.8% to a mean of 152.2 mg/100 g FW for the three cooked treatments. Cooking with trim milk reduced the intestinal soluble oxalate by 77.2% when compared to the level in the raw petioles. In contrast, cooking the rhubarb petioles in standard milk led to a 67.5% reduction of soluble oxalate when compared to the level in the raw petioles.

### INTRODUCTION

Rhubarb (*Rheum rhaponticifolium* L.) belongs to the Polygonaceae family and includes about 60 different species of rhubarb that have been cultivated for thousands of years. Rhubarb originates from China and it is now widely distributed in Europe and Asia. Originally, it was valued for its medicinal properties to treat gastrointestinal dysfunction, ulceration, inflammation, provide renal protection and as an anti-microbial agent (Zargar et al., 2011). The rhubarb petiole is also an important crop in Europe where large amounts are sold fresh and thousands of tonnes are canned annually (Blundstone and Dickinson, 1964). The petiole of cultivated rhubarb can be eaten raw as a source of minerals, vitamins and fibre or cooked as an acid fruit for desserts, as a filler for the jam-making industry in pies, tarts and crumbles (Allison, 1966; Clementi and Misiti, 2010). However, many publications and cookbooks advise that rhubarb should not be consumed regularly as its petioles contain high levels of oxalates (Blundstone and Dickinson, 1964). The total oxalate content in rhubarb petioles was reported to range from 275 to 1336 mg/100 g fresh weight (FW) (Noonan and Savage, 1999). The total and soluble oxalate contents of 10 rhubarb varieties collected from different harvest seasons were investigated by Allison (1966) in which total oxalate ranged from 220-760 mg/100 g FW and soluble oxalate ranged from 42.4-78.2% of the total oxalate. Meanwhile, Blundstone and Dickinson, (1964) found that the soluble oxalate content of four varieties of rhubarb was in a lower range of 83-271 mg/100 g FW and was 56.5 - 67.0% of total oxalate. They also recorded an increase of oxalate content as the plant aged. Much later, Libert and Creed (1985) showed that the total oxalate content of 78 varieties of rhubarb ranged from 3.35 to 9.48% of the dry weight of the petioles. It was concluded that genotype could be the reason for 72% of the variation in oxalate content (Libert and Creed, 1985). Oxalate content in raw rhubarb has been well established; however, the levels of oxalate in cooked rhubarb, the most common way that rhubarb is consumed, are rarely reported. There are only two reports which show the effect of boiling and steaming on oxalate contents of rhubarb petioles. Boiling is the most effective method to reduce soluble oxalate especially if the cooking water is discarded (Savage et al., 2000; Chai and Liebman, 2005). Depending on the cooking method used, the amount of oxalates and the ratio of soluble to total oxalate remaining in cooked food may vary. Since calcium has the capacity to bind to soluble oxalate to form calcium insoluble oxalate, it is thought that the availability of oxalates in food can be changed by adding extra calcium. For instance, adding cow's milk and coconut milk significantly reduced the soluble oxalate content in baked taro leaves while baking alone increased soluble oxalate levels in the food plant (Oscarsson and Savage, 2007; Savage et al., 2009). Similarly, Brogren and Savage (2003) found that baking spinach with products containing high levels of calcium reduced the bioavailability of oxalate compared to spinach consumed alone.

The amount of soluble oxalate, rather than the insoluble form has been shown to be proportional to oxalate absorption (Chai and Liebman, 2004; Holmes and Assimos, 2004). This means that pH changes at different segments of the gastrointestinal tract may affect the form of oxalate present and, therefore, the oxalate available for absorption. In the stomach, where the pH ranges from 1.0-2.5, it is likely that all insoluble oxalate in a plant food can be dissolved. The chyme then passes to the alkaline pH of the small intestine, which ranges from 6.5 to 8. In this region, some of the oxalate solubilised in the stomach will rebind with minerals such as  $\text{Ca}^{2+}$  and  $\text{Mg}^{2+}$  that are also available in the intestine.

This study used an *in vitro* digestion method to simulate the physiological environment of the human digestive tract, this study aimed to investigate whether changes of pH in the digestive system can affect the potential absorption of oxalate from rhubarb in the digestive tract. This study also investigated the effects of cooking and the addition of milk, a good source of calcium, on the composition of the total, soluble and insoluble oxalate content of the cooked mixture. The intention is to provide reliable information on how to minimise oxalate absorption from this widely used food.

## MATERIALS AND METHODS

Rhubarb were grown and harvested from a local garden at the same maturity stage. Petioles of rhubarb were sampled at random from several groups of plants, bulked and frozen at  $-24^{\circ}\text{C}$  for further treatment. Analyses were carried out on raw rhubarb, rhubarb cooked with water, rhubarb cooked with Anchor trim milk (100 ml milk containing 0.5 g fat and 133 mg calcium) and with Anchor standard milk (100 ml milk containing 3.3 g fat and 117 mg calcium) with a 1:1 (w/v) ratio. After boiling, the samples were cooked for a further 5 min to make sure the rhubarb petioles were soft when eaten. The mixtures were analysed by two different extraction methods prior to analysis by HPLC chromatography.

The measurement of total and soluble oxalates was performed following the method outlined by Savage *et al.*, (2000). Total and soluble oxalate contents were determined in triplicate by extracting 5 g of fresh samples with 50 mL 2 M HCl or 50 ml nanopure water, respectively and incubating in a water bath at  $25^{\circ}\text{C}$  for 15 minutes. The extracts were cooled and filtered through 0.45  $\mu\text{m}$  cellulose acetate filter (Sartorius, Göttingen, Germany) prior to HPLC analysis. Insoluble oxalate was calculated as the difference between total oxalate and soluble oxalate contents (Holloway *et al.*, 1989). The measurement of gastric and intestinal available oxalates was performed following the method outlined by Savage and Catherwood (2007) except that the extractions were carried out at  $37^{\circ}\text{C}$ . The gastric available oxalates were extracted with 0.01 M HCl at pH 2 and intestinal available oxalates were extracted with Nanopure water (Barnstead International, Dubuque, USA) at pH 7.0.

Statistical analyses were performed using GenStat version 13 (VSN International Ltd., Hemel Hempstead, UK.) using one way ANOVAs analysis and simple linear regression to assess the effect of cooking treatments and digestive pH on oxalate content and form in the rhubarb petioles.

## RESULTS

Table 1 shows that cooking significantly reduced ( $p < 0.05$ ) the total and soluble oxalate content compared to the original raw rhubarb. The mean reduction of total oxalate in the three cooking trials was 49.7%. The soluble oxalate content was significantly reduced by cooking in water and was further reduced by cooking with either trim milk or standard milk. The lowest concentration of soluble oxalate was measured in the trim milk treatment (117.7 mg/100 g FW), a moderate concentration was obtained in standard milk treatment (158.1 mg/100 g FW) and the highest concentration was observed in water treatment (238.0 mg/100 g FW).

There was a significant effect ( $p < 0.05$ ) of adding different kinds of milk on soluble oxalate content (Table 1). While the total oxalate contents in rhubarb petioles cooked with water, cooked with trim milk and cooked with standard milk were not significantly different, the soluble oxalate contents of the rhubarb petioles in the two milk cooking treatments were significantly lower ( $p < 0.05$ ) relative to boiling in water.

The gastric and intestinal available oxalate contents of the raw rhubarb (739.1 and 420.5 mg/100 g FW, respectively) were lower than the total and soluble oxalate contents measured following the 2M HCL and water extractions, respectively. The gastric available

oxalate was significantly reduced by boiling and by addition of trim and standard milk (means 344.7 mg/100 g FW compared to 739.1 mg/100 g FW for the raw petioles). There were no significant differences between boiling in water or the addition of milk. The intestinal available oxalate content showed a significant reduction ( $p < 0.05$ ) when compared to the level observed in the raw rhubarb petioles. Cooking with trim milk was the most effective treatment to reduce the overall intestinal available oxalate content.

The percentage of soluble oxalate to total oxalate varied depending on the material added to the cooking media. For raw, water, trim milk and standard milk cooking treatments they were respectively, (51.3%, 52.5%; 26.9% and 34.8%). Similarly, for *in vitro* digestive extractions, the percentage of intestinal oxalates to gastric available oxalates followed the same trend as observed for the chemical extraction method. They were 56.9%, 60.9%, 29.6% and 39.9%, respectively.

Linear regression of the total oxalate contents of the four different treatments with the respective gastric oxalate contents gave a significant ( $p < 0.01$ ) linear regression ( $y = 1.1322x + 63.122$ ,  $r^2 = 0.992$ ) while the linear regression of the soluble oxalate contents with the respective intestinal oxalate contents also gave a significance ( $p < 0.01$ ) linear regression ( $y = 1.0639x + 10.66$ ,  $r^2 = 0.998$ ).

## DISCUSSION

The Concise New Zealand Food Composition Tables (2009) place rhubarb in the fruit group even though it is clearly a petiole that is consumed. It is placed in the fruit group in this and many other publications because it is cooked and eaten as a fruit. Rhubarb contains several vital phyto-nutrients such as vitamins, polyphenols, minerals and dietary fibre. It is well known that rhubarb contains high levels of oxalates and this study has shown that the intestinal available oxalates are also very high. The most interesting feature of this study is that a significant reduction in soluble and intestinal available oxalates occurs when rhubarb is cooked with trim milk. This effect occurs because trim milk contains 133 mg Ca/100 ml compared to 117 mg Ca/100 ml in standard milk. Previous studies have shown that addition of calcium especially from milk was an effective way to convert soluble oxalate into insoluble calcium oxalate (Simpson et al., 2009). Trim milk was more effective as it contains 12% more calcium than standard milk but the higher fat content of the standard milk may also have an adverse effect on the binding of soluble oxalate and calcium (Simpson et al., 2009). It has been reported that consuming food containing fat along with oxalate may increase oxalate absorption (Andersson and Jagenburg, 1974; Finch et al., 1981). It is possible that triacylglycerides may bind with calcium in the small intestine to form insoluble soaps. This would reduce the amount of calcium available for binding with soluble oxalate to form insoluble calcium oxalate and, would therefore; leave higher levels of free oxalate available for absorption (Fransson, 1984; Bailly et al., 2000). Therefore, the much higher fat content of the standard milk compared to trim milk (3.3% vs. 0.5%) can also explain the significantly higher ( $p < 0.05$ ) soluble oxalate content of the standard milk cooking relative to the trim milk cooking. Overall, the cooking of rhubarb petioles with trim milk led to a 74.5% reduction on soluble oxalates when compared to the level of soluble oxalate in the raw petioles. In contrast, cooking the rhubarb petioles in standard milk led to a 65.9% reduction of soluble oxalate when compared to the level in the raw petioles.

## CONCLUSIONS

This is the first time the oxalate content of rhubarb cooked with different milk products has been investigated using two different methods to extract the oxalate from the tissue. The present study suggests that oxalate available for absorption in the gastrointestinal tract changes depending on both the pH of the digestive system and materials added to the cooking mix. The *in vitro* extraction method showed that there was less oxalate available for absorption in both the gastric and intestinal regions of the digestive tract compared to the total and soluble oxalates present in raw rhubarb. Although boiling reduced the total and soluble oxalate contents by dilution, the levels of soluble oxalate and intestinal available oxalate were further reduced by the presence of added calcium when the rhubarb was cooked in standard or trim milk.

Table 1: Mean oxalate content of raw and cooked rhubarb compared to gastric and intestine available oxalate contents (mg/100 g FW ± SE)

Treatments	Chemical extraction			<i>In vitro</i> digestive extraction	
	Total oxalate	Soluble oxalate	Insoluble oxalate	Gastric available oxalate	Intestinal available oxalate
Raw rhubarb	902.7 ± 33.5 <sup>a</sup>	463.2 ± 19.7 <sup>a</sup> (51.3) <sup>1</sup>	439.5 ± 31.4 <sup>a</sup>	739.1 ± 28.7 <sup>a</sup>	420.5 ± 10.8 <sup>a</sup> (56.9) <sup>2</sup>
Cooked with water	453.4 ± 4.3 <sup>b</sup>	238.0 ± 7.2 <sup>b</sup> (52.5)	215.4 ± 4.1 <sup>b</sup>	368.4 ± 4.8 <sup>b</sup>	224.2 ± 5.5 <sup>b</sup> (60.9)
Cooked with trim milk	455.2 ± 16.9 <sup>b</sup>	117.7 ± 5.0 <sup>c</sup> (26.9)	337.7 ± 5.6 <sup>c</sup>	323.5 ± 13.1 <sup>b</sup>	95.9 ± 5.2 <sup>c</sup> (29.6)
Cooked with standard milk	454.2 ± 4.4 <sup>b</sup>	158.1 ± 5.9 <sup>d</sup> (34.8)	296.2 ± 8.8 <sup>d</sup>	342.3 ± 10.0 <sup>b</sup>	136.7 ± 4.1 <sup>d</sup> (39.9)

Different letters in each column indicate significant difference ( $p < 0.05$ ).

<sup>1</sup> % soluble to total oxalate.

<sup>2</sup> % intestinal available to gastric available oxalate.

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## Selenium-rich (Seleno) green tea: A possible chemopreventive activity against breast cancer in rats

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### ABSTRACT

**Background:** Green tea has gained a good reputation of being a healthy drink and provides various health benefits including protective effects against cancer.

**Objective:** To assess the chemopreventive potential of selenium-containing green tea (Se-GTE) and regular green tea (R-GTE) against breast cancer induced by 7, 12-dimethylbenz(a)anthracene (DMBA) in female Sprague Dawley rats.

**Design:** Forty, 50-day old, female rats were divided into 4 equal groups. Groups 1 to 3 were orally gavaged with a single dose of DMBA (50 mg/kg body weight) while the fourth group was gavaged with a similar dose of the vehicle (corn oil). One day after DMBA gavaging, the rats in the first group were gavaged with 10 ml of water extract (1% extract) prepared from Se-GTE 3 times weekly for 9 weeks while the rats in the second group were gavaged with a similar dose of R-GTE. The rats in the third and the fourth groups were gavaged with the same volume of water to serve as control groups.

**Outcomes:** Nine weeks after giving DMBA, mammary tumours were found in 8 (80%) and 4 (40%) of rats gavaged orally with water and 10 ml/kg of water extract from R-GTE, respectively. Rats gavaged with R-GTE had significantly fewer ( $p < 0.05$ ) tumours than control group gavaged with water. In contrast, rats gavaged with water extracts from Se-GTE had no mammary tumours. These results are the first to show that oral administration with Se-GTE extract after DMBA-dosing may provide protection against chemical-induced mammary cancer. Thus, Se-GTE may be a useful dietary ingredient with a possible anticarcinogenic capacity.

**Conclusion:** The superiority of Se-GTE over R-GTE may be due to the synergistic effect between polyphenolic compounds and organic selenium which are both potent antioxidants and have antiangiogenic capacities.

### INTRODUCTION

Breast cancer is the most commonly diagnosed malignancy in women worldwide, and exploration of a chemopreventive approach to this cancer is significant as it can potentially identify a dietary ingredient active in the prevention of this disorder. Current treatments including chemotherapy, radiotherapy and surgery are generally associated with a high risk of complications and uncertainty, highlighting the need of new and better therapeutic strategies.

Green tea and selenium have attracted much attention, recently, in both scientific and the public arena because of their pronounced health benefits towards a variety of disorders ranging from obesity to cancer (Clark et al., 1996; Whanger, 2004; Zaveri, 2006; Sagara et al., 2010).

Many studies have shown that both green tea and selenium have anti-angiogenic activities highlighting the potential for the consumption of green tea, especially with a good level of organic selenium as a remedy for various disorders including breast cancer. Although both green tea or its polyphenols and selenium have been reported to exhibit cancer chemopreventive properties against different types of cancers, the tea we are using is unique in having both polyphenols and organic selenium (i.e. the power of two). The synergism between green tea polyphenols and selenium may explain the superiority of Se-GTE over R-GTE in our previous studies regarding the free-radical scavenging, and prebiotic activities (Molan et al., 2009, 2010).

Barros et al. (2004) stated that the experimental animal model of chemical mammary induced carcinogenesis using DMBA closely mimics human breast cancer and can be used to

elucidate the role of biomodulation in mammary carcinogenesis. The purpose of this study was to investigate the chemopreventive effects of physiological concentrations of normal and selenium-containing green teas against DMBA-induced mammary carcinogenesis in female Sprague Dawley (SD) rats

## METHODS

Se-GTE and R-GTE were purchased from the local markets. 7, 12-dimethylbenz[a]anthracene (DMBA) and other reagents were purchased from Sigma Chemical Co. (Australia).

A fresh solution of 1% (w/v) was prepared every other day by dissolving the green tea powder in the proper volume of hot water (100 °C) and allowed to brew for 10 minutes with stirring, set to cool to room temperature and then served to the rats. The tea extracts were supplemented via oral gavage and the dose was based on the assumption that an adult person (75 kg) consumes 3 cups of green tea (750 ml/day) daily which means 10 ml of tea extract per kg body weight (Molan et al., 2010).

Forty female 50-day-old SD rats were used in this study. The rats were hosted two per cage, in a room with a temperature of  $22 \pm 1$  °C and a 12-hr light: dark cycle (light on at midnight) and they had free access to water throughout the study. The animal protocol followed the procedures set by the Animal Ethics Committee of the Massey University (Palmerston North, New Zealand).

After 7 days of acclimation, 30 rats were fasted overnight then orally gavaged with a single dose of 50 mg/kg of DMBA which was dissolved in corn oil. Rats were further restrained from food for 6 hr before being return to normal feeding. It has been found that maximum binding of DMBA to mammary cell DNA takes place between 24 to 48 hr after DMBA dosing. One day after receiving DMBA, rats were randomized into 3 equal groups and gavaged orally either with 10 ml/kg of water (control group) or with the same volume of 1% freshly prepared extracts from R-GTE and Se-GTE (experimental groups), 3 times weekly (Monday, Wednesday and Friday) for 10 weeks. The experiment was terminated at the end of the 9<sup>th</sup> week after DMBA treatment. Water and food (pelleted chow rodent diet) were supplied *ad libitum*. A separate group of 10 rats only received the vehicle for DMBA (corn oil; 4 ml/kg) and used as a negative control.

Food consumption and weight gain were monitored weekly throughout the experiment, in addition to monitoring general health status for signs and symptoms of toxicity. Four weeks after DMBA gavaging, the animals were palpated twice weekly to detect the presence and location of mammary tumours. The time of appearance of the first tumour (latency period) and the relative size and location of every tumour were recorded. The number of rats with tumours (incidence) and the number of tumours/rat (tumour burden) were recorded on a weekly basis and at the end of the experiment. All animals were sacrificed using CO<sub>2</sub> euthanasia. The weight of each tumour was recorded and all tumours from each animal were removed and fixed in 10% buffered formalin.

Data were subjected to ANOVA followed by a Student's *t*-test when significant differences existed among groups. The number, size and weight of tumours (at autopsy) were analysed by a Mann-Whitney U-test, and *p*-values <0.05 were considered significant.

## RESULTS

Average body weights of animals in the four treatment groups were similar throughout the experiment (Table 1). Administration of corn oil, R-GTE or Se-GTE did not produce any gross changes in the liver, kidneys, stomach, or intestinal tract.

### **Effects of tea extracts on mammary tumours**

The chemopreventive effects of the water extracts from R-GTE and Se-GTE were investigated using female rats with mammary cancer, induced by a single oral dose of 50 mg DMBA/kg. The chemopreventive effects of R-GTE and Se-GTE on mammary tumour development are summarised in Table 2. Under the conditions of the present study, Se-GTE produced 100% inhibition of the mammary tumours as no tumours have been found in any of the Se-GTE- treated rats. Similarly, no tumours have been detected in the corn oil-treated group. In the R-GTE group, only 40% of rats developed tumours, and the weights of tumours were significantly smaller (mean weight,  $2.55 \pm 0.29$  g; *p*<0.01) than those recovered from the

control group gavaged with water ( $4.93 \pm 0.52$  g). In contrast, 80% of control animals developed tumours. The administration of R-GTE also prolonged the latency period of tumour induction. In animals receiving the control diet only, the first DMBA-induced tumour appeared at week 5 after the initial DMBA treatment while in the R-GTE-treated group the first tumour was observed at week 7 after DMBA administration.

Research to determine the chemical identity of the organic selenium present in the Se-GTE and the histopathological nature of mammary tumours is underway.

*Table 1: Effects of regular (R-GTE) and selenium-containing (Se-GTE) green teas on body weights of female Sprague Dawley rats.*

Treatment group <sup>a</sup>	Initial weight (g) <sup>b,c</sup>	Final weight (g) <sup>b,c</sup>
Control	125.51 $\pm$ 3.12	295.73 $\pm$ 11.1
R-GTE	122.6 $\pm$ 5.9	289.4 $\pm$ 8.8
Se-GTE	121.03 $\pm$ 2.94	274.6 $\pm$ 11.1
Corn oil	120.4 $\pm$ 3.8	275.4 $\pm$ 6.9

<sup>a</sup>Rats were orally gavaged with 10 ml of either water (control and corn oil groups), R-GTE or Se-GTE tea extract/kg body weight.

<sup>b</sup>Mean weight  $\pm$  SEM.

<sup>c</sup>Differences among treatment means were not statistically significant.

*Table 2: Effect of regular green tea (R-GTE) and selenium-containing green tea (Se-GTE) on the incidence and development of DMBA-induced rat mammary tumours.*

Groups <sup>a</sup>	% incidence <sup>b</sup>	Latency (weeks)	Tumour burden <sup>c</sup>	Tumour weight (g) <sup>d</sup>
Control	80%	5	1.4 $\pm$ 0.34	4.93 $\pm$ 0.52
R-GTE	40%	7	0.4 $\pm$ 0.16*	2.55 $\pm$ 0.29*
Se-GTE	0	-	0	-
Corn oil	0	-	0	-

<sup>a</sup>Rats were orally gavaged with 10 ml of either water (control and corn oil groups), R-GTE or Se-GTE tea extracts/kg body weight.

<sup>b</sup>Cancer incidence is the frequency of rats that develop breast tumours.

<sup>c</sup>Mean number of tumours per rat  $\pm$  SEM.

<sup>d</sup>Mean tumour weight (g)  $\pm$  SEM.

\*Significantly different from control group ( $p < 0.05$ ).

## DISCUSSION

The major aim of this investigation was to evaluate the chemopreventive effects of the R-GTE and Se-GTE against the development of chemically induced breast cancer in female rats. Our results showed for the first time that oral administration of tea extracts containing organic selenium provided 100% prevention against chemical-induced mammary cancer. In contrast, R-GTE exhibited moderate chemopreventive capacity. These results support earlier findings suggesting that green tea or its catechins may have chemopreventive value against breast cancer. It is also important to note that antitumour effects of green tea or its polyphenols have been observed against other types of malignancies (Stuart et al., 2006; Sagara et al., 2010). Moreover, studies on animal models of cancer have suggested a strong inverse correlation between selenium intake and cancer incidence (Clark et al., 1996; Whanger, 2004). The possibility, therefore, exists that green tea especially Se-GTE may have value as a general chemopreventive agent against a wide spectrum of malignancies.

The exact mechanism of action by which Se-GTE inhibits mammary carcinogenesis remains to be clarified. Our hypothesis is that inhibition of angiogenesis by green tea polyphenols and selenium is one of the main mechanisms of inhibiting the establishment of mammary tumours in rats. Inhibitory effects of green tea catechins and selenium on angiogenesis have been demonstrated in various models and suggest that green tea and/or selenium could suppress cancer (Cao and Cao, 1999; Matsubara and Mizushima, 2009).

Both green tea and selenium are potent scavengers of free radicals (Molan et al., 2009) and this may prevent the mutagenesis caused by creation of free radical molecules. Thus, Se-GTE could modulate critical steps in the initiation and promotion of mammary carcinogenesis. It has been noted that patients with breast cancer are under oxidative stress which is indicated by the elevated levels of reactive oxygen species and decreased total antioxidant capacities in the blood (Tas et al., 2005). Moreover, Se-GTE contains higher concentrations of phosphorus, potassium, iron, zinc and copper than R-GTE, but CGT contains more calcium and manganese (Molan et al., 2009).

The superiority of Se-GTE over R-GTE may be due to the synergistic effect between catechins in green tea and selenium which are both potent antioxidants (Molan et al., 2009) and have antiangiogenic capacities (Cao and Cao, 1999; Matsubara and Mizushima, 2009). Minerals, other than selenium (Molan et al., 2009), may also be effective against breast cancer. Recently, Sagara et al. (2010) investigated the effects of green tea polyphenol (GTP) on bladder tumour size and angiogenesis in mice and the results showed that GTP inhibited tumour growth and invasion in mice with established bladder cancer, at least in part through the regulation of angiogenesis.

One of the key advantages of green tea is its non-toxicity and has been consumed in Asian cultures for long time and has not been associated with any significant adverse effects. Recently, Boehm et al. (2009) reported that consumption of no more than 1200 ml (3-5 cups) of green tea per day, which provides around 250 mg of mixed catechins, could indeed be considered safe.

## CONCLUSIONS

Administration of R-GTE suppressed the incidence, multiplicity, and weight of breast tumours induced by DMBA in female rats while administration of Se-GTE resulted in 100% inhibition of tumours. The results suggest that Se-GTE may be an effective chemoprevention agent against breast cancer.

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## Antioxidant capacity of cooked spinach prepared in India styles

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### ABSTRACT

Dietary antioxidants play an important role in reducing the oxidative damage in humans due to their free radical scavenging capacity and thus may help to reduce the incidence of disease. Among dietary leafy vegetables, spinach is widely consumed and contains considerable antioxidant capacity. Here the oxygen radical absorption capacity (ORAC) assay was used to investigate the effects of Indian cooking methods and addition of ingredients such as spices and paneer (Indian milk product) on the antioxidant capacity of spinach. Four different dishes namely palak, plain palak saak, palak saak with ginger and palak paneer were produced. 'Plain palak saak' contained the highest antioxidant capacity ( $163 \pm 39 \mu\text{M TE/g DM}$ ) compared to the other raw samples, while 'palak paneer', which was prepared using paneer, contained significantly lower antioxidant capacity ( $49 \pm 1 \mu\text{M TE/g DM}$ ). This reduction in the antioxidant capacity was completely explained by the dilution of dry matter by paneer. The effect of cooking time on the antioxidant capacity of the Indian spinach dishes was not significant.

### INTRODUCTION

Studies on green leafy vegetables have shown protective effects including anti-diabetic, anti-histaminic, anti-carcinogenic and anti-bacterial activities. These beneficial effects are attributed to bioactive phytochemicals including antioxidants such as flavonoids, flavones, isoflavones, lignans, catechins and isocatechins (Murcia et al., 2009; Subhasree et al., 2009). Since these vegetables are mostly cooked before eating and databases tend to report information for raw food it is important to look at the effects of cooking on these activities.

Spinach (*Spinacia oleracea*) is a commonly consumed plant with a high nutritional value that is rich in antioxidants. Grossman et al. (1994) and Bergman et al. (2001) reported the presence of powerful water soluble antioxidants in spinach leaves and identified a few of them, such as isomers and derivatives of *p*-coumaric acid.

In India some famous dishes of spinach are palak paneer, palak kofta, palak paratha, Kashmiri palak, palak alu and palak curry. The way these dishes are prepared varies from European cooking techniques (steaming, boiling or frying). In India, vegetables are cooked in a wok with the addition of cooking oil, spices and sometimes milk products. The cooking water is not discarded but is evaporated from the dish. The effect of Indian cooking on the nutritional properties of spinach is unknown.

A limited number of papers (Gayathri et al., 2004; Wachtel-Galor et al., 2008; Thomas et al., 2010) have reported the effect of cooking on the antioxidant activities of vegetables and spinach in particular. Wachtel-Galor et al., (2008) reported that boiling and microwaving can reduce the antioxidant activity of vegetables (e.g. cabbage, broccoli and cauliflower) by 39-70% when analysed by a ferric reducing antioxidant power assay. Zhang and Hamazu (2004) found that antioxidants in broccoli flowers and stems were decreased by thermal processing using conventional methods and microwave heating. Ismail et al., (2004) also found that thermal treatment decreased the total phenolic contents in vegetables such as spinach, cabbage and kale. There are a few reports (Sánchez-González et al., 2005; Dupas et al., 2006; Ryan and Petit, 2010) demonstrating that adding milk to tea and coffee reduced the antioxidant activity of these beverages, but there appears to be no studies on the effect of spices, milk or milk products on the antioxidant capacity of cooked vegetables.

In this study, the effects of Indian-style wok cooking on the antioxidant capacity of four spinach dishes, palak, plain palak saak, palak saak with ginger and palak paneer were compared after 15 and 25 minutes cooking. These dishes may be eaten either after 15 minutes cooking as boiled saak or after 25 minutes cooking as saak bhaji. Therefore, the objectives of this study were to analyse the effects of adding spices and paneer (Indian cottage cheese type product) on the antioxidant content of cooked spinach dishes by means of ORAC assay and to analyse the effects of Indian cooking methods on antioxidant capacity of spinach.

## MATERIALS AND METHODS

Fully grown spinach (*Spinacia oleracea*) was purchased from a local grower in New Zealand. Dried spices (turmeric, cumin, Malabathrum leaf, cinnamon, caraway and ginger) and paneer, a typical Indian cheese product made from milk powder and citric acid, were produced by MV Exports, Mumbai, Maharashtra, India and purchased from an Indian supermarket in Christchurch. Ionised table salt (Cerebos-Skellerup Ltd, Auckland) and vegetable oil (Sunfield Canola Salad Oil, Tasti Products Ltd, Auckland) were purchased from a local supermarket.

Table 1: Compositions of the Indian spinach dishes

Palak	Plain palak saak	Palak saak with ginger	Palak paneer
500 g Spinach	500 g Spinach	500 g Spinach	500 g Spinach,
	0.8 g Cumin seeds	0.8 g Cumin seeds	1.2 g Turmeric powder
	1.2 g Turmeric powder	1.2 g Turmeric powder	0.8 g Cinnamon
	1.1 g Malabathrum leaf	1 g Ginger powder	0.8 g Caraway seeds
	1.4 g Salt	1.4 g Salt	1.1 g Malabathrum leaf
	8 mL Oil	8 mL Oil	200 g Paneer
			1.4 g Salt
			8 mL Oil

Four Indian spinach-based dishes, namely, palak, plain palak saak, palak saak with ginger and palak paneer were prepared (Table 1). The wok was preheated to 100°C and then 8 mL of vegetable oil was added followed by spices. After 30 seconds chopped spinach and salt were added. Then, the wok was covered and left to boil. After 15 minutes, the soft spinach was uncovered and stirred for another 10 minutes to allow the moisture to evaporate. Each dish was prepared three times. Palak (the control dish), was prepared using the same procedure but without any spices, salt or oil. Samples were collected from four palak dishes at three different stages, before cooking (i.e. raw), after 15 minutes and 25 minutes of cooking. Samples were freeze dried in a Cuddon freeze dryer (WG Cuddon Ltd., Blenheim, Marlborough, NZ) and then finely ground in a coffee mill (Sunbeam, model EM0400, China).

The dry matter (DM) of each sub-sample of spinach and paneer were determined by drying in an oven (Watvic, Watson Victor Ltd., NZ) at 105°C to a constant weight (AOAC, 2002).

For ORAC assay, triplicate samples of powdered spinach dishes (0.5-1.0 g) were extracted with acetone/water/acetic acid (10 mL, 70:29.5:0.5) (Isabelle et al., 2010). The ORAC-fluorescein assay was performed as described by Boivin et al. (2009). Data were analysed using MARS software and trolox standards to construct calibration curves. ORAC values were expressed as  $\mu\text{mol}$  trolox equivalents (TE) per gram DM.

Statistical analysis was performed using two-way ANOVA using Minitab version 15.1 (Minitab Ltd., Coventry, UK).

## RESULTS AND DISCUSSION

The antioxidant capacities are presented in Table 2. Among the raw samples, plain palak saak contains the highest antioxidant capacity (163  $\mu\text{mol}$  TE/g DM) in comparison to the other samples. In contrast, palak paneer, which was prepared using milk product paneer, contained significantly lower antioxidant capacity (49  $\mu\text{mol}$  TE/g DM). The combination of ingredients had a significant effect ( $p < 0.001$ ) on the antioxidant capacity of spinach dishes. Cooking time had no significant effect ( $p > 0.05$ ) on antioxidant capacity. In the present study, the raw spinach showed antioxidant capacity with ORAC values of 149  $\mu\text{mol}$  TE/g DM. This fell within the range of 103-243  $\mu\text{mol}$  TE/g DM reported for spinach by Ou et al. (2002), but was lower than the ORAC value of fresh spinach reported by Song et al. (2010) and Zhou and

Yu, (2006). The value was greater than the 129  $\mu\text{mol TE/g DM}$  previously observed by Cao et al., (1996). All the samples in the present research were freeze dried before measuring the level of antioxidant capacity. This may have resulted in a reduction in antioxidant capacity.

Table 2: Antioxidant capacity ( $\mu\text{mol TE/g DM} \pm \text{SE}$ ) as measured by ORAC of four palak dishes analysed raw, or wok cooked for 15 minutes (boiled saak) or 25 minutes (saak bhaji)

Dish	Raw	Cooked for 15 minutes	Cooked for 25 minutes
Palak	149 $\pm$ 22	142 $\pm$ 11	136 $\pm$ 17
Plain palak saak	163 $\pm$ 22	134 $\pm$ 22	118 $\pm$ 15
Palak saak with ginger	106 $\pm$ 15	104 $\pm$ 2	108 $\pm$ 6
Palak paneer	49 $\pm$ 1	45 $\pm$ 2	30 $\pm$ 5
Analysis of variation	d.f.	Antioxidant capacity	Isd (5% level)
Cooking time	2	NS	20.67
Dish	3	***	23.87
Dish $\times$ cooking time	6	NS	41.34

\*\*\* Significant  $p < 0.001$ , NS=Not significant ( $p > 0.05$ )

Isabelle et al. (2010) investigated the antioxidant activity of 66 vegetables commonly consumed in Singapore. In general, all leafy vegetables showed antioxidant activity higher than 8.3  $\mu\text{mol TE/g FW}$ . The antioxidant activity of vegetables depended on the cultivar, maturity level and growing conditions such as location, soil state, climate and agricultural practices. As a result, there can be a large variation in antioxidant content within the same vegetable. In the same study, spinach (*Spinacia oleracea* L.) showed H-ORAC activity of 16  $\mu\text{mol TE/g FW}$ , which is similar to the antioxidant capacity of raw spinach (149  $\mu\text{mol TE/g DM}$  or 14.9  $\mu\text{mol TE/g FW}$ ) in the current study. Isabelle et al., (2010) also freeze dried and stored their samples before analysis of antioxidant capacity.

In plain palak saak, made by the addition of Malabathrum leaf (1.1 g), cumin seeds (0.8 g) and turmeric powder (1.2 g) to spinach, the total level of antioxidant capacity marginally increased (9.4%) from 149  $\mu\text{mol TE/g DM}$  to 163  $\mu\text{mol TE/g DM}$  and this was attributed to the antioxidant content in the spices. Based on the individual antioxidant capacity of raw spinach and spices with no interaction between antioxidants, a theoretical calculation showed that both raw plain palak saak and palak saak with ginger should contain the same antioxidant level of 176  $\mu\text{mol TE/g DM}$ . The experimental value of raw plain palak saak is similar to this theoretical value and shows not entirely additive effects of antioxidant capacity. However, when Malabathrum leaf was replaced by ginger powder in the palak saak with ginger, the measured antioxidant capacity decreased, despite the fact that the theoretical capacity should be the same. Interactions between antioxidants can be complex with activities potentially showing straight forward addition of capacity or synergistic interactions where antioxidants work together for greater effect or inhibition. Antioxidants can also act as pro-oxidants and this may be happening in this combination. Moreover, different food matrices may have unpredictable consequences on the overall antioxidant properties.

Among the four boiled saak dishes that were cooked for 15 minutes, the antioxidant capacity was highest in the palak (142  $\mu\text{mol TE/g DM}$ ) followed by plain palak saak (134  $\mu\text{mol TE/g DM}$ ), palak saak with ginger (104  $\mu\text{mol TE/g DM}$ ) and palak paneer (45  $\mu\text{mol TE/g DM}$ ). When the cooking finished after 25 minutes palak showed the highest antioxidant level (136  $\mu\text{mol TE/g DM}$ ) compared to other samples, while the order of ranking for antioxidant capacity was maintained. Ismail et al. (2004) and Amin et al. (2006) both showed significant decreases in antioxidant activity when spinach was boiled. Using bleached  $\beta$  carotene assay, Ismail et al., (2004) noted this decrease after just one minute of boiling in water while Amin et al. (2006) blanched the spinach for 15 minutes and used both bleaching  $\beta$  carotene and DPPH

radical scavenging assays. In contrast, Turkmen et al. (2005) showed an increase in the DPPH radical scavenging activity of boiled, steamed or microwaved spinach while antioxidant levels in other vegetables were either the same or increased by cooking. Chuah et al. (2008) suggested that stir frying was preferable to boiling in water for retaining antioxidant activity when cooking peppers. The current study suggests that wok cooking in an Indian style maintains the antioxidant content of spinach, perhaps because additional water is not added during steaming and the water is evaporated not discarded.

The total antioxidant capacity of palak paneer was decreased by 67 to 78% on addition of paneer (Table 2). Addition of milk has been shown to have a detrimental effect on the total antioxidant capacity of food products like tea and coffee (Sánchez-González et al., 2005; Dupas et al., 2006). Milk predominantly contains casein proteins, which bind phenolic residues of antioxidant compounds. It is assumed that the polar pendant groups (e.g. amino group of lysine and/or carboxylic acid group of aspartic or glutamic acids) of casein proteins chemically interact through hydrogen and/or covalent bonding with the polar group of antioxidants. This hinders the antioxidant capacity and presumably impairs the health benefits (Dupas et al., 2006). It is reported that *in-vitro* antioxidant capacity of coffee can decrease by 40-95% depending on the amount of milk added to the coffee (Sánchez-González et al., 2005; Dupas et al., 2006). Hydroxycinnamic derivatives and their hydrolysed forms, such as caffeic acid, are likely to interact with milk proteins. Similarly, milk reduces the antioxidant capacity of tea extracts by binding with catechins (Dupas et al., 2006). In the present study, the lower level of antioxidants in palak paneer is explained almost entirely by the dilution effect of the added dry matter from the paneer.

## CONCLUSIONS

When expressed on a dry matter basis, plain palak saak contains the highest antioxidant capacity (163  $\mu\text{mol TE/g DM}$ ) among the raw samples. The combination of ingredients has shown a significant effect ( $p < 0.001$ ) on the antioxidant capacity of spinach dishes based on their dry matter. Although, the antioxidant capacity of the spinach samples tended to decrease with longer cooking time, the difference was not significant and so it can be said that Indian cooking method maintains the antioxidant capacity of spinach. The addition of a milk product like paneer did not reduce the antioxidant capacity of spinach, a leafy vegetable, whether it was cooked or uncooked, but diluted the antioxidant capacity of the vegetable in the resulting dish.

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## **Comparative pollen analysis of mānuka and other honeys produced in New Zealand**

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### **ABSTRACT**

New Zealand mānuka honey is recognized for its highly effective antimicrobial activity due to its hydrogen peroxide and antioxidant contents. This study investigated the pollen content of mānuka honey produced in New Zealand and compared it to clover honey from the same region. Seventy four honey samples were used in this study. After pollen analysis, only half the samples labelled as mānuka met the criteria to be called authentic mānuka while most clover honeys could be claimed as authentic clover.

### **INTRODUCTION**

Honey has been used for its highly effective antimicrobial activity because of its hydrogen peroxide content (Bogdanov, 1984; Brudzynski, 2006; Irish et al., 2011; Majtan and Majtan, 2010). In addition, honey is reported to have high antioxidant activity, which is associated with health benefits (Blasa et al., 2006; Gheldof et al., 2003; Schramm et al., 2003) and food preservative properties (Chen et al., 2000; Ergun and Ergun, 2010; Johnston et al., 2005; Krushna et al., 2007; McKibben and Engeseth, 2002; Mundo et al., 2004). Many anecdotal medical studies have demonstrated the use of honey for its wound healing and anti-inflammatory effects (Celepli et al., 2011; Henriques et al., 2006). Kaskoniene and Venskutonis (2010) explained that low levels of pollen could be collected from different flowers and the honey could still be called a monofloral honey. For this reason, it is difficult to use only phytochemical contents in honey as indicators or markers since their compositions might differ. In contrast, Kropf et al. (2010) concluded that some physicochemical measurements could be used to indicate the geographical origin of Slovenian honey. New Zealand mānuka honey is recognised to contain appreciable amounts of peroxide and antioxidants (Allen and Molan, 1991; Lin et al., 2011; Molan et al., 1988). Therefore, some consumers are willing to pay more for mānuka honey compared to a common type of honey, such as clover.

Pollen analysis is a method used to determine geographical and botanical origins of honey. This method can also be used to identify the nectar source of monofloral honeys as indicated by the frequency of different plant pollen in the honey. To date, pollen analysis has been carried out by many different researchers to classify monofloral honey (Abell et al., 1996; Ebenezer and Olugbenga, 2010; Gomes et al., 2010; Lieux, 1981; Malacalza et al., 2005; Ouchemoukh et al., 2007; Pérez-Arquillué et al., 1995; Terrab et al., 2003). However, the frequencies of pollen present in honey can be under- or over-represented in relation to the nectar of the flowers and characteristics of their pollens (Terrab et al., 2002). Pollen from a monofloral honey can be classified into frequency classes as: very frequent >45% for predominant pollen, frequent 16-45% for secondary pollen, rare 3-15% for important minor pollen, and sporadic <3% for minor pollen (Louveaux et al., 1978).

However, the spectrum of pollen count that indicated the same type of monofloral honey could vary because of, for example, geographical origin, forest conditions, floral agriculture where that honey was produced, and contamination from wind-pollinated plants or the use of nectarless plants (Ebenezer and Olugbenga, 2010). Besides this, the filtering method either before analysis or during processing might completely remove pollen from the honey (Moar, 1985; Molan, 1998).

Guidelines for honey quality can be found in the New Zealand Honey Standard for Monofloral Varieties of Honey in the Bee Products Standard Council (2011). Pollen analysis is not the only way that can be used to define a monofloral honey. The codex statement (CODEX STAN 12-1981) (Codex Alimentarius Commission, 2001) "honey may be designated according to floral or plant source if it comes wholly or mainly from that source and has the organoleptic, physicochemical and microscopic properties corresponding with that origin". In commercial practice a honey type is identified using pollen count, colour, conductivity and

profile of sugar contents. Many companies have been carrying out this analysis for a long time and can use their databases (which contain data for up to 26,000 samples) to confirm their identifications (Airborne honey Ltd., Leeston, Canterbury) (Airborne honey, n.d.).

Criteria for pollen of mānuka honey has not been set in these Guidelines (CODEX STAN 12-1981), however, according to a guideline proposed by Moar (1985) to be classified as a mānuka honey, the honey should contain more than 70% mānuka pollen. Unfortunately, some honey brands sold in New Zealand do not reach this minimum standard for mānuka honey. The aim of this study was a preliminary investigation of the pollen count of mānuka honey currently available in the New Zealand market.

## MATERIALS and METHODS

Seventy four honey samples (48 samples labelled as mānuka honey, 25 as clover honey and one as tāwari honey; Figure 1) produced in New Zealand were purchased in 2009 from shops and supermarkets located in Christchurch. Samples were only chosen if they indicated on the label what flower they came from. Clover and tāwari honey samples were purchased to use as comparisons. Pollen analysis was carried out using a method modified from Von Der Ohe et al. (2004). A honey solution was made by mixing 20 g of honey with 20 mL of distilled water and centrifuged (Clements GS 100, NSW, Australia) at 2000 rpm for 5 minutes. The supernatant was decanted off using a pasteur pipette and then the precipitant was diluted to 1 mL with distilled water. This mixture was vortexed for 10 seconds. One drop of solution was put into the Fuchs-Rosenthal Haemocytometer then covered with cover slip and used for the total pollen count. All pollen grains were counted under a microscope (Dialux, Leitz Wetzlar, Germany) and the total pollen calculated:

$$\text{Total pollen} = \frac{80,000}{\text{Number of chambers counted}} \times \text{number of pollen grains}$$

After determining the total pollen count the same tube was centrifuged again at 2000 rpm for 5 minutes and the supernatant was discarded. Then one drop of sediment was placed onto a slide and carefully dried by heating. One drop of warm glycerine jelly (Kaiser's Glycerol Gelatine, Merck & Co, NJ, USA), at temperature of not more than 40°C, was added for mounting. A coverslip was placed on top and left for 1 hour, then the predominant specific pollen was counted from the entire slide or until at least 300-500 pollens were counted. The percentage of predominant pollen was calculated based on total pollen count on this slide.

## RESULTS and DISCUSSION

To be classified as a mānuka honey, the honey should contain more than 70% mānuka pollen (Moar, 1985). The results of pollen analysis (Figure 1) revealed that only 17 samples of the 40 samples labelled as mānuka, could be claimed as authentic monofloral mānuka (ie mānuka pollen exceeded the minimum content of 70%). The rest were categorised into a low mānuka pollen group that included most samples containing mānuka (54% and 10%), tāwari and some samples labelled as clover.

In contrast, most samples labelled as clover honey, including one sample containing 10% mānuka, could be claimed as authentic monofloral clover according to the label as they met the minimum pollen (45%) level required. This is because clover is grown throughout New Zealand and clover honey production covers a wide area compared to mānuka. One honey claimed to be from tāwari flowers, however, tāwari pollen could not be detected in this sample.

The mānuka honey samples were divided into two groups according to the content of mānuka pollen: equal to or more than 70% (17 samples) and less than 70% (35 samples). The high mānuka pollen group (14 samples were labelled as mānuka, two samples were labelled as clover and one sample was labelled as a mānuka honey blend, which contained mānuka 54%) contained pollen in the range of 70-84% (Figure 2). There were 35 samples in the low pollen group. This group contained pollen in the range of 35-69%. Twenty five samples came from samples labelled as real mānuka, two samples were labelled as mānuka 54%, five samples were 10% mānuka, two samples came from clover and one sample came from tāwari (Figure 3).

Honeys with more than 45% clover pollen were classed as clover honey (Figure 4). The clover pollen contents were found to range from 46 to 91%. All samples were labelled as clover honey except for one sample that was labelled as multifloral, which contained mānuka 10%.

The % pollen in high pollen mānuka showed a smaller range than those in the low pollen group or clover (Figure 2, 3 and 4). The average % pollen counts in each category were 75%, 53% and 59%, respectively. Only 36% of mānuka labelled honeys (14 out of the 40) were labelled correctly, while 88% of clover labelled honeys (21 out of the 24) were reliably labelled as clover honey. Thus more than 50% of mānuka labelled honeys purchased in this study could be said to be grossly mislabelled.

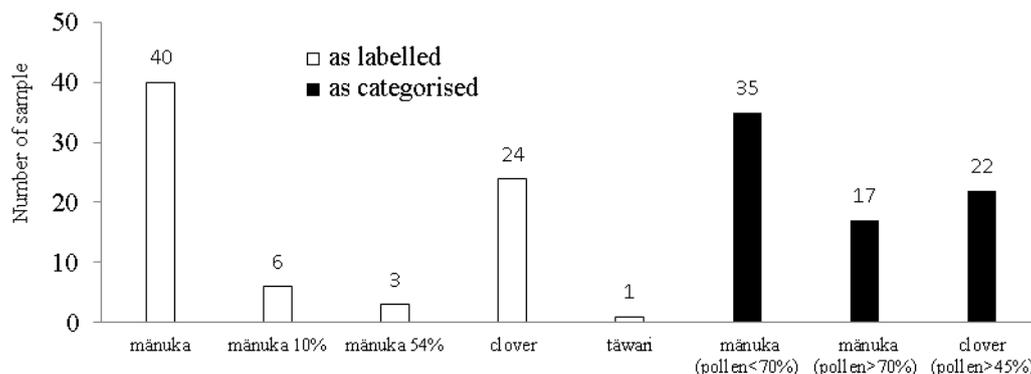


Figure 1: Labelling and categories of honey after pollen analysis

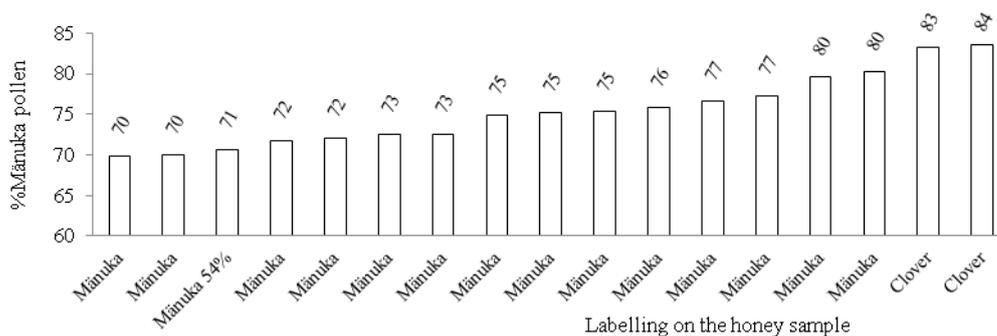


Figure 2: Number of pollen counts in honey classified as mānuka (ie. mānuka pollen > 70%)

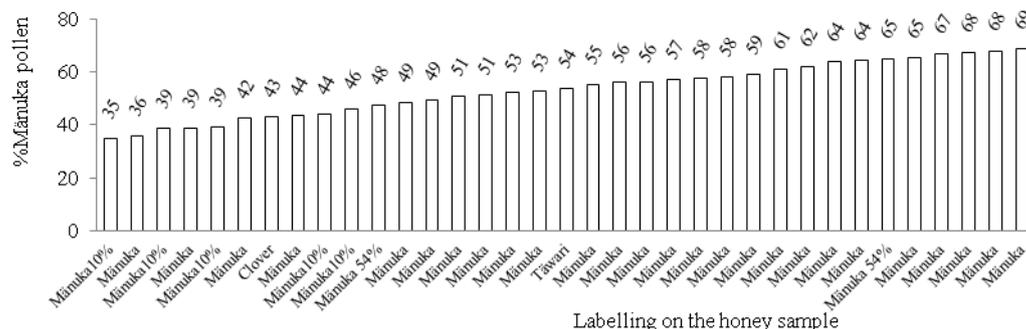


Figure 3: Number of pollen counts in honey classified as mānuka (ie. mānuka pollen < 70%)

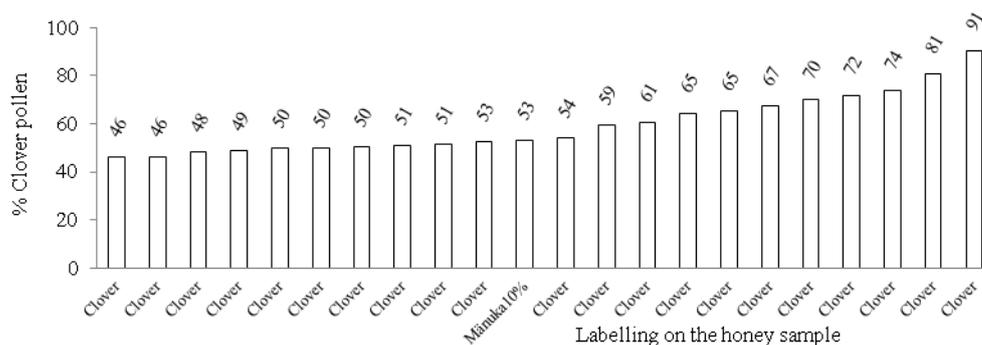


Figure 4: Percentage of pollen counts in honey classified as clover (ie. clover pollen >45%)

## CONCLUSIONS

Some mānuka honey produced in New Zealand could not be trusted to be authentic monofloral honey when using pollen counting guidelines. However, pollen contents are not the only single factor that can be used to discriminate each type of honey. Physicochemical analysis of honey can be used to elucidate the particular characteristics of each honey type and some of these parameters are stated as quality factors according to the Codex Standard for Honey (CODEX STAN 12-1981). Therefore, further investigation into how honey pollen counts relate to the physicochemical properties of different honeys needs to be carried out in order to accurately authenticate the honey source.

## ACKNOWLEDGEMENT

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## DHA supplementation influences cognitive performance in healthy young adults

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**Background:** Docosahexaenoic acid (DHA), a long chain omega n-3 fatty acid, is important for brain structure and function and is dependent on dietary intakes. Individuals following diets low in n-3 may cognitively benefit from increased DHA intake.

**Objective:** To investigate whether a high DHA supplement improves cognitive performance in healthy young adults.

**Design:** Healthy adults (n=176, 18-45 years, non-smoking, low intake of long chain n-3) completed a 6-month randomised placebo controlled double blind trial. Subjects were matched for age and gender and randomly assigned to either DHA (1.16 g DHA/day) or placebo. Cognitive performance was assessed using a computerised cognitive test battery. Z-scores were calculated and the different tests clustered into cognitive domains: memory (word recall, picture and word recognition), working memory, attention, speed of memory and working memory and attention. Intention-to-treat analysis was performed using ANCOVA (controlling for baseline and education) and adding gender as a factor.

**Outcomes:** Memory and working memory improved with DHA compared to placebo in women (mean (95% CI) difference: 0.25 (0.05, 0.45) SD, p=0.01; 0.19 (0.01, 0.36) SD, p=0.04, respectively) but not in men. Speed of working memory improved with DHA compared to placebo in men (reaction time (RT) -0.56 (-0.90, -0.21) SD, p=0.002). Although speed of memory (delayed word and picture recognition) failed to reach significance between treatments (p=0.07), speed of delayed word recognition improved in women (RT -0.34 (-0.59, -0.08) SD, p=0.01). Attention was not affected.

**Conclusion:** DHA supplementation improved memory and speed of memory in healthy young adults whose habitual diet was low in DHA. DHA affected the memory domains differently in men and women with memory and speed of long-term memory improving only in women and speed of working memory improving in men. (Trial registration: ACTRN12610000212055)

**Source of Funding:** Massey University Research Fund and Efamol Ltd, Health & Herbs International Ltd.

## **Validation of a semi-quantitative food frequency questionnaire to assess polyunsaturated fatty acid intakes in the New Zealand population**

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**Background:** Current polyunsaturated fatty acid (PUFA) intakes in New Zealand (NZ) cannot be assessed as there is no valid dietary assessment tool or complete fatty acid database.

**Objective:** To develop, validate and test the reproducibility of a NZ-specific food frequency questionnaire (FFQ) to assess dietary intakes of PUFAs.

**Design:** A 36-item, semi-quantitative NZ PUFA FFQ was developed based on a validated Australian PUFA FFQ. The Australian fatty acid database was adapted to include NZ-specific data for 86% of the major sources of PUFA. Healthy subjects from Auckland, NZ ( $n=48$ ) provided fasting blood samples for biomarker (erythrocyte PUFA) analysis, completed the NZ PUFA FFQ and a 3 d weighed food record (WFR) (validation), and repeated the NZ PUFA FFQ ( $n=41$ ) three months later (reproducibility). The method of triads was used to assess the triangular relationship between the NZ PUFA FFQ, food record and erythrocyte PUFAs; validity coefficients represent the relationship between the NZ PUFA FFQ and true intakes.

**Outcomes:** The NZ PUFA FFQ adequately estimated dietary intakes with strong validity coefficients for eicosapentanoic acid (0.72 [95% CI 0.49, 0.89]), docosahexaenoic acid (0.72 [95% CI 0.53, 0.95]) and total LC n-3 PUFAs (0.68 [95% CI 0.47, 0.89]). Total PUFA, alpha-linolenic acid, linoleic acid, arachidonic acid and total n-6 PUFA were comparable between the NZ PUFA FFQ and WFR. There were no significant differences in intakes of LC n-3 and n-6 PUFAs between repeated implementations of the NZ PUFA FFQ.

**Conclusion:** The NZ PUFA FFQ is a valid and reliable tool to measure PUFA intakes in healthy NZ adults. Using this tool to assess PUFA intakes can provide direction for public health strategies, and will be useful in research and clinical settings.

**Source of Funding:** Institute of Food, Nutrition and Human Health, Massey University.

## Does daily iodine supplementation improve cognition in mildly iodine deficient young adults?

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**Background:** Iodine deficiency during periods of brain development can result in detrimental effects on cognitive ability. In school aged children with mild iodine deficiency, improvements in cognitive test scores have been seen with iodine repletion. The brain continues to develop throughout young adulthood and into the fifth decade of life, however, there is little research into the effect of iodine deficiency on cognition in young adults.

**Objective:** To investigate the effect of daily iodine supplementation on cognition in mildly iodine deficient young adults.

**Design:** A double-blind, randomised, placebo-controlled trial was conducted from July 2010 to September 2011 in young adults aged 18-30y who usually ate <2 servings of bread per day. Participants were randomised to receive 150 µg potassium iodate daily, or placebo, for 32 weeks. At baseline, seven cognitive tests from the Wechsler Adult Intelligence Scale were administered and participants asked to provide a casual urine sample for the measurement of urinary iodine concentration (UIC). All measurements were repeated at the end of 32 weeks.

**Outcomes:** At baseline, 205 young adults (mean age = 21.4 years) participated in the study. The median UIC of the subjects was 65 µg/L (25<sup>th</sup>, 75<sup>th</sup> percentile: 33, 101), between 50-99 µg/L indicating mild iodine deficiency. There were no significant differences in UIC ( $p=0.144$ ) and cognitive test scores for the block design ( $p=0.639$ ), backward digit span ( $p=0.172$ ), matrix reasoning ( $p=0.364$ ), symbol search ( $p=0.465$ ), visual puzzles ( $p=0.641$ ), coding ( $p=0.740$ ), and letter-number sequencing ( $p=0.158$ ) between the placebo and supplemented group at baseline. The results of the intervention will be presented at the conference.

**Conclusion:** Despite mandatory fortification of bread, young adults who consumed <2 servings of bread per day were mildly iodine deficient. An improvement in cognitive test scores in the supplemented group will provide evidence for the continuing role of iodine in brain development in young adults.

**Source of Funding:** Funded by the Maurice and Phyllis Paykel Trust, Laurenson Fund and University of Otago Research Grant.

## **Suboptimal iron status and associated dietary patterns in pre-menopausal women living in Auckland, New Zealand**

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**Background:** Most studies investigating associations between iron status and diet focus on individual nutrients and/or foods. However, individuals do not consume nutrients and foods in isolation, but in a variety of combinations that may interact (for example, ascorbic acid enhances non haem iron absorption). Dietary pattern analysis considers how nutrients and foods are consumed in combination.

**Objectives:** To investigate dietary patterns and their relation to risk of suboptimal iron status in pre-menopausal women living in Auckland, New Zealand.

**Design:** Pre-menopausal women aged 18-44 years (n=375) participated in this cross sectional study. Women completed a 144-item computerised iron food frequency questionnaire (FeFFQ) developed to assess dietary intake patterns over the previous month. The FeFFQ included iron containing foods and foods known to affect iron bioavailability. The 30 most frequently consumed food items from the FeFFQ were entered into a factor analysis. Suboptimal iron status was defined as a serum ferritin <20 µg/L.

**Outcomes:** Seven dietary patterns were identified: refined carbohydrate & fat; Asian; healthy; meat & vegetables; tea & coffee; sandwich; and milk & yoghurt. These patterns explained 44.3% of the variance in intake. Women with high scores on the 'meat and vegetable' dietary pattern were at reduced risk of suboptimal iron status [odds ratio (OR) for quintile 5 versus quintile 1: 0.18; 95 % CI: 0.07, 0.52; p=0.001]. High scores on the 'milk & yoghurt' dietary pattern were associated with increased risk of suboptimal iron status (OR 3.04; 95 % CI: 1.29, 7.16; p=0.011).

**Conclusion:** These results suggest dietary patterns characterised by either a low intake of meat and vegetables, or a high intake of milk and yoghurt, are associated with an increased risk of suboptimal iron status in pre-menopausal women.

**Source of Funding:** This study was funded by the Massey University Research Fund and New Horizons for Women Trust Research Award.

## Iron status and dietary intake of Solomon Island women living in New Zealand

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**Background:** Iron deficiency is a global problem among women of reproductive age, and even more prevalent in women from developing countries. Solomon Island's (SI) recent health and demographic survey reported anaemia (haemoglobin (Hb)<120 g/L) in 44.3% of reproductive aged women. Currently nothing is known about the iron status of women from the SI living in New Zealand (NZ).

**Objective:** To compare iron status and dietary factors influencing iron status of SI and Caucasian women living in NZ.

**Design:** A cross-sectional study comparing 39 SI women with 75 age-matched Caucasian women living in Auckland & Hamilton. Serum ferritin (SF), C-reactive protein (CRP) and Hb were analyzed. Iron status was defined as: SF>20 µg/L + Hb>120 g/L (iron replete), SF<20 µg/L + Hb>120 g/L (low iron stores) and SF<20 µg/L + Hb <120 g/L (iron deficiency anaemia (IDA)). Subjects with CRP>10 mg/L were excluded. A computerized iron food frequency questionnaire was used to assess foods affecting iron status. Anthropometric measurements and blood loss, general health and demographic data were collected.

**Outcomes:** There was no significant difference ( $p=0.899$ ) in prevalence of low iron stores and IDA combined between groups (16.7% SI and 22.5% Caucasian women). The frequency of beef consumed was lower in SI compared to Caucasian women (median (25<sup>th</sup>, 75<sup>th</sup> percentile) 1 (0.5, 2.5) vs 2.5 (1, 2.5) times/wk ( $p<0.001$ ), whereas SI women consumed more kiwifruit 2.5 (1, 7) vs 0.5 (0, 2.5) times/wk ( $p<0.001$ ). Milk as a drink ( $p=0.002$ ), tea ( $p=0.005$ ) and Milo ( $p<0.001$ ) were more frequently consumed by SI women and milk in food ( $p=0.047$ ) and cheese ( $p<0.001$ ) were more consumed by Caucasian women.

**Conclusion:** The iron status of SI women and Caucasian women did not differ, but the intake of foods influencing iron status varied. Prevalence of IDA was lower in SI women living in NZ (developed country) compared to SI (developing country), possibly due to adopting different dietary habits compared to their native country. A different combination of dietary /other factors may be influencing iron status in various groups.

**Source of funding:** IFNHH Postgraduate Research Fund Massey University and New Zealand Aid Programme

## **Can oral supplementation with vitamin B12 reduce vitamin B12 deficiency in South-Asian women of child-bearing age?**

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**Background:** Maternal vitamin B12 (B12) deficiency is associated with an increase in the life-course risk of non-communicable disease for offspring. South-Asian women are particularly susceptible to B12 deficiency due to low or non-meat dietary patterns.

**Objective:** To investigate whether low dose B12 supplementation or individualised B12 dietary advice are effective, appropriate and sustainable in reversing B12 deficiency in South-Asian women of child bearing age.

**Design:** A double blind intervention trial recruiting 63 South-Asian women of child bearing age, stratified by dietary pattern, then randomly allocated to one of three treatment groups (B12 dietary advice, 6mcg B12 supplement or placebo capsule). Participants were tested at baseline, two months and six months for B12 biomarkers (serum B12 and holotranscobalamin [holoTC]), serum folate, glucose, insulin, lipids and full blood count.

**Outcomes:** At baseline, 49.2% of the study participants met the criteria for borderline serum B12 deficiency (B12<222 pmol/L) and 40.7% met the criteria for a low holoTC (<35 pmol/L). None of the participants were very low in serum B12 (B12<110 pmol/L). After six months of treatment the increase in B12 biomarkers was significant for the B12 supplement treatment ( $p<0.05$ ) but not for the placebo or B12 dietary advice treatments. Serum B12 increased by 30% and holoTC by 44% for the supplement group, but decreased by 8.1% (serum B12) and 9.8% (holoTC) respectively for the placebo group. Dietary advice group serum B12 decreased by 2.5%, while holoTC increased by 1.1%.

**Conclusion:** Borderline vitamin B12 deficiency is common in South-Asian women of child-bearing age. A B12 6mcg supplement treatment had a small, but significant effect on increasing B12 biomarkers while placebo and B12 dietary advice treatments had a non-significant effect. Low dose oral B12 supplementation may be an important strategy for preventing B12 deficiency before pregnancy, therefore reducing the non-communicable disease risk from this deficiency for offspring.

**Source of Funding:** Supported by a STAR project PhD Scholarship and a grant from AUT University Faculty of Health and Environmental Sciences Contestable Fund.

## **Skin type and knowledge of vitamin D and sun exposure in New Zealand mothers**

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**Background:** Skin colour is a risk factor for vitamin D deficiency. Darker skin requires longer sun exposure to generate comparable amounts of vitamin D to fair skin.

**Objective:** The objective of this study was to determine whether darker skinned mothers were aware of the relationship between skin colour and risk of low vitamin D.

**Design:** An anonymous, online survey about vitamin D and sun exposure was completed by mothers (n=8004) who had a child 5 yrs or less and living in New Zealand. Questions included knowledge about vitamin D, skin type and sun exposure. Mothers self-identified their own and their child's sun sensitivity type according the Fitzpatrick skin type scale.

**Outcomes:** A sub group of mothers (n=2689) with self-rated Fitzpatrick skin types 3 to 5 (minimally sensitive to sun or sun insensitive) were identified. The majority of these mothers were Caucasian (n=1905) followed by Maori (n=548), Pacific (n=431), Asian (n=158) and Indian (n=121). Most of these mothers (71%) reported that their child also had a Fitzpatrick skin type 3 to 5. When asked whether having darker skin was a risk factor for low vitamin D status 1315 (48.9%) said no and 702 (26.1%) didn't know. The majority of mothers (n=1224, 45.5%) reported that people with darker skin tones did not need to spend longer in the sun to make enough vitamin D and a further 883 (32.8%) were unsure. When asked about their child sunbathing 2193 (81.6%) agreed that it was harmful.

**Conclusion:** These results show that the majority of mothers surveyed, who identified themselves as having darker skin tones, were unaware or unsure of the relationship between skin colour and risk of low vitamin D. This could potentially put mothers and young children with dark skin tones living in New Zealand at risk of vitamin D deficiency and further studies are warranted.

**Source of Funding:** Institute of Food, Nutrition and Human Health, Massey University

## The relationship of vitamin D receptor gene polymorphisms with insulin resistance in vitamin D deficient women

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**Background:** The actions of vitamin D are mediated by the vitamin D receptor (VDR). A number of single nucleotide polymorphisms (SNPs) have been identified on the VDR gene. Genotype may predict risk in diseases related to vitamin D status such as insulin resistance, and response to subsequent supplementation.

**Objective:** To discover any relationships between 5 identified SNPs in the VDR gene and insulin resistance (IR) in women with low vitamin D concentrations; to determine if allelic variants affect the response in IR to vitamin D supplementation.

**Design:** Genotyping of the *Cdx-2*, *FokI*, *BsmI*, *Apal* and *TaqI* SNPs was carried out on 239 South Asian women in Auckland, New Zealand using polymerase chain reaction-based techniques. Associations of these genotypes and 3' end haplotypes with IR were determined using multiple regression analysis. Associations between SNP genotypes and responses in insulin sensitivity to vitamin D supplementation (4000 IU vitamin D<sub>3</sub> per day) were also determined for a subset (81) of these women who participated in a randomised controlled trial.

**Outcomes:** *BsmI* BB, *Apal* AA and *TaqI* tt genotypes were significantly associated with lower insulin resistance compared to *BsmI* bb, *Apal* aa and *TaqI* TT, and homozygosity of the haplotypes baT and BA<sub>t</sub> was associated with higher and lower insulin resistance, respectively, compared to no copies of their respective alleles. Of the subjects supplemented with vitamin D, those with the *FokI* Ff genotype showed a significantly greater improvement in insulin sensitivity (increase of 29.4 (2.9, 38.1)) compared to women with the *FokI* FF genotype (increase of 2.3 (-11.5, 10.1)).

**Conclusion:** This study has highlighted the association of vitamin D responsiveness and insulin resistance with *VDR* gene polymorphisms. This is the first study to determine associations between all three. Genotyping of the *VDR* gene may provide a predictive measure for insulin resistance in response to vitamin D intervention.

**Source of Funding:** Lottery Health, Massey University and AUT University.

## **Mandatory folic acid fortification – who stands to benefit? Results from a New Zealand postpartum survey**

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**Background:** Internationally, poor maternal periconceptional folic acid uptake has been associated with lower socioeconomic status, minority ethnicity status and unintentional pregnancy.

**Objective:** To investigate the effect of proposed mandatory folic acid fortification of bread on associations between maternal factors and recommended folic acid use.

**Design:** Retrospective survey of postpartum women in hospitals and birthing centres across New Zealand using a self-administered questionnaire on supplement use and bread intake in the periconceptional period.

**Outcomes:** Of the 968 women approached, 758 (78%) agreed to participate. Thirty-three percent (33%) of women reported having used folic acid supplements as recommended during the periconceptional period. When the mandatory fortification of bread was modelled, socio-demographic predictors of poor folic acid use, including younger maternal age, increasing parity, non-New Zealand European ethnicity, lower education and less income, were rendered either non-significant or appreciably attenuated. Notably, after controlling for all other significant factors, the odds ratio of pregnancy planning was reduced from 17.23 (95% confidence interval (CI): 8.12–36.55) to 2.60 (95% CI: 1.72–3.92; both  $p < 0.001$ ). Overall, the proportion of women who would achieve adequate folic acid intake increased to 59% with mandatory fortification.

**Conclusion:** Few women comply with periconceptional folic acid recommendations and thus the maximal prevention of neural tube defects is still far from being attained. Health education campaigns to increase awareness of folic acid recommendations will likely widen socio-demographic inequalities in periconceptional folic acid intake, while the data from this study demonstrate that mandatory fortification narrows these inequities.

**Source of Funding:** Funding provided by the Department of Human Nutrition, University of Otago, Dunedin, New Zealand.

## **Bias within the folate microbiologic assay: implications for monitoring folate status**

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**Background:** The microbiologic assay for folate is a “gold standard” assay and is relied upon as an accurate measure of folate status. The microbiologic assay is often used to assess population folate status in national nutrition surveys.

**Objective:** To assess the effect of varying dilution factors on calculated folate concentrations using the microbiologic assay, and to determine the level of agreement between the microbiologic assay and liquid-chromatography tandem mass spectrometry (LC-MS/MS).

**Design:** Plasma and whole blood (WB) folate were measured in samples from 73 participants using the microbiologic assay method of Molloy and Scott (1997). The calibrator used was 5-methyltetrahydrofolate and dilution factors of 1/40 and 1/80 for plasma and 1/800 and 1/1600 for WB were used. WB samples were also measured by a LC-MS/MS method adapted from Fazili *et al.* (2005). Pearson correlation coefficients, Deming regression, and Bland-Altman plots were used to compare the effect of the higher and lower dilution factors used in the microbiologic assay, and to compare LC-MS/MS with the microbiologic assay.

**Outcomes:** Deming regression showed a proportional bias with a slope of 1.79 (95% CI 1.49 – 2.10) and 1.48 (1.33 – 1.64) comparing the lower vs. higher dilution factor for plasma and WB folate respectively. The Bland-Altman relative bias was 31% (limit of agreement -20 – 83%) and 21% (-7 – 48%) for plasma and WB folate respectively. The best agreement was seen at low concentrations, with an increasing difference in calculated folate concentration between dilutions toward higher concentrations. Deming regression comparing the microbiologic assay using a higher dilution factor with LC-MS/MS showed no significant proportional (slope 0.94, 0.77 – 1.11) or constant bias (intercept 23, -17 – 62). The Bland-Altman relative bias was 4% (-25 – 32%) and the Pearson correlation coefficient between these two methods was 0.92.

**Conclusion:** Our data show a discrepancy in the results of the two dilutions whereby the lower dilution (i.e. higher folate concentration) may lead to an overestimation of population blood folate status when using the microbiologic assay.

**Source of Funding:** Supported by an Otago University Research Grant.

## **Food expenditure of food insecure families: a reality check**

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**Background:** The food environment has been proposed as an important factor in terms of influencing diet. A high use and density of fast food outlets in lower socio-economic areas has been proposed as a possible cause of obesity. There is however limited data on actual use of these foods in New Zealand. Information on food purchasing practices can provide some insight into this.

**Objective:** To describe where food insecure households with children are purchasing food and how much they are spending on food.

**Design:** This descriptive analysis uses baseline data from a randomised control trial (Spend Study) conducted in Dunedin between June 2009 and May 2010. Low income (\$45,000/year), food insecure households with children collected food-shopping receipts from all possible sources for four weeks (n=159). When receipts were not obtained households recorded the date the food was purchased, the retail outlet, and an itemised list of the food and associated prices. Food items were classified into groups based on where they were purchased. Many households did not report any expenditure for some food outlets therefore medians and interquartile ranges have been reported.

**Outcomes:** One quarter of this group reported 'low food security' meaning they were reliant on others for their regular food supply. Nearly half received a government benefit as their main source of income and half were single parent families. Weekly median expenditure on food from all food shops was \$40.20 per person (standardised to one adult female). Eighty-five percent of this was attributed to supermarkets, 4% at local food shops and 3% at fast food/takeaway outlets. Median expenditure at restaurants and cafes was zero.

**Conclusion:** Most food purchased by food insecure households with children in Dunedin was from supermarkets with negligible purchases from fast food or takeaway outlets and restaurants and cafes. Further research relevant to the NZ environment into actual consumption of these foods is warranted.

**Source of Funding:** Department of Human Nutrition, University of Otago

**Effects of a free school breakfast programme on children's attendance, academic achievement, and short-term hunger: a stepped-wedge, cluster randomised controlled trial**

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**Background:** Free school breakfast programmes exist in a number of high income countries, but their effects on educational outcomes have rarely been evaluated in randomised controlled trials.

**Objective:** To evaluate the effect of a free school breakfast programme on children's school attendance, academic achievement and short-term hunger.

**Design:** A stepped-wedge, cluster randomised controlled trial was conducted. Schools were randomised to cross over from control to intervention phase (i.e. one-way switch over) in different terms during a full school year.

Setting: 14 New Zealand schools (decile 1-4).

Participants: 424 children, mean age  $9.4 \pm 2$  years, 53% female, 34% Māori, 42% Pacific, 23% New Zealand European/Other ethnicities.

Intervention: A free daily school breakfast programme.

Outcome measures: The primary outcome was children's school attendance. Secondary outcomes were academic achievement; self-reported grades; sense of belonging at school; behaviour; short-term hunger; breakfast habits; and food security.

**Results:** There was no statistically significant effect of the breakfast programme on children's school attendance. The odds of children achieving an attendance rate  $<95\%$  was 0.76 (95% confidence interval [CI] 0.56, 1.02) during the intervention phase, and 0.93 (95% CI 0.67, 1.31) during the control phase, giving an odds ratio of 0.81 (95% CI 0.59, 1.11);  $p=0.19$ . There was a significant decrease in children's self-reported short-term hunger during the intervention phase compared with the control phase, demonstrated by an increase of 8.6 units on the Freddy satiety scale (95% CI 3.4 – 13.7,  $p=0.001$ ). There were no effects of the intervention on any other outcome.

**Conclusion:** A free school breakfast programme did not have a significant effect on children's school attendance or academic achievement, but had significant positive effects on children's short-term satiety ratings. More frequent programme attendance may be required to influence other outcomes.

**Source of Funding:** Health Research Council of New Zealand (09/337). Cliona Ni Mhurchu holds the National Heart Foundation Senior Fellowship (Grant 1380).

## Patterns of stress, mood and eating behaviour in a student population

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**Background:** Daily stress and mood may influence eating behaviour, which may affect health outcomes in the long term.

**Objective:** To investigate the role of daily stress and mood on eating behaviour in a student population and to examine how trait characteristics may moderate these associations.

**Design:** A daily diary survey was carried out over 21 days in 288 undergraduate students at the University of Otago. Students accessed a secure webpage each day and reported the type and intensity of their daily stress, mood, and daily eating behaviours. The moderating effect of Intuitive Eating, Mindfulness, Emotional Eating, BMI, gender, and dietary restriction to lose or maintain weight was measured separately. Hierarchical linear modelling was used to examine associations between daily stressors, mood, and eating behaviour.

**Outcomes:** On days when participants reported greater positive moods, they reported consuming more servings of fruit ( $p < 0.01$ ) and vegetables ( $p < 0.001$ ), yet they also perceived consuming more food than usual ( $p < 0.001$ ). By contrast, the experience of daily stress and negative mood were associated with less healthy eating patterns. Greater total daily stress was associated with a lower number of servings of vegetables per day ( $p < 0.01$ ). Total daily stress was also associated with the self-perception that the amount of food consumed that day was less than usual ( $p < 0.001$ ). On days when participants reported higher negative moods, they also reported consuming more servings of crisps, corn snacks and corn chips ( $p < 0.05$ ), and less fruit ( $p < 0.05$ ). Body Mass Index was positively associated with consuming more unhealthy foods on days when more stress and negative mood was experienced. No other moderators substantively altered the stress, mood, and eating associations.

**Conclusion:** The adverse influence of stress and negative mood is consistent with previous research. A unique finding is the link between positive mood and higher fruit and vegetable consumption. Individuals with a high BMI may be at risk of unhealthy eating behaviours under stress and negative mood.

**Source of Funding:** University of Otago Research Grant.

## **Glycaemic response and glycaemic index for rice in people of European and Chinese ethnicity**

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**Background:** Glycaemic Index (GI) maybe used to guide choice of carbohydrate containing foods. GI is normally determined in small groups of European volunteers and the value thus obtained is assumed to apply to all populations.

**Objective:** Our study aim was to determine whether there are ethnic differences in glycaemic responses and GI to various varieties of rice in people of European and Chinese ethnicity.

**Design:** Sixty-two healthy volunteers, 31 Chinese and 31 Europeans (18-50 yr) consumed 50 g of available carbohydrate portions on separate mornings after a 10 hr overnight fast. Capillary blood glucose was measured at baseline and over a 2 hr period following ingestion of foods (2 x glucose beverage and 1 x five rice varieties: Jasmine, Basmati, Brown, Doongara<sup>®</sup> and Parboiled).

**Outcomes:** Age, height, and sex distribution was not different between the two groups, but body weight and BMI were significantly lower in the Chinese than the European group ( $p < 0.05$ ). Incremental blood glucose areas under the curve (iAUC) of all tested foods were greater in Chinese than in Europeans ( $p < 0.05$ ). The largest difference was for Parboiled rice for which the Chinese iAUC was 77% (95%CI: 38, 226,  $p < 0.001$ ) higher than the European iAUC. In the Chinese and European groups, respectively, the GI of Doongara<sup>®</sup> (67, 55), Jasmine (81, 68), and Parboiled rice (72, 57) were statistically significantly higher in the Chinese.

**Conclusion:** Chinese have a greater glycaemic response to the rice varieties than Europeans. Further study is required to investigate long-term consequences of rice intake in Chinese.

**Source of Funding:** Riddet Institute (Palmerston North, New Zealand) and Performance Based Research Fund (PBRF).

## Breaking prolonged sitting reduces postprandial glycemia & insulinemia in healthy adults

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**Background:** Observational research suggests that breaking sitting time is associated with improved cardiometabolic risk. However, intervention studies are needed to substantiate these findings.

**Objective:** To compare the effects of prolonged sitting on postprandial lipidemia glycemia and insulinemia to a similar duration of sitting combined with either 30 min of continuous physical activity or intermittent short bouts of activity in healthy adults.

**Design:** Seventy (28 males:  $VO_{2max}$  50.8±3.6 ml/kg/min and 42 females:  $VO_{2max}$  37.7±6.6 ml/kg/min) participants completed three, 9 h trials in a randomised order: 1) *Physical Activity (PA)*: participants walked on the treadmill at 60 %  $VO_{2max}$  for 30 min prior to consuming the first meal replacement beverage (MRB) then remained seated for the remainder of the trial; 2) *Breaking sitting time (BS)*: participants walked on the treadmill at the same speed and incline as PA, for 1 min and 40 s every 30 min throughout the trial; and 3) *Prolonged sitting (PS)*: participants remained seated for the duration. In each trial participants were fed three MRBs, each providing 0.46 g fat, 0.54 g protein and 1.12 g carbohydrate per kg of body mass at 60, 240 and 420 min. Venous blood samples were obtained at baseline, and hourly for 9 h, with additional samples taken 30 and 45 min after each feeding.

**Outcomes:** After adjusting for age, sex and BMI, the 9 h triglyceride iAUC was 24% lower in PA, compared to BS ( $p=0.008$ ). The 9 h glucose iAUC was 32.5% and 29.3% lower in BS when compared to both PA ( $p<0.001$ ) and SE ( $p=0.002$ ). The 9 h insulin iAUC was 24.4% and 31.9% lower in BS compared to both PA ( $p<0.001$ ) and PS ( $p<0.001$ ).

**Conclusion:** Regularly breaking sitting time with intermittent short bouts of moderate intensity activity lowers acute postprandial glucose and insulin concentrations in healthy adults. Limiting sitting time by incorporating regular very short bouts of activity may be an important public health intervention

**Source of Funding:** Discretionary funding from the Sports Nutrition and Exercise Metabolism Research Group, Department Human Nutrition, University of Otago.

## **Kernel intactness, carbohydrate digestibility and the relative glycaemic impact of white, wholemeal, multigrain and mixed grain breads**

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**Background:** Breads described in New Zealand (NZ) as wholegrain, multigrain and mixed grain, and promoted for their health benefits, usually contain a proportion of intact or partially intact (cracked) kernels, but whether kernel intactness and the bread descriptor used reliably indicate a low glycaemic impact of the breads is uncertain.

**Objective:** To determine whether or not the terminology used to promote NZ supermarket breads, and the appearance of semi-intact grain kernels in them, is useful to consumers as a guide to breads of low glycaemic impact.

**Design:** Eight NZ breads described as white, wholemeal, multigrain mixed grain and low GI were digested in vitro either intact (I) or after homogenising (H) to remove grain structure, and determine their relative glycaemic impact (RGI) determined as grams of glucose equivalents (GGE) per serving and per 100g. The breads were also digested under gastric conditions to remove and measure intact kernels and kernel fragments from the breads.

**Outcomes:** Digestion profiles of I and H versions of white, wholemeal, wholegrain and low GI white bread were similar, although the breads differed in relative glycaemic impact (RGI; g glucose equivalents/100 g bread) because they differed in digestible carbohydrate content. Intact kernels or kernel fragments made little difference to the digestion profiles of some grainy breads, but made a small difference in others. Based on rapidly available (20 min) carbohydrate, the RGI of I and H samples ranged from I =  $34.3 \pm 0.5$  GGE/100 g and H =  $34.0 \pm 1.6$  GGE/100 g for a white bread to I =  $18.1 \pm 0.3$  GGE/100 g and H =  $23.0 \pm 1.1$  GGE/100 g for a Wholemeal and Grain bread. The main factors determining glycaemic impact per serving were available carbohydrate content, and conversely, water content of the breads.

**Conclusion:** Description as wholegrain and wholemeal, and appearance of intact kernels in breads available in NZ supermarkets, is not a reliably effective guide to breads of low glycaemic impact, because the proportion of intact kernels in the breads available is too low to sufficiently retard carbohydrate digestion.

## Effects of resistance training and detraining and muscle fibre type on insulin responses to a glucose load

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**Background:** Resistance training can enhance insulin sensitivity. Literature is limited in type 2 diabetic (T2DM) populations and in those with a familial link. Muscle fibre types vary in their glucose handling and training and detraining can influence muscle fibre composition.

**Objective:** This study was designed to investigate glucose and insulin responses to an oral glucose load, muscle fibre type and muscular performance, in offspring of T2DM and control subjects, with resistance training and detraining.

**Design:** Six familial insulin resistant (FIR) and 10 control (C) subjects did 9 wk resistance training, then 9 wk detraining. At baseline (T1), after training (T2) and after detraining (T3), performance measures, an oral glucose tolerance test and myosin heavy chain (MHC) fibre composition were taken.

**Outcomes:** 3RM increased similarly in both groups in leg press, leg curl and squat (all  $p \leq 0.001$ ). Wingate peak power also increased ( $p \leq 0.005$ ). Only strength decreased with detraining ( $p \leq 0.001$ ). Training reduced insulin area under the curve (AUC) to a greater extent ( $p = 0.050$ ) in FIR (T1, T2:  $1219 \pm 734$ ,  $837 \pm 284$  pmol/L, resp.) than C (T1, T2:  $647 \pm 268$ ,  $635 \pm 258$  pmol/L, resp.). Detraining increased insulin AUC, with a larger ( $p = 0.018$ ) insulin AUC in FIR (T2, T3:  $837 \pm 285$ ,  $1040 \pm 194$  pmol/L, resp.) than C (T2, T3:  $635 \pm 258$ ,  $625 \pm 213$  pmol/L, respectively) No differences in MHC isoform distribution between groups or with training were observed. MHCIIIX fibres increased with detraining ( $p = 0.026$ ). A positive correlation between fasting insulin concentration and MHCIIA content was observed after training ( $p = 0.046$ ) and an inverse correlation between fasting glucose concentration and MHCIIIX after detraining ( $p = 0.027$ ).

**Conclusion:** Familial insulin resistant individuals exhibit a greater training effect to reduce insulin AUC and, in contrast to controls, an increase with detraining. Fibre type plays a role in insulin and glucose responses, but cannot fully explain differences with training or between groups.

**Source of Funding:** Funding was received from the Sports Nutrition and Exercise Metabolism Research Group, Otago University.

## **The relationship between our food choices and farm animal welfare**

**K BICKNELL**

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Not only do the choices we make about food influence our own health and well-being, but they may also affect the welfare of the animals that are farmed to produce our food. In this presentation I will consider the topic of farm animal welfare from an economic policy perspective. I will begin by identifying some of the factors that contribute to poor versus enhanced animal welfare. With these drivers in mind, I will discuss the role of the government in regulating animal welfare in general, and more specifically what is being done in New Zealand to ensure that our food is produced with an adequate standard of welfare. Given that enhanced animal welfare is likely to be achieved only at an increasing cost, I will also look at the distributional impacts of animal welfare policy, focusing in particular on who ultimately bears the cost of improving animal welfare. Finally I will consider farm animal welfare from a 'global' perspective, addressing the question of relative animal welfare standards, what varying standards might mean from the perspective of international trade and, ultimately, the amount of welfare that is 'embodied' in the food that we eat.

**Source of Funding:** None.

## **Parents know breakfast is the most important meal of the day, so why are so many children not eating breakfast?**

RM WHITING, SJ MURRAY

*Health Sponsorship Council, Wellington, New Zealand*

**Background:** Each school week over 100,000 children aged five to 14 years skip breakfast at least one day of the week. Breakfast has been linked to improved academic and physical performance, and is associated with less snacking and less weight gain. Pacific children are 5.7 times more likely and Maori children are 2.5 times more likely to skip breakfast than NZ European or other children.

**Objective:** To provide insight into the usual morning routines for some low-income Maori and Pacific families in the morning and to examine the barriers to, and facilitators of, breakfast consumption in children aged eight to 16 years.

**Design:** Focus group interviews were conducted with 23 low-income Maori and Pacific parents (two Maori groups, two Pacific groups) who were in charge of the household's morning routine and whose children ate breakfast irregularly or did not eat a cereal or bread-based breakfast. Focus group interviews were analysed using thematic analysis.

**Outcomes:** Parents had heard breakfast is important but many did not know why or did not believe the message. Parents who did not eat breakfast themselves did not encourage their children to eat breakfast. Cost was the main reason for choosing breakfast food – nutritional benefits of the food were never mentioned. Parent's perceptions of why children did not eat breakfast included their children's focus on appearance (e.g. preening), skipping breakfast to avoid weight gain, not having breakfast made for them and parents not eating breakfast themselves. Most parents thought children older than eight or nine could make breakfast themselves and handed responsibility for, and overall decision making about, breakfast to the child.

**Conclusion:** Parental valuing and role-modeling of breakfast is important for increasing children's breakfast consumption. Parents retaining responsibility for their child's breakfast may slow the age-related decrease in breakfast consumption. Messages aimed at increasing breakfast consumption need to focus on fast, easy, low-cost breakfast solutions and the functional benefits of breakfast.

**Source of Funding:** Health Sponsorship Council is funded by Ministry of Health.

## Reliability of a Food Frequency Questionnaire (FFQ) for New Zealand adolescents

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**Background:** As there is currently no up-to-date, validated food frequency questionnaire (FFQ) for use in New Zealand adolescents, there is a need to develop one as a cost-effective way to assess adolescent's food patterns in health and nutrition studies.

**Objective:** This study aims to examine the test-retest reliability of an FFQ to describe food patterns in adolescents aged 14 to 18 years.

**Design:** A non-quantitative FFQ comprising 72 food items from seven major food groups was developed and pretested. Fifty-two subjects (28 males, 24 females) aged  $14.9 \pm 0.8$  years completed the FFQ twice within a two-week period to assess short-term test-retest reliability.

**Outcomes:** Spearman's correlation tests showed that reliability ranged from 0.33 for peaches to 0.87 for non-standard milk. No more than 10% of participants were grossly misclassified for any food, and correct classification ranged from 54% (chocolate confectionary) to 94% (pizza). This high reliability was ascertained by the average weighted kappa value of 0.51 (95% CI: 0.46, 0.55). Among the seven food groups, milk and dairy products showed the best reliability ( $\rho=0.76$ ,  $\kappa_w=0.70$ ).

**Conclusion:** The FFQ exhibited excellent test-retest reliability in the estimates of intake frequency, particularly for foods consumed regularly. Further validation is currently underway to determine the relative validity of this FFQ.

**Source of Funding:** Supported by grants from the National Heart Foundation, Lottery Health NZ and the University of Otago.

## **Factors associated with body mass index in a nationwide cohort of mid-age women**

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**Background:** Autonomous and controlled styles of eating regulation (as described by Self-Determination Theory) and speed of eating may be important in influencing weight gain and degree of obesity. However these characteristics have rarely been investigated in representative samples of adult women.

**Objective:** To examine, in a nationwide sample of women, the association between different styles of eating behaviour regulation (autonomous and controlled) and body mass index (BMI), with specific eating habits as mediators, as well as the association between speed of eating and BMI.

**Design:** A nationwide study of 2500 randomly selected New Zealand women aged 40-50 years, using electoral rolls for the entire country. The survey yielded a 66% participation rate (n=1601).

**Outcomes:** Autonomous forms of regulation were negatively associated with BMI while controlled forms of regulation were positively associated with BMI. When controlling for confounding variable as well as specific food patterns that were possible mediators, there was a statistically significant inverse association between autonomous regulation and BMI (2.0% decrease for every 10-unit increase in autonomous regulation; 95% CI: 1.4%, 2.7%; p<0.001) and a statistically significant positive association between controlled regulation and BMI (1.4% increase for every 10-unit increase in controlled regulation; 95% CI: 0.4%, 2.3%; p=0.005). The relationships between autonomous regulation and BMI and controlled regulation and BMI were partially mediated by the specific food patterns measured. After adjusting for confounding variables, BMI statistically significantly increased by 2.8% (95% CI: 1.5%, 4.1%; p<0.001) for each category increase in self-reported speed of eating.

**Conclusion:** Autonomous styles of eating behaviour regulation are associated with lower BMI in mid-age women, and the associations are partially mediated by the specific food patterns measured. The results also suggest that faster eating is associated with higher BMI in mid-age women.

**Source of Funding:** Supported by the Department of Human Nutrition, PBRF fund, University of Otago, Dunedin, New Zealand.

## Postprandial response of subcutaneous adipose tissue in healthy adults

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**Background:** Adipose tissue (AT) is an important endocrine organ in the regulation of metabolism. It has been identified as one of the first sites of meta-inflammation, which is a key event in the development of insulin resistance. While diets chronically high in saturated fat are known to induce AT dysregulation, less is known about the acute impact of nutrients on AT inflammation and metabolism.

**Objective:** To measure acute, postprandial changes in gene expression of subcutaneous AT in healthy volunteers following the consumption of a lipid or carbohydrate beverage.

**Design:** Thirty-three healthy adults were recruited and randomised to consume one of three beverages; placebo (0 kJ), lipid (1988 kJ) or carbohydrate (1856 kJ). Subcutaneous AT biopsies were taken at baseline (0 h), as well as 2 and 4 h following consumption of the beverage. RT-PCR was performed to measure changes in AT gene expression.

**Outcomes:** Groups were matched for height and weight. No differences were observed in adipokine gene expression between each group (*MCP-1*, *IL-6*, *TNF- $\alpha$*  and *Adiponectin*). Expression of metabolic gene *IRS2* was reduced with carbohydrate consumption at 2 h ( $p < 0.001$ ) and 4 h ( $p < 0.01$ ). In addition *PKD4* was reduced following carbohydrate consumption at 2 h ( $p < 0.001$ ) and 4 h ( $p < 0.001$ ); and after lipid consumption at 4 h ( $p < 0.05$ ). No changes were observed in *LPL*, *PPAR $\gamma$*  or *FAT-CD36*.

**Conclusion:** Carbohydrate and lipid differentially affect gene expression in subcutaneous AT. Understanding the impact of these macronutrients on AT may provide insight into the development of metabolic dysregulation of AT.

**Source of Funding:** Not applicable.

## **Forget schools: how do we tackle the home environment for obesity prevention?**

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**Background:** Considerable public attention and debate has surrounded the role that schools can or should play in obesity prevention in New Zealand. In contrast, the importance of the home food environment and its contribution to weight in children has received relatively little scrutiny.

**Objective:** To discuss the evidence regarding the potential effectiveness of different approaches to tackling obesity within the home.

**Design and Outcomes:** The importance of the home food supply is illustrated by national data reporting that although New Zealand children aged 5-14 years consume about one-third of their total energy intake while at school, the vast majority of this food (80%) comes from home. Important factors which might influence the types and amount of food available within the home, range from macro-level factors including the political and economic environment (advertising, government policies, and food pricing), to micro-levels factors such as parenting style, family eating patterns and work schedules. The contribution of these factors and how they might be manipulated to change the home environment in some way will be discussed.

**Conclusion:** The importance of the home environment should not be underestimated, and efforts to improve dietary intake and maximise health should concentrate where the majority of energy is consumed.

**Source of Funding:** R Taylor is supported by the Karitane Fellowship in Early Childhood Obesity.

## Macronutrient balance and energy intake: the protein leverage hypothesis

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**Background:** A significant contributor to the rising rates of human obesity is an increase in energy intake. The 'protein leverage hypothesis' proposes that a dominant appetite for protein in conjunction with a decline in the ratio of protein to fat and carbohydrate in the diet drives excess energy intake and could therefore promote the development of obesity.

**Objective:** To test the 'protein leverage hypothesis' by comparing energy intakes of humans on manipulated diets containing high, medium and low protein density.

**Design:** Energy intakes were measured for 22 lean subjects studied over three 4-day periods of in-house dietary manipulation. Subjects were restricted to fixed menus in random order comprising 28 foods designed to be similar in palatability, availability, variety and sensory quality and providing 10%, 15% or 25% energy as protein. Nutrient and energy intake was calculated as the product of the amount of each food eaten and its composition.

**Outcomes:** Lowering the percent protein of the diet from 15% to 10% resulted in a 12% higher total energy intake ( $p=0.02$ ), predominantly from savoury-flavoured foods available between meals. In contrast, increasing protein from 15% to 25% did not alter energy intake

**Conclusion:** In our study population a change in the nutritional environment that dilutes dietary protein with carbohydrate and fat promotes overconsumption, thus enhancing the risk for weight gain.

**Source of Funding:** Australian National Health and Medical Research Council; National Research Centre for Growth and Development, New Zealand.

## **Protein and carbohydrate for weight control: what role for hunger and appetite regulation?**

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**Background:** Long term weight loss is notoriously difficult to achieve. While it is clear that maintaining intake below expenditure will lead to weight loss, the challenge is to understand how best to suppress intake long term. A wealth of data points to the suppression of appetite playing an important role in weight control, but whether weight gain is a result of poor satiety and constant hunger or whether sensory aspects of food (hedonics) and ready access overwhelm our regulation of appetite is poorly understood. Certainly many factors encourage over-consumption and we question how much hunger and satiety drive our eating anymore. Our environment is one of varied food choices, 'fast' foods, snack foods and food advertising. While the macronutrient content and composition of the diet still have an important role to play in the regulation of intake, many questions remain unanswered.

**Outcomes:** Any restrictive diet, given sufficient adherence, can be successful but most important to understand is whether there is an optimal composition that best suppresses food intake when eaten freely (*ad lib*). Recent studies have shown diets moderately high in protein and [low glycemic index (GI),] carbohydrate (CHO) may best promote weight loss. Whilst *ad lib* studies show higher protein diets suppress food intake, the role of CHO, and GI in particular, on appetite may be less clear. Also important is the issue of poor weight loss long term. Whether this is a decline in motivation and poor adherence to diet or a physiological adaptation of hunger to a diet-induced energy deficit is also unclear. Recent exercise studies are revealing. In some individuals hunger is stimulated in response to an exercise-induced energy deficit and these individuals fail to lose weight. What are the mechanisms, and who is most susceptible to success and who is susceptible to failure is not yet known.

**Conclusion:** Eating is a behaviour modified by both physiological (eg. hunger, satiety) and psychological (eg. hedonics, motivation) parameters. Targeting appetite suppression through high protein, high CHO (high fibre, low GI), lower energy-dense diets appears a good strategy but individual success may be variable and require changes in the wider food environment before such diets can be successful for all.

**Source of Funding:** Not applicable

## **Lipidomic analysis of chylomicron response following ingestion of high fat dairy- and soy-based breakfasts in men with metabolic syndrome**

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**Background:** Postprandial hyper-triglyceridaemia and delayed chylomicron (CM) clearance have been implicated as independent risk factors for CVD.

**Objective:** The aim of this investigation was to evaluate the effect of high fat breakfast meal (containing 54 g of fat from dairy foods or 54 g fat from soy-based foods) on postprandial TAG composition of CMs in men with or without MetS.

**Design:** A randomised crossover trial where a mixed breakfast with dairy foods or soy-based foods was compared. The molecular species of TAG and selected regioisomers of CM TAGs were measured by LC-MS/MS, and fatty acids by GC.

**Outcomes:** Postprandial CM TAG concentrations were significantly lowered in both control and MetS subjects fed the dairy breakfast relative to the soy-based breakfast at 3 hours. The chylomicrons from the dairy group at 3h contained two thirds of the 80 molecular TAG species found in the meal suggesting that most of the short and medium chain fatty acids were transported after absorption via the portal venous route. Long chain saturated TAG were also lost from the soy breakfast lipids in comparison with CM from the soy group.

**Conclusion:** Dietary TAG composition influences postprandial CM TAG levels. The lowered postprandial response observed after the dairy meal was likely a result of limited short and medium chain FA incorporation into CM suggestive of selective fatty acid and/or monoacylglycerol absorption, formation and/or clearance. The implications of the findings from this acute research study should be explored in longer-term studies.

**Source of Funding:** This research was supported by funding from Dairy Health & Nutrition Consortium, a consortium of Tatura Milk Industries & Bega Cheese, National Foods, Fonterra Australia, Parmalat Australia, Dairy Australia, Geoffrey Gardiner Foundation, Murray Goulburn Co-operative, Warrnambool Cheese & Butter Factory, and Dairy Innovation Australia.

## **Added carbohydrates in children's milk products increase dietary glycaemic load**

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**Background:** Milk is a staple food for children, delivering high quality protein, essential nutrients such as calcium, riboflavin, vitamin A and zinc. In Asian countries a new category of high value milk products for children, 'Growing Up Milks', is well established and these products are now appearing in New Zealand and Australia. Manufacturers often add additional carbohydrates, including sucrose, maltodextrins and corn/glucose syrups which theoretically contribute to a higher glycaemic index (GI) and glycaemic load (GL). Diets with a high GI/GL may increase the risk of developing obesity and type 2 diabetes in adulthood.

**Objective:** To survey growing up milk products in Malaysia and Indonesia to determine the content of added carbohydrate ingredients and to assess the impact on blood glucose responses.

**Design:** A total of 24 products from Malaysia and 32 from Indonesia were surveyed. The ingredient lists and nutrition information panels were used to calculate the percentage of declared carbohydrates coming from added sources, excluding fibre. A representative subset of products were tested for their GI according to International Standards Organisation methodology. The Glycaemic Load was calculated as GI x carbohydrate per serving/100.

**Outcomes:** The range of added carbohydrate content in the products ranged from 0 to 40% w/w. Milk powders without added sources of carbohydrate had similar GI values to liquid milk, ~30, and a low GL. Products containing maltodextrins and corn/glucose syrups increased the GI and GL by more than 2-fold.

**Conclusion:** Growing up milk products targeted at young children may contain excessive levels of added carbohydrates. Irrespective of chain length, they may increase the risk of overweight, obesity and diabetes.

**Source of Funding:** Funding for this work was provided by Fonterra Brands, Asia Middle East.

## Microbial phytase improves bone mineral density (BMD)

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**Background:** Phytate is the major phosphorus store in plants. Phytate can bind complex compounds, particularly divalent cations, present in foods and feedstuffs thereby reducing the availability of dietary minerals and other nutrients when plant-based foods are consumed. Phytase hydrolyses phytate and microbial phytase is often included into pig and poultry diets to enhance nutrient availability.

**Objective:** To explore the impact of dietary microbial phytase supplementation on BMD and Ca retention in poultry.

**Design:** 1-day-old broiler chickens (60 birds per treatment) were fed a corn-soya bean meal-based diet (CS) for 21 days. Treatments consisted of the CS diet and the CS diet supplemented with 1000 Units/kg of either microbial phytase A (expressed in *Aspergillus oryzae*) or phytase B (expressed in *Schizosaccharomyces pombe*). A Ca balance study was conducted over days 18 – 21 to determine Ca retention. On day 22 the birds were euthanized and the left leg removed for tibial BMD analysis.

**Outcomes:** The tibia BMD of the broilers fed the CS diet was 186 mg cm<sup>2</sup>. Dietary supplementation with either phytase A or B led to significantly ( $p < 0.001$ ) higher tibia BMD (225 and 210 respectively). Ca retention in the birds fed the CS diet was 50.4%. Ca retention was significantly ( $p < 0.001$ ) higher for the birds fed the phytase supplemented CS diets (56.6% and 54.7% for phytase A and B respectively). Dietary supplementation with phytase A led to significantly ( $p < 0.05$ ) higher Ca retention and BMD compared to phytase B.

**Conclusion:** Dietary supplementation with either microbial phytase improved Ca retention and BMD in the broiler chickens, but the extent of the improvement was phytase dependent. Other studies using pigs and rats have also reported improved Ca retention and BMD when receiving phytase supplemented diets. Studies have not yet been conducted exploring the impact of dietary microbial phytase supplementation in humans but supplementation may have a role in improving mineral availability and bone health in humans.

**Source of Funding:** Funding from DSM Nutritional Products, Switzerland.

**Nutritional and environmental risk factors for young children in Auckland, New Zealand, developing community acquired pneumonia – a case-control study**

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**Background:** New Zealand has a high pediatric pneumonia hospitalisation prevalence rate (11 per 1000 in children less than five years) compared with other developed countries (0.1 – 2 per 1000). A number of risk factors have been identified in both developing and developed countries for contracting pneumonia; these include nutritional factors.

**Objective:** The objective of this study was to identify the nutritional and environmental risk factors associated with preschool aged children in New Zealand developing community-acquired pneumonia.

**Design:** We conducted a case-control study of children residing in Auckland, New Zealand, aged less than 5 years of age. The cases were children admitted to Starship Children's Hospital or presenting at the emergency department of the same hospital with pneumonia. The controls were randomly selected children living in the community. The children were matched for age and ethnicity. Data was collected by a face to face interview with the child's caregiver; this included a general questionnaire and a specific dietary questionnaire.

**Outcomes:** There were 856 children included in the analysis, 505 cases and 351 control children. The univariate analysis found a number of nutrition-related risk factors; no breastfeeding during infancy (OR 2.56, 1.54-4.25), no sunlight exposure in the previous month (OR 2.56, 1.63-4.02). In the over 2 year old children other risk factors identified were less than 2 serves of breads and cereals per day (OR 3.77, 1.40-10.18), and higher consumption of takeaway-type foods (OR 2.44, 1.05-5.55). In the under 2 year old children, consumption of fruit juice was also a risk factor (OR 1.46, 1.01-2.10). In the multivariate analysis, the only 2 nutrition-related risk factors to remain were no sunlight exposure in the previous month (OR 2.54, 1.47-4.39), and not having been breastfeed during infancy (OR 1.87, 1.02-3.41).

**Conclusion:** Identification of controllable risk factors can lead to improvements in policy, primary care and prevention of hospitalisation.

**Source of Funding:** HRC Research Grant, Top Doctoral Scholarship

## **Micronutrient intake and supplement usage of pregnant and lactating women in New Zealand**

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**Background:** Maternal nutrition status during pregnancy and lactation should be positively optimized to enhance nutritional support during fetal and infant growth and development.

**Objective:** This study explores a range of essential nutrient intakes and how dietary supplement usage impacts upon pregnant and lactating women in New Zealand.

**Design:** Qualifying women were recruited nationally. Face-to-face and telephone interviews provided individually three 24h dietary recalls. Dietary intake data from 66 pregnant (P) women (after 26 weeks gestation) and 88 lactating (L) women (21 days after delivery) was analysed by Foodworks 2009 and SPSS 17 Software.

**Outcomes:** Intakes of total folate (P: 67%; L: 49%), dietary fibre (P: 73%; L: 80%), selenium (P: 72%; L: 66%), iron (P: 53%), and vitamin D (L: 72%) were all below EAR levels. Two intakes were above the upper limit: total folate (P: 14%; L: 13%) and iron (P: 33%; L: 6%). Pregnant (47%) and lactating (42%) women used a range of supplements. Comparisons made between supplement and non supplement users during pregnancy gave statistically significant differences: vitamin D ( $6.77 \pm 5.67$ ;  $2.26 \pm 1.99$ ,  $p=0.000$ ), vitamin E ( $25.64 \pm 42.18$ ;  $9.99 \pm 7.25$ ,  $p=0.036$ ), total folate ( $746.22 \pm 488.44$ ;  $359.83 \pm 155.99$ ,  $p<0.001$ ), iron ( $82.52 \pm 68.67$ ;  $14.01 \pm 5.15$ ,  $p<0.001$ ) and selenium ( $56.59 \pm 49.79$ ;  $44.50 \pm 18.20$ ,  $p=0.040$ ). Lactating women, had similar results for vitamins D, E and iron, and also intakes of zinc ( $16.38 \pm 6.77$ ,  $12.26 \pm 3.76$ ,  $p=0.002$ ) and copper ( $14.70 \pm 75.60$ ,  $1.86 \pm 0.77$ ,  $p=0.02$ ) which were significantly different.

**Conclusion:** Almost half of all participants used supplements. Many women were either below EAR or above the Upper Limit for some micronutrients. Any potential risk during this critical period of human reproduction needs careful understanding prior to dietary supplement use. Enhanced public awareness of appropriate nutritional use of such supplements is essential for all pregnant and lactating women.

**Source of Funding:** Massey University Research Fund.

## Methodology of the 2008/2009 New Zealand Adult Nutrition Survey

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**Background:** National nutrition surveys provide valuable information for comparing changes in the nutrition status of populations over time. The comparability of survey results depend on the methodology used. The 2008/09 New Zealand Adult Nutrition Survey (2008/09 NZANS) is the fourth population-based nutrition survey in adults.

**Objective:** To describe the methodology used in the 2008/09 NZANS, with reference to the unique aspects of the current survey compared with the 1997 National Nutrition Survey (1997 NNS).

**Design:** The 2008/09 NZANS was a nationally representative, cross-sectional survey of 4,721 adults 15 yrs and over. Participants were recruited using a three-stage, stratified approach, with oversampling of Māori, Pacific, and individuals under 19 yrs and over 70 yrs. Interviews were conducted within the participant's home. A computer-assisted, multiple-pass 24 hr recall was used to collect dietary data. Information on dietary habits, nutrition-related health and food security were obtained through interviewer-administered questionnaires. Height, weight, waist circumference and blood pressure measurements were taken for consenting participants. Blood and urine samples were collected at local clinics from 3,348 and 3,315 participants, respectively.

**Outcomes:** The 2008/09 NZANS achieved a higher response rate (61%) and greater numbers of Māori (n=1,040), Pacific (n=757), under 19 yr (n=699) and over 70 yr (n=1,065) participants than the 1997 NNS. In the survey reports, the categorisation of ethnicity and definitions of overweight and obesity for Māori and Pacific differed. Differences in the Estimated Average Requirement for some nutrients influenced the calculation of the prevalence of inadequate intakes between the surveys.

**Conclusion:** The methodologies used for the 2008/09 NZANS and 1997 NNS were largely similar, although some differences exist which should be considered when comparing and interpreting the results from the reports.

**Source of Funding:** The Ministry of Health, New Zealand, funded the 2008/09 NZANS and the 1997 NNS.

## **Disentangling the obesity epidemic: Results from the 2008/2009 New Zealand Adult Nutrition Survey**

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**Background:** Obesity rates continue to increase worldwide. Documenting and explaining national prevalence rates is necessary in order to find solutions to the problem.

**Objective:** To report current prevalence of overweight and obesity in New Zealand, changes in rates since the 1997 National Nutrition Survey (1997 NNS), and to examine relationships with energy intake.

**Design:** Secondary analysis of the 2008/2009 New Zealand Adult Nutrition Survey (2008/09 NZANS) and the 1997 NNS. The same BMI cut-offs were used and prioritised ethnicity.

**Outcomes:** The overall age-standardised prevalence of obesity is significantly higher in the 2008/09 NZANS (27.1%, 95%CI: 25.1, 29.1) than in 1997 (19.1%, 95%CI: 17.6, 20.6). Overweight and obese adults now account for 62% of the total population over 15 yrs compared with 52% of the population in 1997. Increases have occurred in NZ European/Other (NZEO) men (13.2 to 25.2%), Māori women (35.5 to 49.3%) and NZ European/Other women (17.9 to 22.8%). While the prevalence of obesity is highest in Pacific (58.4% versus 45.4% in Māori and 24% in NZEO), 72.1% of all obese adults are NZEO. Mean reported energy intakes appear to have declined. Significant decreases have occurred only in normal weight men and women, and overweight men. There has been no change in reported energy intakes among obese adults. There is no relationship between energy intake and BMI in the NZ population. However when only considering data from those predicted to be reliable reporters (using the McCrory method), total mean energy intakes remain unchanged and there is a clear positive relationship between energy intake and body weight status.

**Conclusion:** The greatest increases in prevalence of obesity have occurred in NZEO men and Māori women and the vast majority of obese New Zealanders continue to be of NZEO ethnicity. The apparent decline in reported energy intakes has occurred in non-obese adults only. Our in-depth analysis of the data provides some evidence that energy intake contributes to increased rates of obesity.

**Source of Funding:** The Ministry of Health, New Zealand, funded the 2008/209 NZANS and the 1997 NNS.

## **The prevalence of diabetes and pre-diabetes in New Zealand – will the health system cope with demand?**

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**Background:** Diabetes is pandemic and the prevalence of diabetes continues to increase worldwide. To date, national diabetes prevalence data for New Zealand have used self-reports of diagnosed diabetes, which underestimate prevalence.

**Objective:** To describe the epidemiology of diabetes and pre-diabetes for New Zealand adults using data from the 2008/09 New Zealand Adult Nutrition Survey (2008/09 NZANS).

**Design:** Diabetes was defined as self-reported diabetes, undiagnosed diabetes as HbA1c  $\geq$  6.5% but not self-reported, and prediabetes as HbA1c between 5.7% and 6.4% but not self-reported. Collectively diabetes (self-reported and undiagnosed) and pre-diabetes were described as glucose metabolism disorders. Data were weighted, and proportions and age-standardised (WHO) rates were calculated.

**Outcomes:** The prevalence of self reported diabetes was 6.0% (95% CI: 4.5, 7.5) among men and 4.0% (95% CI: 3.1, 4.8) among women. The prevalence of undiagnosed diabetes was 2.3% (95% CI: 1.3, 3.2) among men and 1.6% (95% CI: 1.1, 2.1) among women. The proportion of both males and females with diabetes or pre-diabetes increased with increasing age so that prevalence of a glucose metabolism disorder increased from 5.9% in adults 15-24 yrs to 59.3% in adults 75+ yrs among males, and from 3.5% to 58.0% among females. Diabetes (self-reported and undiagnosed) was more common among Maori men (8.8%) and women (9.8%), and Pacific men (15.7%) and women (14.2%), compared with New Zealand European/Others men (7.6%) and women (4.5%). Prevalence of diabetes was higher among the obese (14.2%) compared with the normal weight (2.4%) group, as was prediabetes - 25.3% v 13.5%.

**Conclusion:** Both diabetes and prediabetes are very common in the New Zealand population. The current level of diabetes service provision is unlikely to meet the increasing burden of disease, given the high prevalence of glucose metabolism disorders.

**Source of Funding:** The Ministry of Health, New Zealand, funded the 2008/09 NZANS.

## How much sodium are we eating? Estimates of New Zealand population sodium intake from the 2008/09 New Zealand Adult Nutrition Survey

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**Background:** Spot urinary sodium represents a practical alternative to the 'gold standard' of 24-hour urinary sodium excretion, especially in large population samples.

**Objective:** To quantify estimated population dietary sodium intake in a representative sample of New Zealanders, and compare population sodium intake with nutrient reference values.

**Design:** The 2008/09 New Zealand Adult Nutrition Survey (2008/09 NZANS) analysed sodium excretion from spot urine samples from a representative sample of 3315 adults aged 15 years and over. Results were converted into estimates of 24-hour sodium excretion using WHO formulae. These estimates are compared with dietary sodium intake estimates from 24-hour recall assessment. 24-hour population urinary sodium excretion estimates are analysed by demography, and self-reported dietary habits using regression analysis.

**Outcomes:** Mean estimated population 24-hour urinary sodium excretion was 3544 mg/day (4013 mg/day for men, and 3116 mg/day for women), which reflects a mean dietary intake of approximately 3900 mg sodium per day. Sixty five percent of New Zealanders had an estimated 24-hour sodium excretion that exceeded the recommended Upper Level of 2300 mg sodium/day (74% of men and 57% of women). There are significant differences in mean excretion by age and sex, but not by prioritised ethnicity, or level of deprivation. Mean estimated sodium intake by dietary recall assessment was 2504 mg/day (2953 mg/day for men and 2094 mg/day for women) and is likely to be an underestimate

**Conclusion:** Spot urinary sodium assessment is a useful way to estimate dietary sodium intake in large representative population samples. New Zealand's mean sodium intake is in excess of current nutrient reference values. Public health interventions to reduce population sodium intake in New Zealand are warranted.

**Source of Funding:** The Ministry of Health, New Zealand, funded the 2008/09 NZANS. Dr McLean is funded through the Health Research Council of New Zealand.

**Folate and iodine status of adults prior to fortification of bread: results from the New Zealand 2008/2009 Adult Nutrition Survey**

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**Background:** Additional folic acid consumed peri-conceptionally reduces the risk of neural tube defect-affected pregnancies. The New Zealand population also has a documented history of mild iodine deficiency. For these reasons, the New Zealand government mandated fortification of bread with folic acid, scheduled for early 2012, and iodine, introduced late 2009.

**Objective:** To assess the folate and iodine status of a nationally representative sample of New Zealand adults, prior to the introduction of mandatory fortification of bread.

**Design:** Serum and erythrocyte folate concentrations were measured in non-fasting blood samples collected from 3348 participants of the 2008/09 New Zealand Adult Nutrition Survey (2008/09 NZANS). Median urinary iodine concentration was measured in 3315 survey participants, from a spot urine sample.

**Outcomes:** In total, 1.6% (95%CI: 1.1, 2.2) of participants had serum folate concentrations <6.8 nmol/L, and 2.2% (1.4, 2.9) of participants had erythrocyte folate concentrations <317 nmol/L indicative of deficiency. Mean (95%CI) red blood cell folate concentration amongst women aged 16 to 44 yrs was 796 nmol/L (758, 834); 27.4% (23.0, 31.7) had erythrocyte folate concentrations  $\geq$  906 nmol/L. The median urinary iodine concentration of males was 55  $\mu$ g/L and of adult females was 50  $\mu$ g/L, which indicates mild iodine deficiency. There were no differences in median urinary iodine concentration by age or NZDep2006 quintile.

**Conclusion:** The prevalence of folate deficiency was low. However, more than two thirds of women of child-bearing age had folate status associated with higher risk of neural tube defect-affected pregnancies. Mild iodine deficiency was prevalent. Mandatory fortification of bread with iodine and folic acid may have public health benefits.

**Source of Funding:** The Ministry of Health, New Zealand, funded the 2008/09 NZANS.

## Food security as a predictor of body weight status: results from the 2008/2009 New Zealand Adult Nutrition Survey

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**Background:** Food security and body weight status are each growing concerns for many New Zealanders. The 2008/09 NZANS allows us to assess food security status in relation to weight status.

**Objective:** To determine household food security status within NZ and its relationship to body weight status.

**Design:** Data from the 2008/09 NZANS determined household food security status from responses to eight statements, later collapsed into three categories: fully/almost fully food secure; moderately food secure; low food security status. Body weight status was determined by BMI ( $\text{weight}/\text{height}^2$ ) and obesity ( $\text{BMI} \geq 30 \text{ kg}/\text{m}^2$ ). Using multivariate analysis, the relationship was explored between food security status and BMI/obesity, adjusted for age, sex, ethnicity, NZDep2006, education, income and household size. Survey weights were used.

**Outcomes:** For the NZ population, 59.1% were fully/almost fully food secure, 33.8% moderately food secure and 7.1% of low food security status. Compared with females of fully/almost full food security, females of moderate food secure status had a higher BMI ( $28.3 \text{ kg}/\text{m}^2$  cf  $29.4 \text{ kg}/\text{m}^2$ ,  $p < 0.05$ ) while low food secure females had a higher likelihood of obesity ( $\text{OR} = 1.87$ ). Compared with fully/almost fully food secure females, those of moderate food security had a higher BMI for NZEO ( $26.8 \text{ kg}/\text{m}^2$  cf  $27.8 \text{ kg}/\text{m}^2$ ,  $p < 0.05$ ) and Maori ( $30.3 \text{ kg}/\text{m}^2$  cf  $32.0 \text{ kg}/\text{m}^2$ ,  $p < 0.05$ ). NZEO and Pacific females of low food security were over twice as likely to be obese as the fully food secure ( $\text{OR} = 2.16$  and  $\text{OR} = 2.61$ ). Compared with Maori and Pacific males of fully/almost full food security Maori males of moderate food security ( $29.5 \text{ kg}/\text{m}^2$  cf  $31.3 \text{ kg}/\text{m}^2$ ,  $p < 0.05$ ) and Pacific males of low food security ( $30.8 \text{ kg}/\text{m}^2$  cf  $33.3 \text{ kg}/\text{m}^2$ ) had a higher BMI. Maori males of moderate food security status compared to fully/almost full secure males were over twice as likely to be obese ( $\text{OR} = 2.15$ ).

**Conclusion:** Food security is an important public health issue in NZ. Both low and moderate food security status for females of all ethnicities and among Pacific and Maori males is associated with a higher body weight status.

**Source of Funding:** Ministry of Health

## **Ergogenesis in sport: chasing the drug cheats**

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**Background:** Drug misuse in sport, with strong historical roots, rose to public prominence following the dismissal of Canadian track athlete, Ben Johnson at the 1988 Seoul Olympics. The Government of Canada invoked a national inquiry into sports drug misuse that was to redefine international anti-doping policy for sport.

**Objective:** This presentation will sketch a background to anti-doping strategies in sport, highlight some historical landmarks and reflect on current trends including “designer” drugs and the market for sports supplements. It will also consider the plight of the athlete for whom an existing medical condition may legitimately demand the use of a banned substance.

**Outcomes:** Rich rewards from success in sport have created a ready market for performance-enhancing substances. Today the World Anti-Doping Agency (WADA) represents the allegiance between international sporting federations and governmental agencies that oversees anti-doping policy for major sporting codes. The vision of WADA is “a world where all athletes compete in a doping-free sporting environment”.

## **Metabolic responses to high-fat diets rich in n-3 or n-6 long-chain polyunsaturated fatty acids in mice selected for either obesity or leanness**

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**Background:** Increasing evidence suggests that diets high in n-3 polyunsaturated fatty acids (PUFA) confer health benefits by improving insulin sensitivity and lipid metabolism.

**Objective:** The aim of the present study was to investigate the metabolic responses and insulin signalling pathways to high-fat diets rich in either n-3 or n-6 PUFA in mice with a predisposition for obesity or leanness. We hypothesised that genetic predispositions for obesity or leanness may favour specific metabolic pathways of lipid or carbohydrate metabolism.

**Design:** We examined the effect of feeding n-3 and n-6 PUFA rich high-fat diets in mice selected for either high body weight gain (DU6), obese phenotype, or for high running performance (DUhTP), lean phenotype. At 29 days of age the mice were fed standard chow (7.2% fat), or a high-fat diet rich in n-3 (27.7% fat) or n-6 PUFA (27.7% fat) for 8 weeks.

**Outcomes:** The metabolic responses to diets enriched with n-3 or n-6 PUFA differed markedly between the two selection lines. Plasma leptin and insulin were higher ( $p < 0.01$ ) in DU6 compared with DUhTP mice. The high-fat diets increased ( $p < 0.01$ ) leptin levels, body fat and metabolic parameters of adiposity in DU6 mice. In both mice lines, n-3 PUFA feeding reduced ( $p < 0.01$ ) hepatic insulin receptor beta protein concentration, suggesting decreased insulin action in the liver. In contrast, protein kinase C zeta expression increased ( $p < 0.01$ ) in abdominal fat of n-3 PUFA fed DUhTP mice, indicating enhanced insulin sensitivity in adipose tissue.

**Conclusion:** Health benefits of dietary n-3 PUFA may be explained, at least in part, by changes in insulin action and lipid metabolism. However, important genotype - diet interactions draw attention to the biological importance of genetically determined pathways that contribute to obesity.

**Source of Funding:** Ministry of Food, Agriculture and Consumer Protection Germany, German Research Foundation and Health Research Council of New Zealand.

## **Transtheoretical model mediators of fruit and vegetable intakes in the 5+YourWay study**

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**Background:** The 5+YourWay<sup>®</sup> Study is a randomized controlled trial testing the effectiveness of Transtheoretical Model-based, computer-tailored communications among a nationally representative sample of low fruit and vegetable consumers ages 25 to 60 years.

**Objective:** This study investigates whether there is a mediating relationship between decisional balance and self-efficacy and fruit and vegetable intakes in The 5+YourWay Study.

**Design:** Validated instruments measured fruit and vegetable intakes, decisional balance and self-efficacy. Screening survey data (n=2132) was used to explore, confirm and validate separate fruit scales and vegetable scales. Multivariate analysis of variance assessed changes in intake across the 12-month study in tailored, generic and control groups (n=384-6). Mediation was tested using structural equation models, the Baron and Kenny causal steps methods and the Sobel method.

**Outcomes:** In the tailored group, 0.6 and 0.5 daily serving increases in fruit intake and vegetable intake at three months were maintained at 12 months. Pros and cons did not mediate the effect the intervention had on intake. Fruit self-efficacy accounted for 15% and 31% of the intervention effect on intake at three and six months. "Main meal" self-efficacy accounted for 13% of the intervention effect on vegetable intake at three months, while "other occasion" self-efficacy accounted for 24% and 45% of the intervention effect on vegetable intake at six and 12 months.

**Conclusion:** 5+YourWay-generated tailored communications significantly increased fruit and vegetable intakes in this sample. Self-efficacy partially mediated increases in intake, so it should be targeted in future interventions. Pros and cons had no mediating effect, although these subscales had marginal validity.

**Source of Funding:** This research was funded by a grant from the Heart Foundation of New Zealand with support from the University of Otago.

### **Out of the box: factors preventing households from eating more fruit and vegetables**

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**Background:** Low-income families tend to eat less healthy diets than families with higher incomes. However, cost is only one of a number of resources that can influence whether a household eats healthily or not. Only New Zealand data from this multi-country study is presented.

**Objective:** To explore the breadth of resources necessary for households to eat more fruit and vegetables and identify strategies for resource distribution.

**Design:** An adapted ethnographic approach of qualitative research was used to observe twenty households in their natural setting for three months. An even number of low- and high-income households representing a range of family types were recruited from across New Zealand. Each household received a free box of fresh fruit and vegetables each week, delivered to their home, and were home-visited on two occasions each week by a researcher. Observations, discussions and interventions were documented using field notes and digital technology. Expanded field notes were coded manually to identify themes.

**Outcomes:** Preliminary analysis suggests human, social, natural, financial and physical resources all play a role in food choice decisions. Households with a greater range of resources, especially human and social resources, appear to make better use of the box contents than those with a narrower range of resources. All households require motivation, basic knowledge and skills, and confidence to overcome barriers to make use of the free fruit and vegetables. These human resources seem to develop over time through social learning experiences beginning in childhood.

**Conclusion:** These preliminary findings suggest deficits in a range of resources, not simply financial, contribute to the social gradient in healthy eating. Ongoing analysis will reveal household-tailored strategies needed to make necessary resources available.

**Source of Funding:** The 5+ A Day Charitable Trust, vegetables.co.nz, and the University of Otago funded this research.

## **Kiwifruit proteases enhance digestion of common protein-based foods under simulated gastric and small intestinal conditions**

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**Background:** Kiwifruit consumption has long been thought to assist in the digestion of foods by hydrolysing proteins more completely and more rapidly than mammalian digestive enzymes alone. However there is little published evidence for this.

**Objective:** To investigate the effect of kiwifruit proteases (actinidin) on the digestion of a range of common protein-based foods under *in vitro* gastric and small intestinal conditions.

**Design:** Protein foods including meat and fish muscles, eggs, tofu, cottage cheese, and yoghurt were incubated in the presence or absence of Hayward kiwifruit extract using an *in vitro* digestion model (incubation with pepsin followed by pancreatin, simulating gastric and small intestinal digestion in humans). The digests were subjected to gel electrophoresis (SDS-PAGE) to assess loss of intact protein and the appearance of peptides (>3 kDa) during digestion.

**Outcomes:** Kiwifruit extract improved the digestion of the proteins with molecular weights above 30 kDa,  $\beta$ -casein and  $\beta$ -lactoglobulin in yoghurt; and tropomyosin- $\beta$  chain, troponin T, tropomyosin- $\alpha$  chain in chicken muscles during the simulated gastric digestion phase. Similarly, the digestion of other food proteins was also enhanced under both gastric and small intestinal digestion conditions. In particular, enhanced digestion of hoki, tuna and cottage cheese was observed in the simulated gastric digestion system; and enhanced digestion of chicken, cottage cheese and to some extent yoghurt was observed in the simulated small intestinal system. Kiwifruit extract alone (in the absence of other digestive enzymes) resulted in greater solubilization of the proteins and was capable of digesting some proteins present in foods, particularly yoghurt, cheese, fish and raw eggs.

**Conclusion:** This study provides clear evidence that Hayward kiwifruit can increase the extent of the digestion of food proteins in an *in vitro* digestion system and lends support to a role for dietary kiwifruit as a digestive aid.

**Source of Funding:** Supported by grants from ZESPRI (New Zealand) and Ministry of Science and Innovation (New Zealand) under Future Foods research program.

## Have we changed? National adult nutrition surveys from 1997 to 2008/09

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**Background:** The Ministry of Health has funded two national nutrition surveys in New Zealanders aged 15 years and over; the 1997 National Nutrition Survey (NNS) and the 2008/09 New Zealand Adult Nutrition Survey (NZANS).

**Objective:** To monitor the food and nutrient intake and nutritional status of the New Zealand adult population over time.

**Design:** The 2008/09 NZANS consisted of a 24-hour diet recall and questionnaires, anthropometric and biochemical measurements. Indicators that were considered comparable across the 1997 NNS and the 2008/09 NZANS were compared by gender, using the same definitions and cut-offs. A significant difference was determined by non-overlapping, 95% confidence intervals.

**Outcomes:** From 1997 to 2008/09, the following changes were statistically significant for males and females. Reported nutrient intakes showed a decline in energy intake (males only); a decline in the proportion of energy from saturated fat; an increase in the proportion of energy from protein; a decline in median vitamin A, zinc and potassium intake; and an increase in median vitamin B<sub>6</sub> and selenium intake. There was an increase in the prevalence of obesity; a decrease in mean total blood cholesterol; an increase in mean HDL cholesterol; and an increase in the prevalence of iron deficiency (females only). The proportion of households classified as having low food security increased. There was an increase in the proportion of New Zealanders who reported eating the recommended number of servings of fruit per day.

**Conclusion:** There have been some positive nutrition-related changes in the New Zealand adult population between 1997 and 2008/09 however the surveys also identified areas of concern and areas for further analysis.

**Source of Funding:** Ministry of Health

### **Microbial saccharolytic enzymes, dietary fibre, and gastrointestinal health**

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**Background:** Dietary fibre-enriched foods confer health benefits to the consumer due to their resistance to digestion, and the resulting stimulation of anaerobic microbial fermentation of the undigested carbohydrate. This fermentation results in the production of short chain fatty acids (SCFA: acetic, butyric, lactic and propionic acids) that play an important role in gastrointestinal health.

**Objective:** To measure changes in microbial saccharolytic degradation enzyme activities and SCFA concentrations over time in the caecum of rats fed cellulose, inulin or resistant starch.

**Design:** Male Sprague Dawley rats (3 weeks of age) were fed diets containing 2.5% cellulose, 5% cellulose, 5% inulin, or 5% maize resistant starch. The rats (n=8) were euthanized at 0, 24, 48, 72, 144, and 168 hours after consuming the experimental diets and caecum digesta removed for microbial saccharolytic enzyme activity and SCFA analyses.

**Outcomes:** There was a significant effect of diet on microbial saccharolytic enzyme activity and SCFA content of the caecum digesta. Rats fed the inulin diet had significantly greater ( $p < 0.05$ )  $\alpha$ -arabinopyranoside,  $\alpha$ -galactopyranoside, and  $\beta$ -glucopyranoside activity than the rats fed the cellulose and resistant starch containing diets. There was no effect of time on most of the carbohydrate enzyme activities. Short chain fatty acid concentrations were significantly different between the diets. Lactic and succinic acids were higher ( $p < 0.05$ ) and isobutyric lower in the caecum digesta of the inulin fed rats. Formic, propionic and butyric acids were significantly ( $p < 0.05$ ) higher for the rats fed the inulin and resistant starch diets compared to the cellulose diets. There were some changes in SCFA concentrations over time.

**Conclusion:** Short chain fatty acid concentrations and microbial enzyme activities were significantly different for the rats fed inulin, a well known prebiotic. These measures may be useful experimental biomarkers in future studies evaluating prebiotic effects in new foods or food components.

**Source of Funding:** This study was funded by the New Zealand Foundation for Research, Science and Technology (C02X0703).

## **Energy and macronutrient intake among those susceptible or resistant to obesity**

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**Background:** Despite living in an obesogenic environment not all individuals become overweight/obese; some remain lean with relative ease. Others report having to consume smaller amounts of food to maintain a healthy body weight. Information from the former group may allow us to develop novel strategies to benefit those who continually struggle with weight management.

**Objective:** To examine the energy and macronutrient intake of exponders (self-defined as 'eating as much as they wish without weight gain' versus conservers (self-defined as 'eating small amounts in order to manage their weight').

**Design:** Exponders (n=32) and conservers (n=25) completed a four day weighed food record. Nutrient composition was calculated using nutrient analysis software and New Zealand food composition data. Body composition was measured using dual-energy x-ray absorptiometry (DXA). Participants also completed the Three Factor Eating Questionnaire (3FEQ).

**Outcomes:** When expressed relative to body weight and lean body mass (LBM) respectively, conservers reportedly consumed significantly less total energy ( $p<0.001$ ,  $p<0.001$ ), protein ( $p<0.001$ ,  $p=0.033$ ), fat ( $p=0.001$ ,  $p=0.002$ ), and carbohydrate ( $p<0.001$ ,  $p=0.001$ ). This observation is in agreement with the conservers' self-reported need to consume small amounts of food to maintain a healthy body weight. Dietary restraint ( $p<0.001$ ) and disinhibition ( $p<0.001$ ) were significantly higher among conservers. Food variety and eating frequency will be explored.

**Conclusion:** Despite having similar LBM and a significantly higher body fat, conservers still reported consuming less total energy. The lower than predicted energy intake in conservers may be due to prolonged periods of dietary restraint followed by episodes of potentially obesogenic disinhibition. Managing energy intake by promoting intuitive eating and maintaining satiation may aid these individuals in achieving weight control.

**Source of Funding:** Supported by a University of Otago Research Grant

## **Changes in dietary habits in men after consultation based on the nine-step New Zealand Heart Foundation guidelines**

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**Background:** Dietary modifications and changes in dietary habits are essential in reducing cardiovascular disease risk and have been shown to improve dyslipidaemia.

**Objective:** To investigate changes in dietary habits following consultations using the New Zealand Heart Foundation nine-step heart-healthy eating guide in hypercholesterolaemic men participating in a fruit intervention study.

**Design:** Hypercholesterolaemic men (n=79) participated in an eight week randomised cross-over fruit intervention study requiring a four week healthy diet run-in period. Fasting plasma lipids and % body fat (BodPod) were assessed at baseline (B1) and after 4 weeks (B2 – before starting the fruit intervention). Dietary intake was assessed using estimated food records, followed by individualised nutrition consultations aimed at achieving the nine heart-healthy guidelines. During the last visit the men self-assessed their achievement of each guideline using a questionnaire and a qualitative interview.

**Outcomes:** Lipid profiles and % body fat improved significantly between B1 and B2 (plasma total cholesterol:high-density lipoprotein cholesterol ratio:  $-0.29$  (SD:  $\pm 0.52$ ) mmol/L ( $p < 0.001$ ); % body fat:  $-0.62$  (SD:  $\pm 0.41$ ) % ( $p = 0.001$ )). The mean (SD) percentage of energy from saturated fat and the median (25, 75 percentile) dietary cholesterol decreased from  $12.8 \pm 3.66$  to  $9.97 \pm 2.67$ % and  $327$  (225-464) to  $237$  (182-337) mg/day, respectively; fibre and vitamin C intakes increased from  $25.6$  (21.1-32.9) to  $27.9$  (23.4-31.8) g/day and  $111$  (78.5-164) to  $202$  (171-256) mg/day respectively. In terms of the men's perceptions in meeting the guidelines, best achieved were fruit and vegetable guidelines improving in 43.1% of the men, takeaway guidelines in 40.4% men, milk and spreads/nuts guidelines in 30.4% men each. Many men (60%) ate high fat / sugar / salt snacks at B1, however improvements in choices of snacks (fruit, vegetable, dairy, nuts) were apparent in the following dietary evaluations.

**Conclusion:** Focusing on these identified strategies would assist men to enhance adherence to heart healthy eating and to achieve dietary goals to improve heart health. It may also improve the risk factors for heart disease over the short term.

**Source of Funding:** None.

### Green kiwifruit: effects on plasma lipids and APOE interactions

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**Background:** Diet is a crucial element in the reduction of risk of cardiovascular disease (CVD). Furthermore, response to dietary change may be influenced by genotype. Kiwifruit are a good source of several dietary components shown to improve dyslipidaemia and lower CVD incidence such as soluble fibre and some vitamins and phytochemicals.

**Objective:** To investigate the effect of consuming two green kiwifruit daily in conjunction with a healthy diet on plasma lipids and examine response according to apolipoprotein E (*APOE*) genotype in hypercholesterolaemic men.

**Design:** Eighty-five hypercholesterolaemic men (low-density lipoprotein cholesterol (LDL-C) >3.0 mmol/L and triglycerides (TG) <3 mmol/L) completed an eight week randomised controlled cross-over study, after undergoing a four week healthy diet phase. The study consisted of two 4-week treatment sequences of 2 green kiwifruit/day plus healthy diet (intervention) or healthy diet alone (control). Fasting blood samples were taken at baseline, 4 and 8 weeks for the measurement of plasma lipids (total cholesterol (TC), LDL-C, TG, high-density lipoprotein cholesterol (HDL-C)), serum apolipoproteins A1 and B (apoA1 and apoB).

**Outcomes:** After the kiwifruit intervention plasma HDL-C concentrations were significantly higher (mean difference 0.04 [95% CI: 0.01, 0.07] mmol/L [p=0.004]) and the TC/HDL ratio was significantly lower (0.15 [-0.24, -0.05] mmol/L [p=0.002]), compared to control. In carriers of *APOE4* allele TG concentrations were significantly lower (0.18 [-0.34, -0.02] mmol/L [p=0.03]) after the kiwifruit intervention compared to control. There were no significant differences between the two treatments for plasma TC, TG, LDL-C and serum apoA1 or apoB.

**Conclusion:** The small but significant increase in HDL-C and decrease in TC/HDL ratio and TG (in *APOE4* carriers) suggests that the regular inclusion of green kiwifruit as part of a healthy diet may be beneficial in improving the lipid profiles of men with high cholesterol.

**Source of Funding:** ZESPRI® International

**Trial No:** ACTRN12610000213044

## **Do we need to correct for endogenous material when determining fibre fermentation in the gastrointestinal tract?**

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**Background:** Dietary fibre is a heterogeneous material and depending on its composition can vary in terms of its fermentability in the gastrointestinal tract. In addition, negative ileal fermentability values have been reported for fibre, but these negative values remain unexplained.

**Objective:** Determine the ileal and faecal fermentability of dietary fibre in kiwifruit using the pig as a model for the human.

**Design:** Ileal T-cannulae were surgically implanted into seven pigs (46 kg live weight). The pigs were first fed with a fibre-free semi-synthetic diet for four days followed for 14 days by a semi-synthetic diet containing freshly peeled kiwifruit as the sole fibre source (13% kiwifruit dry matter in the diet). Titanium dioxide was included in all diets as an indigestible marker. Ileal digesta and faecal samples were collected on day 14 for determining the apparent fermentability of soluble (SDF), insoluble (IDF) and total dietary fibre (TDF) (Prosky method).

**Outcomes:** The apparent ileal and faecal SDF, IDF and TDF fermentability of the kiwifruit were 25%, -8% and 0.2% and 88%, 21% and 38%, respectively. For the pigs fed the fibre-free diet, the ileal digesta and faeces contained endogenous material that was determined as SDF, IDF and TDF based on the Prosky method (for ileal 4.0, 2.9 and 6.9 and for faeces 1, 13 and 14 g/kg DM intake, respectively). When the apparent fermentability values were corrected for this interfering endogenous material the resulting values at the ileum were 82%, 5% and 24% and over the total tract were 100%, 85% and 91% for SDF, IDF and TDF. These values were significantly ( $p < 0.05$ ) higher than their corresponding apparent values.

**Conclusion:** Apparent fibre fermentability values based on the Prosky method may underestimate the amount of fibre fermentation that occurs in the gastrointestinal tract. Therefore, apparent fermentability values should be corrected for the endogenous material present in the gastrointestinal tract that interferes with the fibre determination. Further work should focus on characterising this endogenous material.

**Source of Funding:** Supported by Zespri International Ltd.

**Characterisation of kiwifruit (*Actinidia deliciosa* var *Hayward*) fibre digestion**

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**Background:** The high fibre content of kiwifruit suggests that it could contribute to the health of the gastrointestinal tract.

**Objective:** To determine the digestibility of kiwifruit fibre throughout the gastrointestinal tract using the growing pig as a model for the adult human.

**Design:** Semi-synthetic corn starch-based diets containing freeze-dried kiwifruit as the fibre source were fed to growing pigs. Digesta from the stomach, jejunum, ileum and faeces were analysed for fibre content after fractionation. Apparent digestibility of the fibre components was calculated for each gut section.

**Outcomes:** Apparent soluble dietary fibre digestibility of freeze dried kiwifruit increased significantly ( $p < 0.001$ ) as soluble fibre transited the gastrointestinal tract with little or no digestion occurring in the stomach to 37% digestibility of soluble fibre at the ileal level and 87% digestibility at the faecal level. Insoluble fibre digestibility also significantly ( $p < 0.001$ ) increased throughout the gut but to a much lower level (faecal digestibility = 26%). At the end of the tract more than 90% of the gut soluble fibre, hot water soluble fibre and oxalate soluble fibre and 75% of the hemicelluloses and 60% of the cellulose were digested. The gut soluble and oxalate soluble fibre fractions were primarily digested in the small intestine, the hot water soluble fibre was evenly digested between the small and large intestine, hemicelluloses were primarily digested in the small intestine and cellulose was digested along the length of the tract. Inclusion of dietary kiwifruit increased the water holding capacity and faecal bulking capacity of the digesta.

**Conclusion:** Digestion of the fibre fractions in kiwifruit occurred along the full length of the intestines with 80% of the total fibre digested by the end of the gastrointestinal tract. Kiwifruit possess properties that characterise a fibre source that may confer health benefits through its actions in the gastrointestinal tract.

**Source of Funding:** ZESPRI™ International Ltd.

## **Effects of food structure on nutritionally distinct carbohydrate fractions in cereal**

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**Background:** Food structure has an important influence on the rate at which foods are digested.

**Objective:** To examine the effect of cereal food structure, primary and secondary, on distinct carbohydrate fractions generated by digestion and studied microscopically.

**Design:** A range of starchy foods, including bakery products, breakfast cereals, pastas, and pulses, were digested *in vitro* and the time course of pancreatic digestion was followed for 180 min to measure rapidly-available carbohydrate (RAC) and slowly-digested starch (SDS). The digests were then homogenised and further digested to measure inaccessible digestible starch (IDS). Samples were also removed for light microscopy.

**Outcomes:** The digestion profile of the foods differed with food types. Bakery products and processed breakfast cereals disintegrated rapidly in digestion and yielded large amounts of RAC, less SDS and little IDS. Pastas, which were dense and homogeneous, were more gradually digested to completion by superficial erosion, yielding approximately equal proportions of RAC and SDS but little IDS. Pulse particles, which consisted of cotyledon fragments with robust cell walls, yielded a low proportion of RAC, a large proportion of SDS and more IDS than other foods, and their digestion profile was more linear than that of the cereal flour-based foods.

**Conclusion:** The preservation of native “primary” structure, and the use of processing to create “secondary” structure, are both means by which wholeness, in the sense of intactness, can be used to influence carbohydrate digestion to make foods of lower glycaemic impact.

**Source of Funding:** Oxford Brookes University, UK and New Zealand Institute of Crop and Food Fellowship.

## The relationship between physicochemical characteristics of mango and antioxidant activities of mango kernels

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**Background:** Materials discarded during fruit processing are often rich sources of antioxidants. The maturity of the fruit influences the total phenolic content and antioxidant capacity of the flesh itself and presumably other components of the fruit such as kernels.

**Objective:** To determine the relationship between the physicochemical parameters of mangoes and the total phenolic content (TPC) and total antioxidant capacity (TAC) of the kernels. To investigate the relationships between results of different antioxidant assays of kernel.

**Design:** The maturity of fresh mangoes (*Mangifera indica* cv "Tommy Atkins" n=12) was assessed by measuring colour, maturity score, firmness, total soluble solids (TSS), titratable acidity (TA), TSS/TA ratio, Vitamin C and moisture content. The total phenolic content of the freeze dried kernel was determined using Folin Ciocalteu reagent and the antioxidant capacity by ABTS, DPPH, FRAP, Hydrophilic ORAC (H-ORAC) and Lipophilic ORAC (L-ORAC) assays. Pearson correlation analysis and principal component analysis were performed using Minitab 16.

**Outcomes:** Total phenolics content of mango kernel was 139 mg gallic acid equivalents/g of dry matter. Total antioxidant capacity as determined by ABTS, DPPH, FRAP, H-ORAC and L-ORAC were 2228, 2206, 855, 907 and 13  $\mu\text{mol}$  trolox equivalents/g of dry matter, respectively. Maturity score, total soluble solids and TSS/TA of fresh mango showed significant moderate correlations with TPC and TAC of freeze dried kernels. With the exception of L-ORAC, there were significant strong correlations ( $p < 0.001$ ) between all the antioxidant assays used including TPC.

**Conclusion:** Maturity score, TSS and TSS/TA of mango fruit are good indicators of antioxidant activities of mango kernels. The total phenolic content was highly correlated with antioxidant activities, which suggests that the phenolic compounds of mango kernels contribute significantly to the antioxidant activity measured. The highly significant correlations between the results of antioxidant activity assays suggest one assay (e.g. TPC assay) could be chosen to conveniently monitor antioxidant capacity of mango kernels.

**Source of Funding:** None.

## **New Zealand Green-lipped mussels enhance iron absorption in Caco-2 cells and mouse proximal small intestine**

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**Background:** Iron bioavailability can be manipulated by the nutritional composition of a meal. Red meat has been repeatedly reported to significantly improve iron absorption, however because red meat contains a high concentration of saturated fatty acids, promoting red meat consumption in order to improve iron status at a population level may be problematic. For this reason, the identification of an alternative meat/poultry/fish factor source rich in polyunsaturated fatty acids or low in saturated fatty acids is warranted. The effects of oily fish on iron uptake in both rodents and humans have been previously investigated, however protocol inconsistencies have led to contradictory results.

**Objective:** To identify whether extracts of Green-lipped mussels (GLM) enhance non-haem iron absorption compared to egg albumin using Caco-2 cell monolayers and isolated mouse small intestinal sections.

**Design:** Raw GLM homogenate and egg albumin were digested in vitro with pepsin at pH 2, and pancreatin and bile salts at pH 7. Tracer  $^{55}\text{Fe}$  combined with carrier iron in a molar ratio of 1:10 respectively was used to measure cellular iron uptake. Ascorbic acid was used as positive control and combined with the  $^{55}\text{Fe}$ /carrier iron solution in a molar ratio of 4:1 respectively. Caco-2 cell monolayers and the mucosal surface of freshly dissected mouse proximal small intestine were incubated with digestate treatments for 60 minutes. All values were standardised per  $\mu\text{g}$  of treatment.

**Outcomes:** Results are expressed as percent absorption compared to egg albumin. Ascorbic acid significantly enhanced non-haem iron absorption compared to egg albumin by ~450% in Caco-2 cell monolayers ( $p=0.001$ ) and 700% in isolated mouse small intestine ( $p<0.0001$ ). GLM digestate significantly enhanced iron absorption by ~300% compared to egg albumin digestate in both Caco-2 cells ( $p=0.01$ ) and small intestinal segments ( $p=0.02$ ).

**Conclusion:** GLM may be a healthy alternative to red meat in order to improve non-haem iron absorption. Further investigation into the mechanism of enhancement is justified.

**Source of Funding:** Institute of Food, Nutrition and Human Health, Massey University.

## **Updating nutrition compositional data for New Zealand beef and lamb to re-establish a credible scientific resource**

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**Background:** An up-to-date, comprehensive set of compositional nutrition data is one essential element of the nutritional 'toolkit'. At present, the nutrient data available on New Zealand beef and lamb is incomplete and out-of-date, possibly misrepresenting New Zealand product available worldwide. Establishing a credible technical resource, comprising a set of data for cooked and raw products, provides the fundamental platform on which all future activities relating to nutritional composition can be based.

**Objective:** To establish an up-to-date set of compositional data for New Zealand beef and lamb, to be used globally.

**Design:** Twenty three cuts of beef, including offal, and 25 cuts of lamb were selected for analysis, in both the raw and cooked state. Enough samples (usually 10) to yield 3kg for each analysis were collected from different parts of New Zealand over a 6-month period. Sample preparation and analysis methods were chosen based on a literature review and the requirements of international databases.

**Outcomes:** Twenty five nutrients and 40 fatty acids were analysed on both raw and cooked product. Basic proximate analysis, yielding protein, water, fat and ash, enabled the calculation of energy content. Additional data in this updated set includes figures for LCPUFAs and vitamin D. Lean tissue and fat were analysed separately, allowing calculation of cut composition for a variety of trim levels, according to market.

**Conclusion:** This analysis provides a set of up-to-date, comprehensive data for all user groups, including industry, nutrition scientists, health professionals and consumers. Its use helps to ensure nutrition information is accurate, current and science-based.

**Source of Funding:** Funding by Beef + Lamb New Zealand Ltd, Wellington, from farmer levies.

## **Test-retest reproducibility of a food frequency questionnaire for New Zealand adults**

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**Background:** Food frequency questionnaires (FFQs) are a relatively simple, cost-effective method of assessing diet in population-based studies. However, there are no up-to-date, validated multi-nutrient FFQs available for use in New Zealand (NZ) adults.

**Objective:** To develop and assess the reproducibility of a multi-nutrient FFQ for use in New Zealand adults.

**Design:** A semi-quantitative 143-item FFQ was developed to assess diet over the past year. This FFQ was tested in 135 adults aged 30-59 years. As part of a wider validation study, diet records were collected for eight days (8DDR) and the FFQ was administered twice over a year. Test-retest reproducibility of the FFQ was assessed using Spearman's correlations and weighed Kappa values for energy, vitamins A, C and E, and retinol and beta-carotene.

**Outcomes:** Preliminary analysis showed that mean energy intake was 9.2MJ for the 8DDR and 9.9MJ for the FFQ on both occasions. Spearman's correlations were 0.66 for energy, 0.61 for vitamin A, 0.66 for beta-carotene, 0.69 for retinol, 0.71 for vitamin C, and 0.68 for vitamin E. Weighed Kappa values were 0.36 for energy, 0.46 for vitamin A, 0.34 for beta-carotene, 0.48 for retinol, 0.44 for vitamin C, and 0.46 for vitamin E

**Conclusion:** The FFQ showed reasonable reproducibility for the selected nutrients. Further work will investigate the effect of energy adjustment as well as modification of the nutrient database on improving reproducibility and validity of this FFQ.

**Source of Funding:** University of Otago

## **Correlates of fruit and vegetable consumption in secondary students in New Zealand**

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**Background:** Healthy eating, including daily fruit and vegetable (F&V) consumption, is important for healthy weight maintenance and prevention of chronic disease. However, only a small proportion of New Zealand adolescents consume the recommended servings of fruits and vegetables.

**Objective:** The current study investigated the social and environmental factors correlated with fruit and vegetable intake in secondary school students.

**Design:** A web-based survey of 1858 students (13-18 years) from 19 secondary schools in Otago was conducted to collect information on food consumption (using a short, validated FFQ) and attitudes, beliefs, perceptions and other factors that may influence food choices. We examined associations between F&V consumption and potential correlates using the chi-squared test.

**Outcomes:** Only 53.9% of participants ate F&V daily (50.9% of boys and 57.2% of girls,  $p < 0.001$ ) and a higher proportion of students from urban schools ate F&V daily compared to those from rural schools. A higher proportion of girls (79.4%) considered the healthiness of food important compared to boys (63.9%,  $p = 0.001$ ). The ease of getting food was important for more boys (62.6%) than girls (55.7%,  $p = 0.009$ ), as was ease of eating. A higher proportion of those who consider healthiness of food important eat fruit daily (67.3% vs. 41.6%,  $p < 0.001$ ). A higher proportion of those who consider the easiness of food to get important are less likely to eat fruit daily (56.8% vs. 64.4%,  $p < 0.001$ ). Parental encouragement to eat F&V and the parents' own F&V consumption was also associated with students' F&V consumption.

**Conclusion:** F&V consumption differed significantly by gender and school location. Both students' and parents' beliefs and attitudes are related to F&V consumption.

**Source of Funding:** Supported by grants from the University of Otago, Dunedin City Council, and Otago Regional Council. Maria Polak was supported by a studentship from the Otago Medical Research Fund.

## The effect of glycaemic load on satiety in healthy adult males

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**Background:** This study examines the feasibility of using glycaemic load as a predictor of appetite, satiety and hunger and its physiological and psychological impact. Blood glucose levels are an important determinant of food drive with low blood glucose signalling meal initiation and hunger. Low glycaemic load (GL) foods result in sustained glucose release and are thought to increase satiety.

**Objective:** To examine if individuals consuming a low GL diet will have a sustained blood glucose response and increased feelings of satiety when compared with those individuals consuming a high GL diet.

**Design:** In a randomised blinded cross-over trial, 24 healthy male individuals were asked to consume two diets, one low GL and one high GL, for a 12-hour period on two occasions at least 7 days apart. Diets were matched for macronutrient content and fibre and supplied an energy content of approximately 9000 kJ/day with 15% protein, 30% fat, 55% energy and 25 g fibre per day in line with the current New Zealand dietary recommendations. Over the test period, participants were monitored for their blood glucose response and completed subjective ratings (VAS) of satiety. Subjects also completed a 3-day diet record prior to each intervention.

**Outcomes:** The results of this study show that although blood glucose response was lower with the low GL diet (iAUC), the reduction was not significant ( $p \geq 0.05$ ) similarly visual analogue scale ratings (iAUC) showed that satiety was not significantly greater when participants were fed the low GL diet ( $p \geq 0.05$ ).

**Conclusion:** From the results of this study it is not possible to conclude that low glycaemic load diets result in greater satiety over a 12 hour period in a young male population.

**Source of Funding:** The project was funded through a Foundation for Research, Science and Technology programme, Foods for Energy Management.

### **Correlates of dieting or trying to lose weight differ between male and female Otago adolescents**

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**Background:** There is increasing pressure for adolescents to be thin and this may not always be acted upon in healthy ways, for example restriction of certain foods or by skipping meals.

**Objective:** To identify correlates of dieting or doing something to lose weight (DOTTLW) in adolescents from Otago, New Zealand.

**Design:** A web-based survey of students from school years nine and ten (mean age 14.1±0.7 years) from 19 Otago secondary schools was conducted in 2009. The survey collected information on food consumption and factors influencing this. Associations between DOTTLW and potential correlates were examined using odds ratios.

**Outcomes:** 10.2% of the 1379 participants reported that they were DOTTLW. 14.7% of girls reported that they were DOTTLW, compared with 7.0% of boys ( $p<0.001$ ). 17.0% of those who were overweight were DOTTLW compared to 8.1% of those of normal weight ( $p<0.001$ ). Girls who reported that they were DOTTLW were more likely to consider the healthiness of food that they eat as important (OR: 3.02, 95% CI: 1.34-6.80), report liking vegetables (OR: 1.47, 95%CI: 1.03-2.08) and wanting to eat vegetables everyday (OR: 1.64, 95%CI: 1.14-2.36) compared to those who were not. However, boys who reported that they were DOTTLW were less likely to report eating three meals a day (OR: 0.46, 95%CI: 0.26-0.82) and more likely to report they thought that "consuming fruit and vegetables daily makes you better looking" (OR: 2.09, 95%CI: 1.19-3.69).

**Conclusion:** These findings suggest that girls who were DOTTLW were more likely to be aware of the importance of doing so healthily. However, boys were more likely to engage in unhealthy practices. This suggests that healthy weight loss messages may need to be more widely promoted to boys.

**Source of Funding:** University of Otago, Dunedin City Council and Otago Regional Council.

## **The relationship between dietary patterns and nutrition knowledge and taste preferences in adolescents from Otago, New Zealand**

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**Background:** Only a small proportion of New Zealand adolescents consume the recommended servings of fruits and vegetables (F&V). However, limited information is available on the correlates of F&V consumption.

**Objective:** The current study aimed to identify if dietary patterns determined using principal component analysis (PCA) are associated with nutrition knowledge and taste preferences in Otago adolescents.

**Design:** A web-based survey of students from school years 9 and 10 (mean age 14.1±0.7 years) from 19 Otago schools was conducted in 2009. Information was collected on food consumption using a short, validated FFQ, and potential correlates of F&V consumption. Dietary patterns were determined using PCA. Associations between dietary patterns and nutrition knowledge and taste preferences were examined using Generalised Estimating Equations.

**Outcomes:** Two dietary patterns were identified using data from 847 boys and 660 girls. The first was a 'Treats' pattern that was associated with higher intakes of confectionery, sugary drinks, crisps and chips. The second was a 'F&V' pattern that was associated with higher intakes of fruit and vegetables. A 1SD unit increase in the 'F&V' score was associated with a 0.05 unit increase in the liking of F&V score (CI: 0.04, 0.06), a 0.17 unit increase in the liking starchy foods score (CI: 0.12, 0.21) and a 0.14 unit increase in the nutrition knowledge score (CI: 0.09, 0.19). A 1SD unit increase in the 'Treats' score was associated with a 0.15 unit increase in liking sweet foods score (CI: 0.04, 0.06) and a 0.18 unit decrease in the nutrition knowledge score (CI: -0.23, -0.13).

**Conclusion:** Interventions focusing on improving food intake in these adolescents should include nutrition education and exposure to a wide range of F&V.

**Source of Funding:** Supported by grants from the University of Otago, Dunedin City Council, and Otago Regional Council. Maria Polak was supported by a studentship from the Otago Medical Research Fund.

## **The relationship between dietary patterns and meal frequency and sleep in adolescents from Otago, New Zealand**

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**Background:** Dietary patterns have been associated with some health related outcomes such as BMI, but to date their relationship with sleep remains understudied.

**Objective:** To identify if two dietary patterns determined using principal components analysis (PCA) are associated with meal frequency and sleep duration in Otago adolescents.

**Design:** A web-based survey of students from school years nine and ten (mean age 14.1±0.7 years) from 19 Otago schools was conducted in 2009. Information was collected on food consumption (using a short, validated food frequency questionnaire) and potential correlates of fruit and vegetable (F&V) consumption. Dietary patterns were determined using PCA. Associations between dietary patterns and meal frequency and sleep duration were examined using Generalised Estimating Equations.

**Outcomes:** Dietary pattern, meal frequency and sleep duration data were available for 1346 participants. PCA produced two dietary patterns. The first "Treats" pattern was associated with higher intakes of confectionery, sugary drinks, crisps and chips. The second "F&V" pattern was associated with higher intakes of fruit and vegetables. Girls had a lower "Treats" score (-0.27) compared to boys (0.21,  $p < 0.001$ ) but there were no gender differences in "F&V" score. A SD unit increase in the "F&V" pattern score was associated with a five minute increase in average daily sleep time (CI: 2, 8), and an increased weekly frequency of consuming breakfast, lunch and evening meals. A SD unit increase in the "Treats" pattern score was associated with a four minute decrease in average daily sleep time (CI: 1, 7) and a decreased weekly frequency of consuming breakfast.

**Conclusions:** A more healthful dietary pattern was associated with more beneficial health behaviours. Interventions focusing on adolescent behaviours should focus on multiple outcomes.

**Source of Funding:** University of Otago, Dunedin City Council and Otago Regional Council.

## Encouraging increased at-home evening meal preparation: an in-home study

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**Background:** Research suggests there have been increases in both expenditure and consumption of food away from home. Food prepared away from home is generally less healthy than foods prepared at home. Increased consumption of food away from home has been associated with a higher BMI, while frequency of at-home food preparation is positively associated with nutrition profile quality.

**Objective:** To examine barriers to and facilitators of preparing evening meals at home and to test the acceptability of homemade takeaway-style meals.

**Design:** In-home interviews were conducted with the primary food provider from 10 low-income New Zealand households (four Maori, four Pacific, and two NZ European) who had indicated they bought takeaways on at least two weekday evenings in the previous week. Following the interview, one of six takeaway-style replacement meals (eg pizza) was chosen, then prepared and consumed at home. All members of the household completed a post-meal questionnaire. Interviews were analysed using thematic analysis.

**Outcomes:** Families saw takeaways as an easy, hassle-free option the whole family enjoyed. However, given the opportunity to make and taste new meals, families enjoyed their homemade takeaway-style meals at least as much as store-bought takeaways. Takeaways were often purchased as a supplement to a meal in order to make home-prepared food 'go further', reduce the cost of purchasing takeaways for the whole family, reduce preparation time and dishes, or because parents knew children would eat takeaways. Although parents were keen to try new meals and add to their set repertoire, they were concerned that if their family didn't like the meal they would have wasted time, money, food and effort.

**Conclusion:** Participants' initial perceptions of a meal do not necessarily match their opinions once they have cooked and consumed the meal, emphasising the need for an experiential component in nutrition interventions. Understanding people's actual reactions to programme elements, such as recipes, aids message tailoring.

**Source of Funding:** Health Sponsorship Council is funded by Ministry of Health.

## Spring in2it! – a workplace wellness strategy

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**Background:** Over 50% of the New Zealand adult population is overweight or obese. The workplace has been found to be an ideal environment to implement wellness programs as staff working together often have similar work environments, hours of work and conditions.

**Objective:** To assess an innovative self-monitored workplace wellness strategy, within a medium sized work environment. In particular to assess whether there can be change in risk factors for a number of lifestyle type diseases.

**Design:** Staff members were approached to participate in a 10 week wellness challenge. The challenge required people to complete a health screening and assessment prior to starting. The assessment included measures of body composition, blood pressure, blood lipids and glucose. Any significant issues were referred for medical assistance. Staffs were asked to record online their daily water intake, daily number of fruit and vegetables servings and daily minutes of exercise. At the end of 10 weeks they completed the same health screening and assessment.

**Outcomes:** A total of 84 participants completed the 10 week challenge. Over the 10 weeks the average exercise was 29 minutes per day, fruit and vegetable intake was 3 servings per day and water intake was 1 litre per day. Over this time there was a significant decrease in total cholesterol (5.37 mmol/L<sub>pre</sub>, 5.17 mmol/L<sub>post</sub> p=0.018), systolic (142 mmHg<sub>pre</sub>, 139 mmHg<sub>post</sub> p=0.024) and diastolic (86 mmHg<sub>pre</sub>, 81 mmHg<sub>post</sub> (p<0.001)) blood pressure, weight (79.3 kg<sub>pre</sub>, 78.4 kg<sub>post</sub> (p<0.001)), girth (95 cm<sub>pre</sub>, 94 cm<sub>post</sub> (p=0.008)), fat mass (27.6 kg<sub>pre</sub>, 27.05 kg<sub>post</sub> (p=0.019)) and BMI (28.6 kg/m<sup>2</sup><sub>pre</sub>, 28.1 kg/m<sup>2</sup><sub>post</sub> (p<0.001)).

**Conclusion:** These findings suggest that a workplace wellness programme can have a positive effect on markers of health status, particularly cardiovascular disease and metabolic syndrome. The levels of exercise, fruit and vegetable intake and water intake were below the recommended levels as advised by the health authorities, and suggest that any change can be positive.

**Source of Funding:** Otago Polytechnic.

### **Actinidin-containing kiwifruit extract enhances the stomach protein digestion of some dietary proteins in rats**

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**Background:** Kiwifruit (*Actinidia deliciosa* cv. Hayward) contains the protease actinidin and anecdotally kiwifruit is believed to aid digestion through the action of actinidin.

**Objective:** To study the effect of actinidin on the gastric digestion and stomach emptying rate (SER) of selected dietary proteins.

**Design:** A total of 96 Sprague-Dawley male rats were fed with semi-synthetic diets containing either beef-muscle protein, whey protein isolate (WPI), soy protein isolate (SPI), gelatin, zein or gluten protein, as the sole nitrogen source, in either the presence (+A) or absence (-A) of an actinidin-containing kiwifruit extract. Titanium dioxide was used as an indigestible marker. Rats were fed freshly prepared diets, euthanized around five hours after meal ingestion and the gastric contents collected for electrophoresis (SDS-PAGE), densitometry and titanium dioxide analysis. Additionally, SER were measured in beef-muscle protein and WPI using a magnetic resonance spectroscopy (MRS) technique. Eight adult rats were fasted overnight and received a single gavage dose of the dietary mixture (diet, AlCl<sub>3</sub> [marker detectable by MRS only in an acidic environment] and acidified water [pH 2], 7:1:9, w:w:v). The rats were immediately placed in the MRS and SER was estimated over 130 minutes from the disappearance of AlCl<sub>3</sub>.

**Outcomes:** The presence of dietary actinidin increased ( $p < 0.05$ ) the gastric digestion of beef-muscle protein, gelatin, SPI and gluten (40, 60, 27 and 29% units, respectively). In contrast, actinidin did not affect the gastric digestion of zein and WPI ( $p > 0.05$ ). The SER was greater for the actinidin-containing beef muscle-based diet (10.6% per hour, for [-A] and 18.5% for [+A],  $p = 0.001$ ). In contrast, no difference was found for the WPI-based diet (21.3% for [-A] and 24.0% for [+A]  $p > 0.05$ ).

**Conclusion:** Kiwifruit extract increases the stomach digestion and SER for some proteins possibly through the action of actinidin, which may have a positive effect on feelings of gastric over-fullness associated with high protein diets.

**Source of Funding:** Supported by Zespri International Ltd.

**Available (ATP) energy contents of two varieties of kiwifruit (*Actinidia deliciosa* var *Hayward* and *Actinidia chinensis* var *Hort16A*)**

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**Background:** Current methods for calculating the energy content of foods using estimated food energy values (Atwater factors) may not be accurate for high fibre or high protein foods due to the diversity in chemical composition and digestibility of these macronutrients. A model which combines an *in vivo-in vitro* digestibility assay and stoichiometric relationships describing nutrient catabolism has been developed to allow prediction of the available energy content of a food in terms of its ATP yield.

**Objective:** To determine the available energy of two varieties of kiwifruit using the *in vivo-in vitro* digestibility assay.

**Design:** The *in vivo-in vitro* model uses the growing pig as a model for upper gastrointestinal tract digestion in humans and ileal digesta incubated *in vitro* with human faecal inocula to simulate large intestine fermentation. Kiwifruit was fed to growing pigs and ileal digesta collected. Ileal nutrient digestibilities were determined. A sample of ileal digesta was then incubated with a human faecal inoculum and the fermentable organic matter determined.

**Outcomes:** The predicted available ATP energy contents of the Hayward and Hort16A kiwifruits were 5.9 and 6.2 kJ g<sup>-1</sup> dry matter respectively, approximately 47.2-50.8% of the determined metabolisable energy content. The available energy content of the kiwifruit expressed relative to the available energy content of dextrin (a highly digestible source of glucose) was 0.57 and 0.61 for Hayward and Hort16A respectively. Comparable ratios for metabolisable energy were 0.74 and 0.73. The energy content of kiwifruit in relation to dextrin was higher for Hayward kiwifruit compared to Hort16A when based on metabolisable energy but lower when based on available energy.

**Conclusion:** The metabolisable energy values overestimate the energy content of kiwifruit that is available to the cell. In addition, the two energy systems ranked the kiwifruit varieties differently in terms of energy supply to the body.

**Source of Funding:** ZESPRI™ International Ltd.

## **The effect of falcarinol on platelet aggregation: assay development using carrot cultivars**

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**Background:** Platelets are specialized cells that play central roles in physiological responses, including haemostasis, inflammation and wound healing. These processes involve platelet aggregation. Falcarinol is a polyacetylene that is present in carrot (*Daucus carota* L.), and known to inhibit platelet aggregation, which may reduce aggregation and maintain healthy blood flow.

**Objective:** The aim of this study was to determine the inhibitory effects of falcarinol on platelet aggregation in the presence of the agonists, adenosine diphosphate (ADP) and arachidonic acid (AA), which trigger platelet aggregation by separate pathways.

**Design:** An AA concentration of 62.5 µg/mL, an ADP concentration of 10 µM, and 80 % (v/v) platelet-rich plasma (PRP) were selected from the relevant dose response curves to give optimal platelet aggregation responses. A carrot cultivar with mean falcarinol content of 358 µg/g ± (90) dry weight was used for a preliminary human feeding trial. Healthy individuals (n = 3) received a dose of approximately 18 mg of falcarinol in 50 g of freeze-dried carrots for breakfast. Platelet aggregation induced by ADP or AA were measured using platelets obtained from the subjects before breakfast and 2 h after breakfast.

**Outcomes:** Consuming carrots caused a 40 % decrease in platelet aggregation induced by ADP and a 70 % decrease in aggregation induced by AA.

**Conclusion:** These preliminary results demonstrate inhibition of platelet aggregation across multiple pathways and support further investigation of falcarinol-rich carrot cultivars for their potential to help maintaining healthy blood flow.

**Source of Funding:** Not applicable.

## Comparison of carbohydrate composition in sweetpotato- and maize-based infant foods

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**Background:** Novel sweet potato-based complementary foods (oven-toasted, extrusion-cooked and roller-dried ComFa), which can be processed at both the household and industrial levels have been developed by the authors as an alternative to cereal-based infant foods, which are usually high in phytate, which limits the bioavailability of some nutrients. These ComFa formulations contain low levels of phytate and appreciable amounts of  $\beta$ -carotene (provitamin A) when compared with traditionally used maize-based infant food.

**Objective:** To compare the carbohydrate composition in the sweet potato-based formulations with the maize-based complementary food (Weanimix) because carbohydrate affects viscosity, and consequently the nutrient density.

**Design:** Total carbohydrate (by difference), maltose, sucrose and free D-glucose (using K-MASUG 10/04 assay kit) and free D-fructose (estimated) levels were determined in the formulations.

**Outcomes:** The total carbohydrate in the formulations were significantly different ( $p=0.0001$ ), ranging from 50 to 60 g/100 g. The levels of the maltose and sucrose in the heat-processed sweet potato-based foods were more than 65% of the total carbohydrate compared to 6.0% for the maize-based product ( $p<0.0001$ ) indicating a higher starch level in the maize-based formulation. Both free D-glucose and free D-fructose were relatively high in the ComFa formulations by a difference of more than 100% ( $p<0.0001$  and  $p=0.002$ , respectively) than in the maize-based product.

**Conclusion:** The significantly higher levels of simple sugars (maltose, sucrose and fructose) and lower starch content of the sweetpotato-based complementary foods have significant nutritional implications. Porridge prepared from sweet potato-based complementary foods would not be as viscous as from the maize-based product, so it would not require extensive dilution with water, leading to "nutrient-thinning" (i.e. nutrient density dilution). The sweet potato-based foods would be naturally sweeter than the maize-based product which is likely to result in higher food intake by infants.

**Source of Funding:** Nutricia Research Foundation (Project number: 2011-30).

## **The feasibility of baby-led weaning: formative research for a randomised controlled trial (BLISS)**

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**Background:** Baby-Led Weaning (BLW) is an alternative method of introducing solid foods to babies that is baby-led rather than parent-led and thus has the potential to reduce excessive infant weight gain.

**Objective:** To describe the characteristics and popularity of BLW, and attitudes towards it, in New Zealand in order to determine its feasibility as an alternative infant feeding option.

**Design:** Phase 1: Interviews with 20 parents who had used BLW. Phase 2: Internet survey (n=230) to assess whether parents would be prepared to try BLW. Phases 3 and 4 followed families weekly for 3 months and compared the experience of following BLW (Phase 3; n=15) with that of following BLISS – Baby-Led Introduction to SolidS – a modified version of BLW to address iron and choking (Phase 4; n=14).

**Outcomes:** Parents who had used BLW liked the convenience and family-oriented mealtimes, and viewed BLW as being healthier than current practices, although they were worried about iron. Over 30% of our survey sample had tried BLW and 51% of those who had not would be willing to try it with a subsequent child. Parents who trialled our resources to support families implementing BLISS reported that “*they helped our level of 'parental anxiety', the resources were practical, easy to follow and engaging*”. Those following BLISS offered considerably more iron-containing foods than those following BLW (2.1 vs 0.6 serves/day).

**Conclusion:** BLISS, a modified form of BLW, appears to be a viable way of introducing solids to infants and will be tested within a large randomised controlled trial commencing in 2011.

**Source of Funding:** Otago University

## **The impact of mandatory fortification of bread with iodised salt in New Zealand school children**

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**Background:** In response to the re-emergence of widespread mild iodine deficiency observed in New Zealand in the 1990s and early 2000s, the New Zealand government legislated the mandatory fortification of bread with iodised salt, effective from October 2009.

**Objective:** To evaluate the effect of the mandatory fortification of bread with iodised salt on the iodine status of schoolchildren.

**Design:** Primary schools in the cities of Dunedin and Wellington were randomly selected to participate. All children aged 8-10 years from each school were invited to take part. The following data were collected from each child: a casual urine sample for the determination of urinary iodine concentration (UIC), a 1 mL fingerprick blood sample for the determination of serum thyroxine, and general socio-demographic characteristics. In conjunction with a parent or caregiver, children were also asked about the usual consumption of foods that are typical sources of iodine in the diet, including fortified bread.

**Outcomes:** A total of 150 children took part in the study from 8 schools in Wellington (n=80) and 9 schools in Dunedin (n=70). The median UIC of the children was 113  $\mu\text{g/L}$  (25<sup>th</sup>, 75<sup>th</sup> percentile: 78, 159); 40% of children had a UIC <100  $\mu\text{g/L}$  and 12% <50  $\mu\text{g/L}$ . The mean (SD) serum thyroxine concentration was 114 (17) nmol/L; the normal reference range for children of this age group is 69-154 nmol/L. Children consumed, on average, 1.5 servings of bread made with iodised salt each day.

**Conclusion:** The mandatory fortification of bread with iodised salt has improved the iodine status of New Zealand schoolchildren, however, this improvement is relatively modest. The use of iodised salt in a wider range of manufactured foods than bread should be considered to safeguard the iodine status of school children in New Zealand.

**Source of Funding:** Funded by Department of Human Nutrition PBRF Fund.

## **Engagement of schools with the Project Energize nutrition and physical activity programme**

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**Background:** The environment provides an ideal setting for programmes to improve child health by encouraging daily moderate and vigorous physical activity and healthier eating patterns. Project Energize has been operating in Waikato (NZ) primary schools since 2005. The programme, delivered by Sport Waikato, currently includes 40,000 children, 244 schools, 27 “Energizers” and 1 dietitian. Energizers are assigned 8-12 schools each and act as a “one stop shop” to support activities that promote and coordinate improved nutrition and physical activity within schools. The programme was evaluated in early 2011.

**Objective:** To identify factors associated with the engagement of schools with the Energize programme.

**Design:** In a subsample of the 2011 Project Energize study the lead teacher in 25 of the 192 schools was interviewed. Independently, scores for engagement were derived from stocktakes of the nutrition and physical activity environment (n=192) Energizers rated the schools (n=192) and the schools were scored from the interview (n=25). Scores were compared using Spearman rank coefficients.

**Outcomes:** The strongest agreement was between the Energizer ratings and the interview score ( $\rho=0.676$ ,  $p<0.000$ ,  $n=24$ ). Higher engagement was associated with higher socioeconomic status and time in programme. Themes identified from the interviews were: perception of a school community health need, committed school leaders and effective interaction. Challenges to engagement were related to lack of parental support, fundraising with unhealthy food, transience and limited time with the Energizer.

**Conclusion:** Information obtained from the interview supported the engagement rating of the Energizers. The Energize rating should be used to determine where additional support would be most productive in reducing disparity.

**Source of Funding:** Not applicable.

## Hydration status 24-hours following dehydrating exercise

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**Background:** Exercise places the body under stress and results in the loss of water through sweat, which is not always appropriately replaced. As adequate hydration for athletes is highly important for sporting performance both inadequate and excessive rehydration can result in decreased performance and impact on health, especially as many athletes train at least once a day.

**Objective:** The two objectives of this study were, first, to investigate whether individuals adequately rehydrate, ad libitum, in the 24-hours following a dehydrating exercise session. Second, to determine which markers of hydration status are most applicable to field studies.

**Design:** A descriptive study of 25 physically active men and women (aged  $22 \pm 3$  yr) was conducted to assess the efficiency of individuals to rehydrate following a dehydrating exercise session. Body mass change and total body water (TBW) measurements were taken via bioelectrical impedance at three time points during a 24-hour period; at baseline, then, following a 50-minute session of dehydrating exercise (post-exercise) and 24 hours following baseline. Urine specific gravity (USG) was measured at baseline and the 24-hour time point. All baseline measures and 24-hour measures were taken in a fasted state. Participants completed the 50-minutes of cycle exercise (ergometer) at a self-selected pace in a climate chamber set at 30°C and 55% relative humidity.

**Outcomes:** A significant decrease in body mass was observed between pre-exercise (baseline) and post-exercise measures ( $71.5 \pm 10.5$  kg and  $70.6 \pm 10.3$  kg respectively) ( $p < 0.001$ ). By the 24-hour time point there was a significant increase in body mass ( $72.1 \pm 10.8$  kg) compared to baseline ( $p < 0.001$ ). No significant differences were observed for other measures between baseline and the 24-hour time point.

**Conclusion:** According to body mass change, USG and TBW, rehydration does occur in the 24 hours following a dehydrating exercise session when ad libitum techniques of hydration occur. Body mass is seemingly the most sensitive of these three markers used to detect a change in hydration status, as no significant differences were seen in total body water or urine specific gravity measures.

**Source of Funding:** Not applicable.

## **Vitamin D supplementation, serum 25-hydroxyvitamin D, body composition and performance in rugby union: a randomised controlled intervention trial**

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**Background:** Vitamin D may affect athletic performance through effects on muscle mass and muscle hypertrophy.

**Objective:** To determine whether vitamin D supplementation improves vitamin D status and affects body composition, and improves strength and speed performance in elite rugby union players during pre-season training.

**Design:** A randomised, double blind, placebo controlled intervention trial in 57 elite rugby union players was conducted to evaluate whether supplementation of 3,333 IU/day vitamin D for 10-12 weeks affects vitamin D status, body composition, speed and strength during pre-season training. Fasting blood samples were collected at baseline, 5-6 weeks and 10-12 weeks for measurement of serum 25-hydroxyvitamin D, parathyroid hormone and insulin growth factor-1. Standardised rugby performance tests were conducted at the same time points. At baseline and weeks 10-12, the sum of skinfolds at eight anatomical sites, plus body composition using dual X-ray absorptiometry (DXA) was determined.

**Outcomes:** Players' mean (SD) age, height, weight and BMI was 21 y (2.8 y), 185 cm (6.9 cm), 97.3 kg (11 kg), and 28.3 kg/m<sup>2</sup> (2.0), respectively. The sum of eight skinfold measurements was 78 mm (25 mm). Percent body fat estimated by DXA was 15.0% (4.7%), lean tissue mass was 78.2 kg (8.8 kg), and bone mineral density was 1.47 g/cm<sup>2</sup> (0.08 g/cm<sup>2</sup>). The sum of eight skinfolds was significantly correlated with percent body fat ( $r=0.923$ ,  $p<0.01$ ). The effects of vitamin D supplementation on body composition, on serum 25-hydroxyvitamin D concentration and on strength and speed will be reported.

**Conclusion:** The effect of vitamin D supplementation on serum 25-hydroxyvitamin D status in young men with high BMI and low body fat is unknown.

**Source of Funding:** Supported by a grant from the Department of Human Nutrition Performance Based Research Fund (PBRF), University of Otago.

## **Do sugary drinks have any role in the development of the metabolic syndrome in New Zealanders?**

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**Background:** Fructose consumption has increased throughout the world in parallel with metabolic disorders including obesity and diabetes. Important dietary sources of fructose are table sugar and sugary drinks which account for most of this increase. Observational studies show a clear association between high intakes of sugary drinks and development of the metabolic syndrome (MS) and diabetes.

**Objective:** To examine the effect of excessive sugary drink consumption on development of the MS in New Zealand.

**Design:** A review of the literature relating to the effect of high fructose intakes on the risk factors associated with the MS was undertaken and the contribution of sugar and sucrose to the diet of New Zealanders was analysed using data from the 1997 New Zealand Nutrition Survey (NNS97). A randomised trial to investigate whether exchanging sugary softdrinks for either milk, fruit juice or diet softdrink for eight weeks, in individuals who consume more than 500 mL of sugary beverages per day and have risk factors for the MS, positively influences risk factors associated with the MS will be described.

**Outcomes:** The NNS97 reported that non-alcoholic beverages, including softdrinks and fruit juice, contributed on average five percent of the total energy intake of New Zealanders. Total sugar intake was highest in overweight males aged 15-24 years who consumed on average 176 g/day. Fifty-three percent of this group consumed carbonated drinks at least three times per week. However such cross-sectional data show no clear association between metabolic risk markers and sugary drink or total sugar consumption. Preliminary data from the randomized trial will be presented to describe the metabolic characteristics of very high consumers of sugary drinks.

**Conclusion:** Some New Zealanders consume very high quantities of fructose through excessive softdrink consumption. Our randomized trial will help to determine whether or such consumption plays a causal role in the pathogenesis of the MS.

**Source of Funding:** Funded by the Riddet Institute, New Zealand.

## Nutritional optic neuropathy in Papua New Guinean prisoners

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**Background:** Ophthalmologists working in Madang, Papua New Guinea suspected a high incidence of optic neuropathy (ON) occurring in a local facility, Beon prison.

**Objective:** To determine the prevalence and severity of ON in Beon prison and to determine demographic, dietary or toxic risk factors in order to provide advice for treatment.

**Design:** All 264 prisoners detained in Beon Prison were invited to participate. Consenting prisoners were interviewed regarding demography, general and ocular health, diet and lifestyle. Participants underwent a vision and ocular examination, a physical examination, and gave a blood sample.

**Outcomes:** Of the 135 consenting prisoners, 14 had 'definite' or 'likely' ON and 30 had 'possible' ON. The prisoner diet predominantly consisted of rice, canned corned beef, canned tuna, crackers, tea and water. Dietary analysis suggested that less than 25% of prisoners met the estimated average requirement (EAR) for vitamin A, folate, vitamin C, vitamin E, potassium and calcium. Over half of the prisoners fell below the cut-offs values of deficiency for biochemical indicators of vitamin A, folate and vitamin C. A significant inverse trend between ON severity was found with both whole blood folate and red blood cell folate concentrations using linear regression when adjusted for age and time of incarceration. On average, those with 'likely' or 'definite' ON had whole blood folate concentrations 26 nmol/L lower than those without ON and had red blood cell folate concentrations 46 nmol/L lower. Exposure to alcohol, cassava, tobacco, and lead toxicity were not correlated to disease.

**Conclusion:** Folate deficiency has a likely aetiological role in the ON found in this prisoner population. The involvement of other B-vitamins, nutrient deficiencies or toxic agents may however be causal or contributing to the onset and development of ON. Vitamin A supplementation has been procured to all prisoners of Beon jail and recommendations for dietary changes in order to halt disease progression and to prevent further cases.

**Source of Funding:** Supported by the NZ Agency for International Development, the University of Otago and the Fred Hollows Foundation.

## The influence of brassicas on people with crohn's disease in a New Zealand cohort

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**Background:** Individuals with Crohn's disease have an aberrant immune function, which sometimes makes them react adversely to food items that would not normally trigger an immune response. Brassica intake contributes to immune function, possibly through release of a compound called sulforaphane during cooking or chewing. This makes it important to identify, for people with Crohn's disease, which forms of Brassica ameliorate their symptoms and which exacerbate them.

**Objective:**

1. To identify the effects of different forms of Brassicas consumed by people with Crohn's Disease as reported by New Zealand adults from the 'Genes and Diet in Inflammatory Bowel Disease' based in Auckland study.
2. To Identify if there were differences depending upon whether the Brassicas were cooked or not.
3. To investigate the possible reasons for and consequences of these differences using nutrition, clinical and genotype information.

**Design:** The consumption patterns of selected vegetables from the *Brassicale* group were identified in the adult subjects in the 'Genes and Diet in Inflammatory Bowel Disease' based in Auckland study. Symptoms associated with their consumption were identified. Nutrients that were missing and required to maintain normal homeostasis were identified through this and also through micronutrient analysis of serum samples. Key genotypes interacting with top listed nutrients and gene- nutrient interactions were ascertained.

**Outcomes:** There were significant differences in responses between individuals to some of the different species, group and forms of *Brassicales*.

**Conclusion:** One form of *Brassicale* if not tolerated can be substituted with another form and thus allow an individual to avail themselves of the nutritional benefits of *Brassicales* and thus improve their nutritional status.

**Source of Funding:** Fellowship from Nutrigenomics NZ, Auckland University.

## **Performance benefits of carbohydrate ingestion during high intensity exercise are not mimicked by mouth rinsing with a carbohydrate solution**

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**Background:** The ergogenic effects of carbohydrate (CHO) ingestion during high intensity exercise are well established. However, studies investigating performance benefits of mouth rinsing with a CHO solution are inconclusive; positive effects were reported in some studies but no benefits were observed in the fed state.

**Objective:** The present study investigated the effects of CHO mouth rinsing in comparison with CHO ingestion or fluid ingestion on time trial performance in a fasted and glycogen-reduced state to investigate whether mouth rinsing with a CHO solution has any performance benefits.

**Design:** A randomised, double-blind, counterbalanced trial with 8 moderately-trained cyclists aged 18-55 years was performed. Four trials were arranged, separated by a week, and included a 90 minute glycogen reducing exercise protocol, immediately followed by a low CHO meal and a subsequent overnight fast. The following morning a 1-hour time trial performance test was conducted. Participants performed a set amount of work as fast as possible using an electromagnetically braked cycle ergometer. The trials included CHO mouth rinse, CHO ingestion, placebo mouth rinse and placebo ingestion. Nine blood samples were taken per trial to investigate metabolic responses.

**Outcomes:** Performance time was not influenced by any treatment. However, power output was significantly increased ( $p < 0.01$ ) with CHO ingestion. Mouth rinsing with a CHO solution or the placebo fluid was without effect. Furthermore, plasma glucose and insulin levels were increased ( $p < 0.01$ ) and circulating concentrations of lactate were also elevated ( $p < 0.05$ ) in the CHO ingestion trial towards the end of the performance test.

**Conclusion:** In a fasted and glycogen-reduced state ingestion of a CHO solution during high intensity exercise enhanced performance through stimulation of insulin-mediated glucose uptake. The CHO mouth rinsing had no ergogenic effect.

**Source of Funding:** We gratefully acknowledge support from the Massey University Research Fund.

## Hydration status of male football players during training and competition

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**Background:** Acute hypohydration has frequently been observed during football and can negatively impact performance, particularly in the heat. Less well studied is hydration status over consecutive days encompassing football training and competition.

**Objective:** The aim of this study was to measure day to day hydration status, as well as acute losses, during training and competing in males participating in the New Zealand Football Championship league during the summer.

**Design:** Daily hydration status was determined by urine specific gravity (USG) from first void urine samples collected over 8 days, including 4 days of training and 1 day of competition. Acute changes in hydration status as a result of training and competition were estimated from body mass change. Sweat loss was calculated from body mass change taking into account fluid intake and urinary loss.

**Outcomes:** Mean net change in body mass during training for all players who completed training (n=12) was  $+0.40 \pm 0.75\%$  and all who completed a full game (n=7) was  $-3.02 \pm 1.08\%$ . Mean net body mass change in players who completed both training and competition (n=5) was  $-1.03 \pm 0.28\%$  and  $-3.45 \pm 0.96\%$ , respectively. Body mass change, fluid intake, sweat loss and sweat rate were greater ( $p < 0.05$ ) during competition than training in those who completed both. Players were hypohydrated (USG  $> 1.020$ ) on all mornings except competition day. Training did not affect hydration status, as indicated by USG the following morning.

**Conclusion:** Hydration status across an extended period of repeated training and competition appears to be suboptimal. Net fluid balance during training and competition is highly variable, with some players over-hydrating in training. Acute fluid loss is inevitable during football competition and is best managed by individualised drinking strategies that prevent weight gain or large loss during training and competition. Monitoring daily hydration status prior to subsequent endeavour may help achieve euhydration

**Source of Funding:** Conference funding was received from the Sports Nutrition and Exercise Metabolism Research Group Discretionary Fund (Department of Human Nutrition, Otago University).

## The effect of a high protein, high fibre diet on insulin sensitivity measured using the Dynamic Insulin Sensitivity and Secretion Test (DISST)

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**Background:** Insulin resistance is a significant factor in the development of type 2 diabetes. While there is considerable evidence to show that weight loss can improve insulin sensitivity in individuals with insulin resistance or at risk of diabetes and cardiovascular disease, the lack of a cost effective, high resolution method of assessment has meant very few studies have examined the effect of macronutrient composition on direct measures of insulin sensitivity.

**Objective:** To examine the effects on directly assessed insulin sensitivity of dietary advice including moderate increases in protein and fibre without specifying energy intake.

**Design:** Eighty-nine overweight or obese women aged 18-65 years were randomised to either a standard diet that was intended to be low in fat and relatively high in carbohydrate (n=42) or to a relatively high protein (up to 30% of energy), relatively high fibre (>30g/day) diet (HPHFib) (n=47) for 10 weeks. Advice regarding strict adherence to energy intake goals was not given. Insulin sensitivity was assessed by a novel method - the Dynamic Insulin Sensitivity and Secretion Test (DISST) which has a correlation with the gold standard glucose clamp method of 0.82 and provides additional information regarding insulin secretion with less clinical intensity and lower cost.

**Outcomes:** In contrast to the improvements in body composition, indirect insulin sensitivity indices and other metabolic risk factors in those on HPHFib, DISST insulin sensitivity was reduced by 19.3% (95% CI: 31.8, 4.5%; p=0.013) in comparison with the standard diet. Basal insulin secretion and fasting plasma glucose were reduced, and first phase insulin secretion was increased on HPHFib, though the difference between diets did not reach conventional levels of statistical significance.

**Conclusion:** An ad-libitum diet relatively high in both protein and fibre indicated reduced insulin sensitivity when measured with our novel DISST method. This was in contrast to evidence suggesting metabolic improvements on the diet. Use of the DISST method in future studies will provide new information about the effect of dietary interventions on insulin sensitivity.

**Source of Funding:** Contributions from Fonterra Co-operative Group Ltd, New Zealand Foundation of Research Sciences and Technology, Riddet Institute and Health Research Council of New Zealand.

## **Evidence-based practice or practice-based evidence?**

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**Background:** Evidence-based nutrition lifestyle interventions can improve health outcomes when implemented in the general practice setting.

**Objective:** Evidence from the five International Heelsum Workshops is reviewed to determine which European approaches may be applicable in New Zealand and Australia

**Design:** Since 2004, a move initiated by the International Heelsum workshops saw the Cochrane Diet and Nutrition Sub Field accommodated within the Primary Health Care Field of the Cochrane Collaboration. This Sub Field includes evidence from non-randomized studies, not usually included in Cochrane reviews, but which form an important part of the evidence for the role of nutrition in health outcomes. The multidisciplinary group of experts formed from the Heelsum workshops contribute to this Sub Field by their research to support the foundation of practice-based nutrition in the consulting room.

**Outcomes:** General practitioners patients' preferred source of nutrition information, require many resources to implement nutrition interventions. For their own nutrition education, nutrition updates electronically distributed are required, such as Australian 'Health Faxes' or the Cochrane Library's PEARLS. Other health practitioners, such as practice nurses, dietitians and physiotherapists can contribute to the collaborative approach. Tailoring the nutrition counselling, using 'patient centred' communication along with motivational interviewing facilitates movement through the stages of behavioural change and increases the programmes' effectiveness. With the involvement of large patient groups and limited resources, the use of an online tailoring expert system also shows promise. Here, computer tailored responses utilize the immediacy of electronic technology, analyzing patient data and queries personalizing responses and can be supported with interactive social support technology, such as chat forums.

**Conclusion:** To facilitate effective nutrition and communication in the General Practice setting, interventions based on the Heelsum Workshop findings need to be trialled in New Zealand and Australia.

**Source of Funding:** University of Auckland.

## **Assessing the prevalence of malnutrition in hospitalised children, evaluating the validity of three newly developed screening tools: a cross-sectional study in Iran**

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**Background:** Hospitalised children are often at increased risk of nutritional impairment at or during admission. Three nutritional risk screening (NRS) tools (STAMP, STRONGkids and PYMS) have been validated in Europe but have not yet been utilised in a developing country.

**Objective:** The aims of this study were to (1) define the nutritional state of children admitted to an Iranian hospital with comparison to healthy children and (2) compare and contrast the three NRS tools for hospitalised children in terms of the ease of completion and the validity of scores with comparison to current nutritional status.

**Design:** Children admitted to a tertiary paediatric teaching hospital located in Mashhad, Iran, were enrolled over a 24 day period along with healthy control children from the same community. Nutritional state was assessed by anthropometry and classified as moderate/severe malnutrition according to WHO criteria. The three NRS tools were applied to all inpatients which classified patients from low to high risk.

**Outcomes:** One hundred nineteen inpatients and 100 controls (mean ages of 4.45 ( $\pm$  3.58) and 4.75 ( $\pm$  1.68) yr) were recruited. The prevalence of moderate and severe malnutrition (under-nutrition) in the inpatient group was 17.64% and 7.56% while it was 1% and 2% respectively in the control group. In contrast, the prevalence of overweight/obesity in the control group was 22%, contrasting to 2.5% in the inpatient group ( $p=0.04$ ). NRS tools were able to identify most of the malnourished patients in the moderate to high risk groups. STRONGkids showed a better overall correlation with all anthropometric measurements.

**Conclusion:** Hospitalised children have higher rates of under-nutrition than healthy children from the same community. The three NRS tools were able to identify children at nutritional risk, but with differing utility. In this context, STRONGkids appeared to be the most useful and reliable tool.

**Source of Funding:** Not applicable.

## Changes in the sodium content of bread in Australia and New Zealand between 2007 and 2010 - implications for policy

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**Background:** Bread is the largest contributor to dietary sodium intakes in Australia and New Zealand. In 2007, the Australian Division of World Action on Salt and Health in Australia, and the Heart Foundation in New Zealand (NZ), each sought to engage industry in new programs to reduce the sodium content of bread.

**Objective:** To define the effectiveness of recent efforts to reduce salt levels in breads in Australia and New Zealand.

**Design:** Sodium data were collected from product labels for 157 packaged bread products in 2007 and 167 in 2010. Mean sodium values and proportions meeting targets were calculated overall, by bread type, and by manufacturer. Findings were compared across years and between countries.

**Outcomes:** Overall mean sodium content in bread in Australia was 434 mg/100 g in 2007 and 435 mg/100 g in 2010. Corresponding values for NZ were 469 mg/100 g and 439 mg/100 g. The proportion of Australian breads meeting the 400 mg/100 g national target increased from 29% in 2007 to 50% in 2010. The proportion of NZ breads meeting the 450 mg/100 g national target increased from 49% in 2007 to 90% in 2010. There were clear differences between the results achieved by different companies.

**Conclusion:** Voluntary efforts by non-governmental organisations have had some impact on sodium levels in bread, particularly in NZ. There remains, however, substantial room for further improvement. If additional reductions are not achieved under the current voluntary arrangements legislated approaches may be required.

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- Smith AB, Brown CD (1982) A multienzyme technique for estimating protein digestibility. *New Zealand Journal of Science* 5, 123-130.
- Smith AB, Jones EF, Brown CD (1979) Problems in the determination of soluble fibre. *Journal of Animal Science* 47, 81-85.

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