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Gut microflora, probiotics and immune function

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Background

The human gastrointestinal tract is home to dense and complex communities of microbes (over 10^{14} bacteria) representing over 800 different species. While a great majority of these bacteria are beneficial, performing numerous physiological functions vital for optimal health, some exhibit the potential to cause disease under certain conditions. Consequently, a fine balance between these health-promoting bacteria and potential pathogens is pivotal for intestinal homeostasis. A disruption of this delicate balance is associated with enhanced susceptibility to enteric infections, cancers, immunoinflammatory disorders and even the obesity. Several experimental and clinical intervention studies have shown that supplementation with probiotics could be used to modify gut microflora-host interactions and thus optimise immune system function. Probiotics are defined as live microorganisms which, when administered in adequate amount confer a health benefit on the host (1).

Review

Evidence from experimental studies has shown that gut microflora plays a critical role in the development, maturation and functioning of the immune system. Germ-free animals, born and raised under sterile conditions, have poorly developed immune systems, fail to develop tolerance to dietary antigens and exhibit increased susceptibility to microbial pathogens when compared with their conventionally reared counterparts. Introduction of microflora is able to effectively restore these deficits in immune development and function. Abundant laboratory and clinical research has also shown that specific strains of probiotics are able to optimise immune function in both health and disease. For example, studies with healthy subjects have revealed that intake of probiotics is associated with augmentation of both non-specific and specific immune responses (particularly responsiveness to vaccines), thereby enhancing resistance to intestinal and extra-intestinal infections. In subjects with aberrant immune responses (responses polarised towards TH1 or TH2), such as allergies and inflammatory bowel disease, probiotics have been shown to correct immunological imbalances through the induction of T regulatory cells. However, not all strains are created equal; significant differences in the efficacy of different probiotic strains against different conditions do exist. The efficacy also depends on the dosage employed and the viability of organisms. The precise mechanisms by which probiotics mediate their disparate immunological effects are not fully understood and are the focus of active investigation. This presentation will provide an overview of the current scientific evidence related to immunomodulatory effects of probiotics and their significance to human health, and discuss some of the opportunities and challenges that lie ahead to enable us *to* harness the immunomodulatory potential of probiotics as novel prophylactic/ therapeutic agents or dietary adjuncts.

Conclusions

A balanced microflora is pivotal for optimal health. Intake of specific strains of probiotics is effective in enhancing immune function in healthy individuals and restoring immunological homeostasis in subjects with polarised Th1 and Th2 responses. The mechanisms by which probiotics influence immune function still remain to be fully understood.

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Report of a Joint FAO/WHO Expert Consultation on 'Evaluation of Health and Nutritional Properties of Probiotics in Food Including Powder Milk with Live Lactic Acid Bacteria, Córdoba, Argentina, 2001

Probiotics prevent the development of eczema in early childhood

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Background

The role of probiotics in the prevention of allergic disease has been controversial. Early reports suggested that children with allergic disease were less likely to have lactobacilli in their faeces[1]. This was followed by a study from Finland which found that administration of Lactobacillus rhamnosus over the first six months of life halved the development of eczema in the first two years of life in children with a family history of allergic disease[2]. However subsequent studies have not always confirmed these findings. The reason for the conflicting results has not been clear. We undertook a randomised, controlled trial of supplementation with probiotics to address this question[3].

Objective

To determine the effect of supplementation with two different probiotics on the development of eczema and allergic sensitization at 2 years of age in children who were at high risk of developing allergic disease.

Design

This was a double-blind, randomized placebo controlled trial of infants who had a family history of allergic disease (with one or both parents having required treatment for eczema, allergic rhinitis and/or asthma). The study was conducted in two centres (Auckland and Wellington). Pregnant women were randomised to take *Lactobacillus rhamnosus* HN001 (6 x 10^9 colony forming units/day), *Bifidobacterium animalis* subsp *lactis* HN019 (9 x10^9 colony forming units/day) or placebo daily from 35 weeks and to continue while breastfeeding for a maximum of 6 months. Their infants (n=474) were randomized to receive the same treatment daily from birth until two years of age. The children were assessed for the presence of eczema at 3, 6, 12, 18 and 24 months of age. Eczema severity was assessed using SCORAD. Stool samples were obtained at 3, 12 and 24 months. PCR was used to identify the presence of the two organisms in the faecal samples. Allergic sensitization was assessed with skin prick testing for food and inhalant allergens and was performed at 2 years of age.

Outcomes

Infants receiving *L.rhamnosus* had a significant reduction in the cumulative prevalence of eczema from birth to two years of age (Hazard Ratio 0.51; 95% CI 0.30-0.85; p=0.01). The prevalence of eczema was 14.8% in the *L. rhamnosus* group compared with 26.8% in the control group. In contrast there was no difference in the prevalence of eczema between the placebo group and the children receiving *B. animalis* subsp *lactis* (Hazard Ratio 0.90; 95% CI 0.58-1.41; p=0.64). Neither *L. rhamnosus* (HR 0.74; 95% CI 0.46-1.18; p=0.21) or *B.animalis* (HR 0.82; 95% CI 0.52-1.28; p=0.38) had a significant effect on the prevalence of allergic sensitization. *L. rhamnosus* (71.5%) was more likely than *B. animalis* (22.6%) to be present in the faeces at 3 months but they were detected with a similar frequency at 24 months.

Conclusion

We found that supplementation with *L. rhamnosus* but not with *B. animalis* subsp *lactis* halved the cumulative prevalence of eczema at two years of age. The difference that we observed with the two probiotics may explain the conflicting results reported by studies that have used a variety of different probiotics. Although it is not clear how *L. rhamnosus* mediates its protective effect it does not appear to be through preventing the development of allergic sensitization. It is possible that the greater detection of *L. rhamnosus* at 3 months of age may contribute to its efficacy in preventing the development of eczema. We plan to follow-up the children at 4.5 and 6 years of age to determine if treatment with the probiotics has any effect on the subsequent development of asthma and allergic rhinitis.

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Impacts of dietary resistant starch and protein on colorectal health

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Population studies suggest an association between high intakes of red and processed meats and colorectal cancer (CRC) risk, while greater dietary fibre consumption, as well as other factors (e.g. exercise) appear to be associated with reduced risk. Our research efforts in the Preventative Health and Food Futures Flagships include developing a deeper understanding of how forms of dietary protein and fibre modulate biological activities associated with the initiation, progression and inhibition of CRC. Dietary fibre includes a wide spectrum of components that can differ in terms of their physicochemical properties and in their actions within the large bowel. A key difference in fibre components is their susceptibility to microbial hydrolysis and fermentation, as well as the profile of short chain fatty acids (SCFA) and

other metabolites which are produced. Of these SCFA, butyrate is considered to be particularly important for the maintenance of normal colonocyte function, apoptosis, and protection against cellular damage. Over the last 5 years, we have conducted a number of studies in humans and rats to examine the effects of different forms of dietary protein and fibre, especially resistant starches (RS), on putative biomarkers of large bowel health, including faecal butyrate concentrations. In a trial with 46 healthy volunteers we found that faecal butyrate concentrations at entry varied widely (3 to 33 mmol/kg). We also found that dietary RS was more effective than non-starch polysaccharides (NSP) in increasing the concentrations of faecal butyrate in our cohort, especially in most subjects who at entry were found to possess faecal butyrate concentrations below the "low risk" threshold for CRC. The diets had significant effects on the colonic microflora and we have shown for the first time that phylotypes related to *Ruminococcus bromii* are abundant in the large bowel of humans and that numbers increase in response to RS. Our studies conducted in rats have shown that increased levels of dietary protein as red meat, casein and soy increase colonocyte DNA damage, and also increase the production of toxic protein fermentation products. However, dietary protein as white meat (chicken) resulted in less damage, and whey protein did not elicit damage any different to basal levels. We have also shown that the inclusion of RS in diets not only raises large bowel butyrate levels in rats, but also reduces protein-induced DNA damage, irrespective of protein source. Similarly, the inclusion of RS in diets fed to rats consuming a high protein, high fat (western) diet also elicits protection against colonic DNA damage. Also, there did not appear to be adverse effects on markers of colorectal health in humans when a weight loss diet high in red meat contained fibre. In conclusion, the combination of our human and animal studies confirm that foods high in RS can raise faecal butyrate concentrations, and our animal studies provide evidence that RS is also effective in protecting against protein-induced colonic DNA damage, apparently via a mechanism involving increases in large bowel SCFA, especially butyrate.

Impacts of diet on diseases of the large intestine

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Background

There are a number of diseases and conditions of both man and animals that implicate 'diet' in their aetiology. The notion that nutrients, components and (or) ingredients of the diet can ameliorate or prevent diseases and conditions occurring is highly attractive, for a large number of reasons. It is beyond the scope of this paper to discuss these and the impacts of diet on every disease and condition recognized in the large bowel, but rather we wanted to focus on a particular dietary component/ingredient – *prebiotics* - that are implicated in large bowel structure, function and 'health' in both animals and man. Prebiotics have been defined as 'non-digestible food ingredients that beneficially affect the host by selectively stimulating the growth, and/or activity, of one or a limited number of beneficial bacteria in the colon and thus improve host health' (1). Research on the potential health benefits of prebiotics in both animals and humans has occurred over the last 15 years or so, with a recent interest in the effects on the immune system, the host's ability to fight infection, and inflammatory processes and conditions. The β2-1 fructans, which include inulin (IN) and fructo-oligosaccharides (FOS), fulfill the criteria for prebiotics (1). IN is a linear carbohydrate molecule that contains β-(2→1) fructosyl–fructose linkages with a terminal glucose and an average of 12 fructose residues. The partial enzymatic hydrolysis of IN yields a FOS, known as oligofructose (OF). In OF there can be 2-8 (average 5) fructose residues with a terminal glucose residue or a chain of 3-8 (average 5) fructose residues. As a result of intestinal fermentation and promotion of growth of (purportedly) beneficial members of the gut microbiota, prebiotics may influence host defence, immune function, inflammatory proceeses and (or) resistance to infection (2).

Objective

The objective is to briefly review empirical evidence from the literature, in addition to presenting some studies in pigs, in relation to prebiotics on diseases and conditions of the large intestine.

Design and Outcomes

Human literature (2) suggests that data conveying the health benefits of prebiotics are often difficult to compare, due to inconsistencies in methodology and the heterogeneity of the subjects used. Despite this, evidence suggests that β2-1 fructans influence some aspects of host immunity. Most studies in man have found little effect of β2-1 fructans on innate immune function, but with mixed results regarding modification on the adaptive immune system. There is also evidence that β2-1 fructans may reduce the incidence and duration of certain infections in infants and children, but supplementation in adults has not generally produced beneficial results. In animal models of inflammation, β2-1 fructans have shown benefits in models of colitis and necrotising entercolitis, e.g., one study showed that the degree of colonic aberrant crypt foci (ACF) inhibition in rats was more pronounced in animals given IN than in those fed OF (5). In pigs, recent interest in prebiotics has focused on swine dysentery (SD), a mucohaemorrhagic colitis of growing/finishing pigs caused by the anaerobic spirochaete *Brachyspira hyodysenteriae,* which can cause morbidity, mortality, and inflict significant economic losses. Danish work (4, 6) investigated disease/physiological data coupled to molecular analysis of bacterial communities in sick and healthy pigs. Feeding a fructan-rich (from chicory roots, a source of IN) and galactan-rich (from lupins) diet inhibited both the percentage of pigs showing clinical symptoms of SD and the percentage of pigs shedding the spirochaete; feeding the diet with dried chicory root and sweet lupins also reduced total numbers of the whipworm *Trichuis suis* being shed, which was co-infected with *B. hyodysenteriae*.

Conclusion

The use of specific dietary nutrients, components and (or) ingredients to ameliorate or prevent large intestinal diseases and conditions in man and animals will continue to attract attention from scientific, pharmaceutical and commercial interests. The interactions between nutrients, components and (or) ingredients and the structure and function of the large bowel to affect a range of diseases and conditions are complex, and remain a rich source of investigation and curiosity.

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Concurrent Session 4: Gastrointestinal Tract

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Gastrointestinal determinants of acute energy intake in healthy lean males

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Background

The presence of nutrients in the small intestine modulates gastrointestinal (GI) functions, eg the release of GI hormones (incl. cholecystokinin (CCK), peptide YY (PYY) and glucagon-like peptide-1 (GLP-1)) and stimulation of pyloric and suppression of antral and duodenal pressures, and suppresses appetite and energy intake.

Objective

To evaluate which of these GI functions are determinants of acute energy intake in healthy, lean men.

Design

Data from eight previously published studies involving a total of 89 healthy males were pooled together. Antropyloroduodenal pressures, plasma CCK, PYY and GLP-1, and appetite perceptions (desire-to-eat, hunger) were measured during various intraduodenal (nutrient) or intravenous (CCK and GLP-1) interventions ranging from 50 - 150 min in duration. Energy intake at a buffet lunch was quantified immediately after each of the infusions.

Outcomes

Bivariate analyses, using within-subject correlations adjusted for repeated-measures, showed that energy intake was significantly correlated with several measures of GI motility and hormone release: specifically, energy intake was related inversely to the area under the curve (AUC) of basal pyloric pressures, total number of isolated pyloric pressures (both $r<-0.24$, $P<0.001$) and AUC of CCK, PYY and GLP-1 (all $r<-0.19$, $P<0.05$), and directly to the number of duodenal pressures ($r=0.29$, $P<0.001$) and AUC of desire-to-eat and hunger profiles (both $r>0.21$, $P<0.001$). A multivariable mixed-effects model identified AUC of pre-lunch, and total number of, isolated pyloric pressures (both $P<0.05$), peak plasma CCK ($P<0.001$) and AUC of nausea profiles ($P<0.001$) as independent predictors of energy intake. In a subset of 5 studies, in which plasma PYY was measured, time to peak for plasma PYY was also significant ($P=0.01$), while GLP-1 was not a significant independent predictor of energy intake.

Conclusion

These findings suggest that specific modulations of GI motor and hormone functions contribute to the suppression of acute energy intake, which could have implications for novel approaches to the prevention and management of obesity by identifying interventions that specifically target these functions.

Gluten as a cause of gastrointestinal symptoms in patients who do not have coeliac disease

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Background

Despite increased prescription of a gluten-free diet (GFD) as a treatment for functional gastrointestinal symptoms in those who do not have coeliac disease, there is minimal evidence that gluten is a trigger.

Objective

To determine whether gluten ingestion can induce symptoms in non-coeliac individuals and to examine the mechanism.

Design

A double-blinded, randomised, placebo-controlled rechallenge trial was undertaken in patients with irritable bowel syndrome in whom coeliac disease was excluded (histology or gene typing) and who were symptomatically controlled on a GFD. Participants received gluten or placebo as two bread slices plus one muffin per day together with a GFD for six weeks. Symptoms were evaluated by a visual analogue scale and markers of intestinal inflammation/injury and immune activation were monitored.

Outcomes

34 eligible patients (29-59 yr, 4 men) were randomised. 56% had HLA-DQ2 and/or DQ8 haplotype. Adherence to GFD and supplements was 100%. The mean change after one week of therapy of overall symptoms ($P=0.05$), pain ($P=0.02$), bloating ($P=0.03$), wind ($P=0.05$), stool satisfaction ($P=0.02$) and tiredness ($P=0.001$) were higher for those consuming the gluten. Symptom severity of pain, stool satisfaction and tiredness ($P=0.02$, $P=0.03$, $P=0.001$ respectively; linear mixed effects model) were higher for gluten. Anti-gliadin antibodies were not induced. There were no changes in faecal lactoferrin, ultrasensitive CRP or intestinal permeability. There were no differences in any end-point in those with and without DQ2/DQ8.

Conclusion

'Non-coeliac gluten-intolerance' does exist, but no clues to the mechanism were elucidated. Clarification of the phenotype of such patients, the mechanisms by which gluten induce symptoms and clinical significance is required.

An investigation of unsubstantiated 'low GI' claims for Australian foods

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Background

The glycemic index (GI) concept is widely used as a mark of product differentiation by food manufacturers and consumers. In Australia, several cases of deceptive practice have been investigated by the Australian Competition and Consumer Commission (ACCC) and by Australia's consumer advocate organisation, *Choice*. Under the wider perspective of the protection of public health, a false GI value claim of a product could affect consumers that rely on that statement as part of their therapeutic management of a specific disorder. Food Standards Australia New Zealand (FSANZ) developed a new draft Standard for Nutrition, Health and Related Claims (P293), in which GI claims are treated as a hybrid nutrition content/health claim, and must include a scientifically substantiated numerical GI value that is determined using the Australian Standard methodology.

Objective

To determine the glycemic index (GI) of 10 products in the Australian market with unsubstantiated GI claims to verify the validity of such claims.

Design

Ten healthy subjects consumed 50 g available carbohydrate portions of the reference food (glucose sugar) and the 10 products. Fingerprick blood samples were collected at -5, 0 (fasting), 15, 30, 45, 60, 90 and 120 min and analysed for plasma glucose concentration. The incremental area under the plasma glucose response curve (iAUC) was used to calculate the GI value of each test food, using glucose as the reference food.

Outcomes

GI values for the products varied from 31 ± 4 to 85 ± 12. Only two out of 10 products were identified as low GI (GI ≤ 55). Eight of the 10 products were therefore found to make a false claim.

Conclusion

Many products on the Australian market make misleading and false claims around their GI. Greater surveillance and monitoring of GI claims with appropriate regulatory action is necessary for the safeguard of public health.

Microbiota and protein profiles of interleukin-10 gene-deficient mice are altered when fed diets enriched in n-3 and n-6 polyunsaturated fatty acids

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Background

Inflammatory bowel diseases (IBD) are characterised by chronic intestinal inflammation due to an abnormal immune response towards intestinal microbiota. The effect of dietary fats in relation to microbial communities, especially in relation to IBD, is not well defined.

Objective

The aim was to identify differences in the large bowel microbiota in response to diets containing polyunsaturated fatty acids (PUFA) and to correlate those with histology and protein data of interleukin-10 gene-deficient (*Il10*$^{-/-}$) vs. C57BL/6 control mice fed PUFA diets.

Design

Denaturing gradient gel electrophoresis (DGGE), qRT-PCR and cluster analyses of caecal bacterial DNA as well as histopathological, 2D-DIGE LC/MS-MS and pathway analyses of the colon were performed.

Outcomes

The DGGE profiles of *Il10*$^{-/-}$ and C57 mice fed AA or EPA diet showed differences ($P<0.05$) in the presence of DNA fragments identified as *Bacteroides vulgatus* and *E. coli* ssp. AA and EPA showed only mild reduction in colon inflammation. Bacteria-influenced proteins associated with actin cytoskeleton and tight junction signalling were more up-regulated with AA which might affect tight junction integrity and migration of bacteria.

Conclusion

These data clearly underline the necessity for a "multi-omics" approach to define the complex bacteria-host interaction networks and to identify mechanistic effects of dietary fats in colon inflammation.

The beneficial effects of a probiotic bacterium, *Propionibacterium jensenii* 702, when consumed with soy foods

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Background

Epidemiological studies have revealed a beneficial role of isoflavones in several hormone–dependent diseases and cardiovascular diseases. However, isoflavones that are naturally present in food are in non-active forms that humans cannot utilise until they are degraded by bacteria in the small intestine. Since microflora play an important part in the isoflavone bioavailability it has been suggested that introduction of probiotics can modify gastrointestinal microbiota and therefore increase the production of bioactive isoflavones namely aglycones.

Objective

To investigate the efficacy of *Propionibacterium jensenii* 702 to produce bioactive isoflavone in soymilk and to influence gastrointestinal microbiota and short chain fatty acids in humans.

Design

A randomised, parallel, placebo-controlled trial was conducted. Forty healthy subjects were randomly allocated to one of the four groups receiving: soymilk with probiotic capsule (MT group), soymilk with placebo capsule (MC), fermented soymilk with probiotic (YT), fermented soymilk without probiotic (YC). Baseline, 7 days and 28 day blood samples and faeces were collected and tested for aglycone levels, SCFA and faecal microbiota.

Outcomes

P. jensenii 702 in combination with starter culture (YT) significantly increased the concentration of aglycones in soymilk from 1.00mg to 4.35mg per100mL ($p < 0.05$). The numbers of bifidobacteria increased significantly in YT group and levels of SCFA also increased significantly when compared with other groups. *P. jensenii* 702 was only detected in the treatment groups (YT and MT) with significantly increased recovery when consumed in fermented state (YT) than as a capsule. Serum aglycone levels increased significantly after 7 days of treatment across all study groups.

Conclusion

The fermented soymilk with the probiotic had beneficial effects on the microecology of the colon of healthy adults by increasing the concentration of short chain fatty acids and increasing the numbers of bifidobacteria. Also addition of combination cultures of YC380 and *P. jensenii* 702 to soymilk increased isoflavone aglycone content and produced equol levels delivering expanded nutritional quality of soymilk.

Addition of dietary enzymes and sorghum protein digestibility

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Background

Composition and availability of nutrients in sorghum grains are highly variable, especially protein content and digestibility. Chickens fed on sorghum based diets often under perform and this may reflect low nutrient availability due to the presence in sorghum of polyphenols, phytate and kafirin which are antinutritional factors.

Objective

To assess the effects of antinutritional factors on the ileal protein digestibility of sorghum with different dietary enzymes

Design

Eight mash diets were prepared with sorghum (918g sorghum/kg diet) as the sole protein source. Celite (20 g/kg) was added to allow acid insoluable ash (AIA) to be used as an indigestible marker. Diet one was a control and the other diets had either: xylanase; phytase; protease; xylanase+phytase; xylanase+protease; phytase+protease or xylanase+phytase+protease added. Broiler chicks, 36-days-old, were housed (7birds/cage) in an environmentally controlled shed and randomly assigned to replicated (n=4) dietary treatments with free access to feed and water. On day-42, birds were euthanized and contents of the lower half of the ileum were pooled/pen, frozen and lyophilized. Nitrogen and AIA content of all feed and ileal samples were determined using standard laboratory protocols and protein digestibility coefficients were calculated.

Outcomes

The ileal protein digestibility coefficient of the control diet was 0.768±0.011 and this was significantly ($P<0.05$) improved by protease (5.0%), phytase (4.4%) and the phytase+ protease combination (5.8%) Xylanase alone had no influence on digestibility but in combination with phytase or protease numerically improved ileal protein digestibility by 3.3 and 2.2% respectively. The diet with the three enzyme combination had no significant effect (0.784±0.01)

Conclusion

These findings demonstrate that strategic application of enzymes to sorghum based poultry diets can reduce the negative influence of phytate and kafirin on protein digestibility.

Characteristics of fermented goat's milk containing probiotic bacteria

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Background
Goat's milk and goat's milk products are attractive to some consumers due to their special nutritional and therapeutic properties. Yogurt is a popular fermented dairy product that is widely consumed throughout the world. Due to potential health benefits probiotic bacteria such as Lactobacilli and Bifidobacteria have been increasingly incorporated in fermented dairy products. Incorporation of newly identified probiotic *Propionibacterium jensenii* 702 (PJ 702) in developing fermented goat's milk products may result in interesting outcomes due to their already demonstrated health promoting, nutritional and sensory advantages.

Objective
This study was designed to establish the suitability of fermented goat's milk as a delivery vehicle for probiotic bacteria PJ 702 with or without *Lactobacillus acidophilus* (LA) and *Bifidobacterium lactis* BB 12 (BB 12) and to evaluate the microbial, physico-chemical and sensory characteristics of the product.

Design
Fermented goat's milk was produced with PJ 702 and following probiotic bacterial combinations: PJ 702 x BB 12, PJ 702 x LA, PJ 702 x LA x BB 12. Products were assessed for viability of probiotics at refrigerated storage (4 ^{o}C) over 21 days using spread plate techniques in triplicate. Physico-chemical properties of products were measured according to standard methods. Sensory properties were also evaluated.

Outcomes
Viable counts of PJ 702 in all products were above 10^{-6} cfu/ml at the end of storage and were able to maintain suggested therapeutic minimum at the time of consumption. Titratable acidity was increased while pH was decreased in all products over the shelf life. The final titratable acidity of products fell within 0.33±0.00 – 0.63±0.01%. Other physico-chemical properties were not demonstrated significant changes (P>0.05) throughout the storage except syneresis. The products received consumer acceptance according to a nine point hedonic scale used for the sensory evaluation.

Conclusion
This study contributes to a better understanding of successful application of newly identified probiotic PJ 702 in fermented goat's milk production in addition to common lactic acid bacteria such as LA and BB 12.

The effect of dietary vegetable and fruit fibres on gut health in healthy rats

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Background
Dietary compounds remaining undigested at the end of the small intestine become substrates for fermentation by the colonic microflora. The main fermentation products of dietary fibre, short chain fatty acids, acetic, propionic and butyric, are rapidly absorbed in the colonic mucosa and have positive physiological effects on gut health.

Objective
To measure the effect of dietary fibre from apple and broccoli on gut health parameters in healthy rats.

Design
Sixty-four male Sprague Dawley rats (6 weeks of age) were fed eight experimental dietary treatments (16 rats per treatment) for 6 weeks. The dietary treatments were control fibre (2.5% cellulose), control + pectin (2.5% cellulose + 2.5% pectin), broccoli fibre (5%) and apple fibre (5%). The apple and broccoli fibres were prepared from locally sourced fresh produce.

Outcomes
There was a significant effect of dietary fibre (*P*=0.055) on colon crypt depth. The crypt depths were greatest for the rats fed the apple fibre and least for the control fibre diet fed rats. Colon goblet cell numbers were significantly (*P*=0.014) increased in the rats fed the broccoli and apple fibre diets. There was no significant effect of diet on *Bifidobacterium* spp copy numbers.

Conclusion
There were positive gut health benefits when the rats were fed broccoli and apple fibre.

Effect of two diets with varying insulin demand on day-long profiles of blood glucose and insulin concentrations

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Background
A food insulin index (FII) based on testing iso-energetic portions of single foods has been shown to be useful in predicting the dietary insulin demand evoked by composite meals. However the utility of FII has not been validated in predicting the day-long insulin profiles.

Objectives

To investigate the effect of two diets with varying insulin demand as predicted by FII of the component foods on day-long blood concentrations of glucose and insulin.

Design

Ten lean, healthy subjects consumed a high and a low FII diet in a randomized, crossover design, consisting of three consecutive meals over an 8 h period. Two diets were matched for macronutrients, fiber and GI and differed only in the predicted insulin demand (predicted FII: 65 vs 30). Capillary blood was sampled every 30 minutes from 0830 h until 1630 h and assayed for glucose and insulin concentrations.

Outcomes

The low FII diet resulted in 53% lower mean insulin incremental area under the curve (AUC, mean ± SEM) than did the high FII diet (31940 ± 4060 vs 68110 ± 11360 pmol/L*min, P = 0.003) over the course of total 8-h session despite no significant difference in glucose AUC (360 ± 88 vs 387 ± 70 mmol/L*min respectively, P = 0.73). The same pattern of insulin secretion was also found in individual meals between the two diets. The pre-meal insulin concentrations had no effect on postprandial insulin AUC for both diets (P = 0.24).

Conclusion

A low FII diet produced significantly lower day-long insulin secretion than did a high FII diet even when matched for macronutrients, fiber and GI. A food insulin index may be helpful in the management of diabetes.

Concurrent Session 5: Micronutrients

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Improving vitamin D status abrogates age-related change in bone markers in older women

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Background

Evidence that improving vitamin D status, without changing calcium intake, has a positive effect on bone turnover is limited.

Objective

To measure the effect of 25 hydroxyvitamin D_3 ($25(OH)D_3$) supplementation, in vitamin D deficient women, on markers of bone turnover , namely osteocalcin (OC), a biomarker of bone formation, and C-telopeptide (CTX), a biomarker of bone resorption.

Design

The study design was a randomised controlled double-blind intervention administering 4000 IU vitamin D_3 (cholecalciferol) (n=42) or placebo (n=39) daily for 6 months to South Asian women, aged >20 years, living in Auckland. Subjects had serum 25(OH)D concentration < 50 nmol/L, and mean dietary calcium intake of 700±300mg/day. Exclusion criteria included vitamin D supplementation > 1000 IU/day.

Outcomes

Median (25^{th}, 75^{th} percentile) serum 25(OH)D increased significantly from 21 (11, 40) to 75 (55,84) nmol/L with supplementation and also in the placebo group from 22 (15, 32) to 32 (24, 36) nmol/L (due to seasonal variation). There were no changes in serum calcium or parathyroid hormone. In women who were older than 49 years or post menopausal (n=26), who were not supplemented (n=13), CTX levels increased from 0.317±0.18 to 0.372±0.19 µg/L (P= 0.001), and OC increased from 20.00±6.56 to 23.38±8.03 µg/L indicating an increased rate of bone turnover.

Conclusion

Supplementation appeared to abrogate the age-related increase in bone turnover; CTX decreased from 0.39±0.15 to 0.36±0.17 (P= 0.012) and there was no significant change in OC. In women who were under 49 years and premenopausal (n=55; 29 supplemented), there was no significant response to supplementation in either CTX or OC. Increasing serum vitamin D levels in older women who are vitamin D deficient suppresses the age-induced increase in bone turnover and reduces bone resorption which would normally be exacerbated in conditions of low serum 25(OH)D.

Consumption of salmon *vs* salmon oil capsules – effects on selenium and omega-3 PUFA status

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Background

Salmon provides long chain (LC) omega-3 PUFA and selenium which are well recognized for their health benefits. The omega-3 and selenium status of the New Zealand population is marginal and consuming salmon might be more beneficial to increase both the omega-3 and selenium status compared to capsules.

Objective

To compare the effects of consuming salmon with salmon oil capsules on red blood cell (RBC) LC omega-3 levels and plasma selenium concentrations.

Design

Healthy volunteers (n=44) were randomly assigned to one of 4 groups consuming 2x120g servings of salmon/week or 2, 4 or 6 salmon oil capsules/day, for 8 weeks. Linear regression analysis predictive models were fitted to the capsule data to predict changes in RBC LC omega-3 levels with intakes of LC omega-3 from capsules in amounts equivalent to that consumed from salmon. Changes in selenium status (plasma selenium and whole blood glutathione peroxidase (GPx)) were compared between groups consuming salmon and capsules (3 groups combined).

Outcomes

Salmon, 2, 4 and 6 capsules provided 0.82, 0.24, 0.47 and 0.68g/day of LC omega-3 fatty acids. Salmon provided 7 µg/day and capsules <0.02 µg/day of selenium. The predictive model (R^2=0.31, p=0.003) showed that increases in RBC LC omega-3 levels were similar with intakes of 0.82g LC omega-3 from salmon and capsules (1.92[95%CI: 1.35-2.49] vs 2.31[1.75-2.87]%). Plasma selenium increased significantly more with salmon than capsules (10.6[5.73-15.6] vs 2.48[-0.71-5.66]µg/L, p=0.008). Whole blood GPx concentrations did not differ between groups.

Conclusion

LC omega-3 status was similarly improved with consumption of salmon and capsules. However consuming salmon had the added benefit of increasing plasma selenium concentrations which is of particular relevance to the New Zealand population who has a marginal selenium status.

Micronutrient status in children with cerebral palsy

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Background

Cerebral palsy "CP" is associated with undernutrition and subsequent growth deficits. Much research has focused on anthropometric data and energy intake with little information available on other parameters.

Objective

Investigate a variety of micronutrients and other biochemical parameters including oxidation and antioxidant levels in children with CP, either orally or enterally fed and compare with typically developing children.

Design

We are currently conducting an observational study in children 4-12 years with severe CP (n=25) compared to typically developing children (n=20). Data is being collected on 3 occasions at 6 week intervals.

Outcomes

Preliminary data indicate differences in height z-scores (mean (±SD)= -1.7 (0.9); 0.0 (0.8), P<0.00) and weight z-scores (mean= -1.5 (1.6); 0.6 (0.8), P<0.00) between CP subjects and controls, respectively. No significant differences were noted between enterally and orally fed children with CP. The lower bicarbonate (mean=22.5 (3.0 mmol/L); 24.3 (1.4), P=0.01), lower phosphorus z-scores (mean=0.3 (0.9); 1.0 (0.6), p=0.01) and raised anion gap (mean=10.2 (2.2 mmol/L); 8.9 (1.6), P=0.04) in CP compared with controls, may indicate metabolic acidosis and warrants further investigation. Raised mean cell volume z-scores (mean= 1.2 (1.5); -0.7 (1.1), P<0.00) and lower urate levels (mean=0.19 (0.05 mmol/L); 0.23 (0.03), P=0.01) were also found, which may indicate megaloblastic anaemia, despite large elevations in red cell folate levels (mean=1059 (403 nmol/L), 786 (147), P=0.01) as high as over the 95th percentile in all of the enterally fed children. It is hypothesised that the methyl-folate trap may be responsible for the increased symptomatology associated with severe CP. Remaining data are yet to be analysed and will be discussed, including methylmalonic acid, methylation cycle intermediates and oxidation markers.

Conclusion

Detecting and correcting biochemical abnormalities may have significant public health implications, as care for these children can typically cost 1.5 million dollars per lifetime, and may lead to improvements in immune, nervous system and cognitive function, as well as catch up growth in these children. Healing outcomes may also be augmented via alleviating any deficiencies and supporting optimal body function.

Insulin sensitivity is improved with vitamin D supplementation in South Asian women who are vitamin D deficient and insulin resistant – a randomised, placebo-controlled trial

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Background

Despite previous evidence of vitamin D supplementation improving insulin secretion and/or resistance, two recent supplementation trials of short duration (4 and 6 weeks) produced inconclusive results.

Objective

To investigate the effect of improving vitamin D status over a period of 6 months, on insulin resistance in vitamin D deficient South Asian women, aged 23-68 years, living in Auckland, New Zealand.

Design

Randomised controlled double-blind intervention administering 4000IU vitamin D_3 (n=42) or placebo (n=39) daily for 6 months. Subjects were insulin resistant (homeostasis model assessment 1 (HOMA1) >1·93) and had serum 25(OH)D concentration < 50 nmol/L. Exclusion criteria included use of diabetic medication and vitamin D supplementation > 1000 IU/day. The HOMA2 computer model was used to calculate insulin sensitivity, insulin resistance and beta cell secretion. Subjects were tested at baseline, 3 months and 6 months (endpoint).

Outcomes

Median (25th, 75th percentile) serum 25(OH)D increased significantly from 21 (11, 40) to 75 (55, 84) nmol/L with supplementation, and also in the placebo group from 19(13, 29) to 29(23, 36) nmol/L P < 0.001. Significant improvements were seen in insulin sensitivity and insulin resistance (P = 0·003, P = 0·02 respectively), and fasting insulin decreased (P = 0·02) with supplementation compared to placebo. There was no change in C-peptide with supplementation. Insulin resistance was most improved after 6 months when endpoint serum 25(OH)D reached ≥80 nmol/L, but changes were not significant at 3 month time point. Secondary outcome variables (lipid profile and high sensitivity C-reactive protein) were not affected by supplementation.

Conclusion

Improving vitamin D status in insulin resistant women resulted in improved insulin resistance and sensitivity but no change in insulin secretion. Optimal vitamin D concentrations for reducing insulin resistance were shown to be 80 – 119 nmol/L. The beneficial effect of supplementation increased with time, and a period of

longer than three months supplementation appears to be desirable.

Folate status; a recognised determinant of colorectal neoplasia may be modified by bitter taste perception and genetics

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Background

Humans perceive bitterness as a consequence of signalling mediated by transmembrane G protein-coupled receptors. The seven genes encoding these proteins are collectively termed TAS2Rs. The TAS2R38 gene has a polymorphic locus known to alter bitter perception. Recent studies have suggested that folate intake levels are inversely associated with colorectal neoplastic risk, and in addition to this other studies have shown that the ability to perceive bitterness as tested with PROP sensitivity is significantly correlated with adenomatous polyp number, and reduced vegetable consumption.

Objective

Given the importance of vegetables as a major source of folate, and folate playing a key role in colorectal neoplastic risk, we examined patients with adenomatous polyps and controls, for a relationships between folate intake and/or red cell folate status with respect to TAS2R38 genotype.

Design

A preliminary cohort of 27 adenomatous polyp cases and 28 controls were recruited into the study. TAS2R38 genotype was determined using PCR. Bitter taste phenotype was assessed using different PROP concentrations. A food frequency questionnaire was used to quantify folate intake. Red cell folate was measured using routine competitive binding assay.

Outcomes

Analyses showed that TAS2R38 taste genotype is significantly associated with PROP sensitivity (Spearman rank correlation $p<0.0001$; ANOVA of taste phenotype by taste genotype $p<0.0001$). TAS2R38 genotype also modifies red cell folate level (ANOVA of red cell folate by taste genotype $p=0.0170$). This is consistent with a hypothesis that bitter taste may affect dietary intake of folate, and is further supported by TAS2R38 approaching significance when examined for its effect on dietary intake of folate (ANOVA of dietary folate intake by taste genotype $p=0.0557$).

Conclusion

Bitter taste genetics may modify the character of the food we eat, particularly folate rich vegetables, and hence alter folate status and therefore risk for colorectal neoplastic change.

Prevention of obesity in mice by the Australian fruit Illawarra Plum

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Background

The prevalence of obesity is increasing worldwide, which also increases the risk of the development of type 2 diabetes, cardiovascular disease, certain cancers, as well as other disease. Prevention and treatment of obesity is therefore relevant to health promotion. Phytochemicals, such as anthocyanins, show promise as natural agents for preventing obesity. Therefore nutritional supplementation with phytochemicals that can assist weight control and prevent the pathological consequences of obesity represents a novel approach to the problem.

Objective

To determine if the anthocyanin-rich Australian fruit Illawarra plum will prevent the development of obesity in mice.

Design

Mice were fed a western diet with or without the addition of Illawarra plum (n = 14 / group) from weaning for 10 wk. Diets were calorically matched. Mice were weighed three times a week throughout the study, food intake and faecal output were measured, and fat pad size was assessed at the end of the treatment period.

Outcomes

Weight gain was significantly reduced in mice that consumed the Illawarra plum diet (for both male and female mice, $P<0.05$). Daily food intake was not different between the groups, however faecal output was greater in the Illawarra plum fed group ($P<0.05$). Epidydimal fat pads were almost half the weight in the Illawarra plum fed group compared to the group without plum ($P<0.05$).

Conclusion

Illawarra plum is able to prevent the development of obesity in mice. We are currently investigating the molecular mechanisms underlying this effect. This is a novel use for a traditional Australian food.

Is the level and form of iron in muscle influenced by finishing systems in lambs?

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Background

Iron deficiency is a major nutritional disorder affecting populations of all ages across the world. Green leafy materials, grain legumes and animal products such as liver and meat are good sources of iron. Within the agrifood

sector in Victoria one of newly developed future farming research strategies is designed to boost the productivity and quality (bio-fortification) of farm products, while remaining sustainable and competitive and also remaining adaptive to climate change.

Objective

To investigate the variation in total-, heme-, and nonheme-iron content in lamb finished by either short term grain feeding or grazing.

Design

Seven month old second cross (Poll Dorset x Border Leicester Merino) lambs were run on a property in southern Victoria. The lambs were divided into two groups of 15. One group of lambs was maintained under continuous grazing while the other group was finished for five weeks on grain feeding. Lambs on the grain diet had hay available all of the time. The grain diet consisted of barley (80%) and lentils (20%) and the grass diet consisted predominantly of rye grass and barley grass. Both groups were slaughtered at a commercial abattoir and 50 g of loin muscle was collected for iron determination.

Outcomes

Total iron (18.34 vs 17.37; $P>0.05$) and nonheme iron (10.86 vs 11.06; $P>0.05$) concentrations in lamb muscles (mg/ kg of muscle) were similar between both pasture and grain groups, respectively. Meat from lambs fed grain had a significantly lower level of heme iron (6.31 vs 7.48; $P<0.05$) content compared to lambs grazed on pasture.

Conclusion

Application of grain in lamb finishing systems to improve the productivity, during seasons of drought reduced muscle heme iron content, but not the total iron or nonheme iron content. Further research is needed to confirm whether this effect is consistent in a larger number of animals.

Are a substantial proportion of reproductive aged females undertaking hormonal contraceptive practices that might alter their risk of iron deficiency?

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Background

Iron deficiency most commonly affects reproductive-aged females, with one in three being diagnosed by the age of 43. Menstruation contributes greatly to iron deficiency risk and hormonal contraceptives (HC) may reduce menstrual blood and iron loss. The current iron RDI does not account for HC use and the potential reduction in bleeding frequency and volume, and subsequently iron loss, as a result.

Objectives

This study was designed to determine the rate of current and intended contraceptive practices, menstrual bleeding frequency and medically diagnosed iron deficiency amongst female university students.

Design

A 20 item questionnaire conducted in female Gold Coast University students. Descriptive and chi-squared analyses were conducted.

Outcomes

Of the 1010 respondents, over half (59%) reported currently using the oral contraceptive pill (OCP). Two thirds of OCP users reported skipping menstruation, primarily for convenience. HC users self-reported less regular menstrual bleeding frequency ($p=0.054$). Thirty percent of HC users had been on their current regimen for 3-5 years and two thirds intended to be on it for up to in excess of 5 years. Sixty-two percent of respondents were interested in a HC regimen designed to further reduce blood loss volume and frequency. Thirty percent of respondents self-reported having had medically diagnosed iron deficiency over their life and two thirds were undertaking behaviours that may increase iron deficiency risk.

Conclusions

OCP use was extremely prevalent. Given that HC's may reduce volume and frequency of menstrual blood loss and many females were interested in, and have undertaken, practices that reduce menstrual bleeding frequency the RDI's for iron in this population might benefit from accounting for HC use.

Folate intake and status in young Australian women

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Background

Folate plays an important role in the prevention of neural tube defects (NTDs) and potentially in modifying risk factors for chronic disease. Mandatory folate fortification will be introduced in Australia in September 2009. Therefore, an update on folate intake and status in young women is warranted.

Objective

To investigate the relationship between folate intake and status in young women, and to compare serum folate concentrations to those in other countries where folate fortification programs are in place.

Design

Females (n=256; age 22.4±3.8; BMI 21.4±2.6 kg/m^2; mean±SD) not taking vitamin or mineral supplements were recruited to participate in a cross-sectional study. Fasting blood samples were analysed for folate in serum and red blood cell (RBC). A 235-item food frequency questionnaire was used to measure dietary intake.

Outcomes

Mean dietary intakes of folate and folic acid were 504±257 and 113±123 µg/day, respectively. The Estimated Average Requirement was met by 76%, and those who met the national guidelines for serves of cereals and fruit had significantly higher folate intake (P<0.001). Both dietary folate and folic acid were positively correlated with serum and RBC folate (P<0.001). Only 1.7% of women had low serum folate (<7.0 ηmol/L), while 88% and over a quarter had RBC folate concentrations >500 ηmol/L and >1000 ηmol/L, respectively. RBC folate concentration associated with a reduced risk of NTDs (≥906 ηmol/L) was achieved by 26% of participants, who had significantly higher folic acid intake (167±149 vs. 88.8±96.7 µg/day). Mean serum folate (23.5±10.4 ηmol/L) was comparable to post-fortification levels in young women in the USA (26.9 ηmol/L), Canada (33.1 ηmol/L), UK (22.1 ηmol/L) and Ireland (24.2 ηmol/L), but higher than previous Australian data (16.7 ηmol/L).

Conclusion

The majority of young women in this study met dietary folate recommendations and achieved optimal folate status. Serum folate values were similar to countries where fortification is mandatory and optimal status was achieved by the consumption fruit and fortified cereals. Folate status should be monitored, as there is the possibility of undesirable effects of very high folate status in some women.

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Long-chain omega-3 polyunsaturated fatty acids may reduce obesity

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Background

Current recommendations for reducing obesity advocate dietary energy-restriction and increased physical activity, but many individuals have difficulty complying with these recommendations. Simpler strategies are required.

Objectives

To review the literature regarding potential benefits of the long-chain omega-3 polyunsaturated fatty acids (LC n-3) eicosapentaenoic acid and docosahexaenoic acid for reducing obesity.

Design

Electronic database search for articles related to n-3 and obesity.

Outcomes & Conclusion

Increased consumption of n-3 protects against obesity in animals fed a high-fat diet[1] and reduces body fat in already obese animals[2]. There is also evidence of body fat reduction in humans but studies are few and of short duration, with small sample sizes and lack of adequate control making it difficult to draw conclusions[3]. LC n-3 may reduce obesity via appetite suppression, adipocyte apoptosis and changes in gene expression which reduce fat deposition and increase fat oxidation and energy expenditure[3]. LC n-3 might also suppress genes in skeletal muscle regulating catabolic pathways[4] and increase expression of those regulating anabolic pathways [5], thus aiding retention of lean tissue mass and metabolic rate. Well-controlled long-term human trials are needed to confirm the anti-obesogenic efficacy of LC n-3 supplementation and identify mechanisms of action.

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Losing weight the healthy way – how do omega-3 fatty acids really work?

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Background

Adipose tissue is a lipid storage depot which is highly responsive to nutritional signals or energy deprivation. It is largely responsible for lipid mobilisation regulating energy homeostasis via triacylglycerols (TAG) accumulation and breakdown to non-esterified free fatty acids (NEFA) and glycerol. An imbalance in lipid mobilisation causes dramatic changes in the circulating NEFA, leading to inflammation, weight gain and consequently obesity.

Objective

Polyunsaturated fatty acids, mainly the omega-3 fatty acids, have been known to have an anti-obesity effect in humans and animals, though the underlying mechanism involved is still not clearly defined.

Design

In this study, we examined the effect of eicosapentaenoic acid (C20:5n-3; EPA) and docosapentaenoic acid (C22:5n-3; DPA) in comparison to saturated (STA; stearic acid, C18:0) and monounsaturated fatty acids (OA; oleic acid, C18:1n-9) in 3T3-L1 adipocytes.

Outcomes

As hypothesised, omega-3 EPA and DPA produced smaller multilocular brown adipocyte-like droplets and significantly suppressed expression of D9D/SCD1 desaturase, a diet-induced obesity marker, distinct from the effects of STA or OA. In addition, omega-3 fatty acids significantly decreased lipolytic (LPL, HSL and FABP), apoptotic (UCP2 and Caspase3) and pro-inflammatory adipokines (MCP-1 and IL-6), and increased β-oxidation (CPT1) expression upon bacterial endotoxin lipopolysaccharide challenge. It is speculated that omega-3 fatty acids influence the TAG accumulation and circulating lipids via modulation of lipid droplets associated proteins to decrease fat storage in adipose tissue.

Conclusion

These data suggest that omega-3 fatty acids could be potential therapeutic agents for a healthy approach for weight loss.

Plasma fatty acid profiles are different in obese versus non-obese asthma

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Background

The mechanisms leading to the association between obesity and asthma are unknown. We hypothesise that obesity is associated with changes in the circulating fatty acid profile, which contributes to a worsening of inflammation and clinical markers in obese asthma.

Objective

To assess circulating levels of fatty acids in asthmatic subjects with and without obesity; and to determine whether fatty acid levels are associated with inflammation in obese asthma.

Design

Non-obese (BMI<30kg/m^2, n=60) and obese (BMI≥30kg/m^2, n=43) adults with asthma were recruited via ambulatory care clinics at John Hunter Hospital, NSW. Plasma levels of total fatty acids were measured by gas chromatography. Plasma inflammatory markers, including CRP and IL-6, were assessed by ELISA.

Outcomes

Obese and non-obese asthmatic subjects had similar total plasma fatty acid levels. However, compared to non-obese subjects, obese subjects had higher mean [SD] % saturated (29.5 [2.1] vs 28.6 [1.7] %, p=0.01) and % monounsaturated (27.5 [3.2] vs 24.4 [3.4] %, p<0.00001) fatty acids. A weak association was observed between % saturated fatty acids versus CRP and % monounsaturated fatty acids versus IL-6. Conversely, the mean [SD] % polyunsaturated fatty acids was significantly lower in obese versus non-obese asthma (42.9 [4.5] vs 47.0 [4.2] %, p<0.001). The median [IQR] % omega-3 fatty acids was higher in non-obese (7.7 [6.6, 9.2] %) than obese asthma (6.9 [6.5, 7.7] %, p=0.02), and was negatively associated with CRP.

Conclusion

This study suggests that in asthma, obese subjects have a different fatty acid profile to non-obese subjects, which may contribute to increased inflammation and worsening clinical markers in obese asthma.

Funding

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Higher erythrocyte LCn-3 PUFA content is associated with a healthier body composition

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Background

There is growing evidence to suggest that an increased intake of long-chain omega-3 polyunsaturated fatty acids (LCn-3 PUFA) may be associated with improved body composition.

Objective

to determine the relationship between body composition and LCn-3 PUFA erythrocyte content.

Design

Height, weight and erythrocyte LCn-3 PUFA (eicosapentaenoic acid (EPA), docosapentaenoic acid (DPA) and docosahexaenoic acid (DHA)) content were measured in 335 participants (135 male (M) 200 female (F); 46 ± 12 y, BMI 31 ± 5 kg/m^2 and 35 ± 7 kg/m^2 respectively). Waist circumference (WC, n=239) and percentage body fat (BF% by DEXA, n=238) were assessed in sub-groups of volunteers. Data are presented as mean ± SD.

Outcomes

The Omega-3 Index (EPA+DHA) was 5.1 ± 1.2% while total LCn-3 PUFA status was 7.4 ± 1.4%. (EPA: M, 0.80 ± 0.28%, F, 0.86 ± 0.39%; DPA: M, 2.42 ± 0.37%, F, 2.28 ± 0.34%; DHA: M, 4.17 ± 0.96%, F, 4.28 ± 1.02%). In women, BMI was inversely related to total erythrocyte LCn-3 PUFA (r=-0.20, P=0.01), particularly DHA (r=-0.23, P<0.001). A smaller WC was associated with higher erythrocyte total LCn-3 PUFA in both sexes (M, r=-0.26, P<0.02; F, r=-0.32, P<0.001). These associations were most strongly influenced by DPA in men (r=-0.29, P<0.01) and DHA in women (r=-0.36, P<0.001). BF% was inversely related to erythrocyte EPA (M, r=-0.24; F, r=-0.18), DPA (M, r=-0.35; F, r=-0.19) and DHA (M, r=-0.27; F, r=-0.34), (P<0.03 for all).

Conclusion

Higher erythrocyte LCn-3 PUFA status is associated with a healthier body composition in men and women.

Associations between baseline erythrocyte n-3 polyunsaturated fatty acids and weight indices in volunteers for a weight loss dietary intervention

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Background

Research indicates that different types and amounts of dietary fat influences the accumulation or possible reduction of excess body fat. In particular n-3 PUFA may affect weight status and abdominal obesity in humans. We are conducting a 12 month dietary intervention trial to test this effect [ACTRN12608000425392].

Objective

The aim of this study was to identify any associations between erythrocyte n-3 PUFA composition and weight status in overweight and obese volunteers at baseline of the trial.

Design

Volunteers for the trial were recruited by advertising in the Wollongong community. Data from fasting blood samples, and anthropometric (weight, height, waist, hip) and percent body fat measures were available for 51 overweight (BMI 25-29.9kgm^{-2}), 43 obese (BMI 30-34.9) and 23 more obese (BMI >35kg) subjects. Erythrocyte fatty acids were determined using standard laboratory procedures in a quality assured pathology laboratory (Analytical Reference Laboratories (ARL) Pathology, Melbourne). After examination of the data, associations between erythrocyte n-3 PUFA (% total fatty acids) and weight indices across the study sample and within BMI group were examined using Spearman's correlation co-efficient.

Outcomes

For each group, the mean age was 45±8; 44±8 and 46±6yr; mean waist was 96.9±6.1; 109.1±8.4; 118.2±13.7cm, and mean fat mass was 34.3±6.6; 36.4±8.0 and 43.4±6.1% respectively. For the whole sample, there were no associations between n-3 PUFA status and any of the weight indices. In the overweight group, a significant ($P<0.05$) negative relationship was found between hip measurements and total n-3 PUFA (r = 0.-340) and 22:6n3DHA (r = -0.333), and in the more obese group a significant positive relationship was found between BMI and 22:6n3DHA (r=0.453).

Conclusion

There was no association between erythrocyte n-3 PUFA and weight indices in this sample of study volunteers. Within group relationships may reflect differences in dietary intake which will be further explored.

Funding source

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Baseline fish consumption patterns in overweight volunteers participating in a trial examining dietary fat and weight loss

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Background

Increased fish consumption is associated with a number of favourable health outcomes, including a decrease in the incidence of cardiovascular disease, stroke and blood pressure. Current Australian health organisations recommend consuming two 150 gram serves of fish per week, however, population based research suggests this recommendation is not being met. Investigation of habitual fish consumption patterns is also required to understand changes in health outcomes when conducting a dietary trial in which fish consumption is important.

Objective

To describe the habitual fish consumption patterns of a group of overweight volunteers involved in a trial of dietary fat in weight loss [ACTRN12608000425392], and compare results to current fish recommendations.

Design

Diet histories (n = 118) were taken at baseline. Fish and fish-containing foods were categorised according to the methods used in the 1995 National Nutrition Survey (NNS). Data was analysed to determine mean daily fish (g), which was compared to the current recommendations for fish intake. The contribution of different categories of fish to total fish intake was calculated. The results of these analyses were then compared to the findings of the NNS.

Outcomes

Mean daily intake of fish was 36.8g, with men found on average to consume 25.4g more fish per day than women. When compared to the current health recommendation of consuming two 150g serves of fish per week, it was found that 72% of participants did not meet this recommendation. Canned fish made up 49.6% of the total fish consumed, followed by cooked fin fish (34%), fish products such as fish fingers or cakes (7.5%), crustaceans and molluscs (4.9%), and mixed dishes containing fish (3.8%). In the NNS, only 12.8% fish consumed was canned or packed.

Conclusion

The majority of participants did not consume the recommended serves of fish per week, leaving room for change during the dietary intervention. Canned fish accounted for almost half of all fish consumed, which was higher than that found in the NNS, which may reflect the variety of canned fish available today compared to 1995.

Funding source

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Differential effects of a single oral dose of EPA or DHA rich fish oil on platelet aggregation in healthy human subjects

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Background

Increased platelet aggregation is the major cause of heart attacks, stroke and deep vein thrombosis. Long chain omega-3 fatty polyunsaturated acids (LC n-3 PUFA; eicosapentaenoic acid, EPA; docosahexaenoic acid, DHA) are known to reduce platelet aggregation; however studies in the published literature involving EPA and/or DHA supplementation have yielded equivocal results. Recent *in vitro* studies have demonstrated that inhibition of platelet aggregation by LC n-3 PUFA is gender specific.

Objective

The objective of this study was to examine effects of dietary supplementation with a single dose of EPA or DHA rich oils on platelet aggregation (ex-vivo) in male versus female subjects over a 24 hour period.

Design

A placebo controlled trial was conducted in a total of 90 healthy male and female adults (males *n* = 45; females *n*= 45). Platelet aggregation was measured at baseline and 2, 5 and 24 hours post supplementation with either a placebo (olive oil) or EPA or DHA rich oil. The relationship between LC n-3 PUFA and platelet activity at each time point was examined according to gender vs. treatment.

Outcomes

EPA was significantly and progressively most effective in reducing platelet aggregation at 2, 5 and 24 hours post supplementation (-3.6%, -8.8%, -13.3 %, respectively). DHA was equally effective at 24 hours post supplementation (-11.9%). When grouped by gender, males showed a greater reduction in platelet aggregation at 2, 5 and 24 hours following EPA supplementation (-11%, -10.6%, -20.5%) compared with placebo, whereas DHA was not significantly effective. In contrast, DHA was significantly most effective in reducing platelet aggregation at 24 hours (-13.7%) in females while EPA was not effective compared with placebo.

Conclusion

Significant gender differences exist to reduce platelet aggregation in response to EPA or DHA. Males benefit more from EPA supplementation while in females, the platelets are more responsive to DHA.

Dose of omega-3 PUFA required to lower plasma triglycerides in pre-menopausal women

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Background

The hypotriglyceridemic effects of long chain (LC) ω-3 PUFA are well established for men and post-menopausal women; however the benefits for young women are unknown.

Objective

We aimed to determine the effective dose of LC ω-3 PUFA for lowering plasma triglycerides (TG) in pre-menopausal women through a dose-response intervention with low doses of fish oil.

Design

A randomized, double-blind, placebo-controlled trial of 8 weeks duration was conducted in 29 women, using 0, 0.35, 0.7 or 1.0 g/day LC ω-3 PUFA from DHA-rich tuna oil and/or placebo capsules. Fasting plasma TG and erythrocyte LC ω-3 levels were determined using enzymatic colorimetry, and direct transesterification followed by gas chromatography respectively.

Outcomes

There was a direct relationship between the supplemental dose of LC ω-3 PUFA and subsequent changes in erythrocyte EPA + DHA levels (R^2 = 0.64, $p<0.0001$). A weaker relationship was observed between changes in erythrocyte LC ω-3 PUFA and those in plasma TG levels (R^2 = 0.15, $p<0.05$). Erythrocyte EPA + DHA levels rose from a mean baseline level of 4.9±0.2% to 5.0±0.4%, 5.4±0.3%, 6.9±0.2% and 7.4±0.4% after supplementation with 0, 0.35, 0.7 and 1.0 g/day LC ω-3 PUFA respectively. Similarly plasma TG levels were unchanged from a mean baseline level of 1.1±0.1 mmol/L after 0 and 0.35 g/day, but decreased to 0.8±0.1 mmol/L after supplementation with 0.7 or 1.0 g/day LC ω-3 PUFA. Both the TG-lowering effect and incorporation of EPA and DHA into RBC appeared to plateau between 0.7 and 1.0g/day LC ω-3 PUFA.

Conclusion

This study suggests that 0.7g/day of LC ω-3 PUFA, but not 0.35g/day, is effective for TG-lowering in pre-menopausal women. Previous studies suggest greater than 1.0 g/day LC ω-3 PUFA is required for TG-lowering of a similar magnitude.

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DPA (22:5n-3) reduces the expression of genes involved in fatty acid synthesis in liver cells

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Background

Docosapentaenoic acid (DPA) is a long chain n-3 (LCn-3) fatty acid found in fish oil and lean red meat along with other two n-3 fatty acids, namely eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Both EPA and DHA have been demonstrated decrease lipogenesis and triglyceride synthesis in the liver. However, the effect of DPA on lipogenesis in the liver is currently not known.

Objective

The aim of this study was to elucidate the effect of DPA, compared with EPA and DHA, on lipid synthesis using FAO rat liver cells as an *in vitro* model.

Design

The FAO cells were treated with 50µM of EPA, DPA and DHA for 24 and 48 hours. Oleic acid (50µM) was used as a fatty acid control, while ethanol was used as vehicle. Each treatment was performed in triplicate. The fatty acid analysis of the cells was conducted using gas chromatography. Real time-PCR was used to measure gene expression levels and western blot analysis was conducted to study the protein expression.

Outcomes

It was observed that supplementation of cells with DPA led to a significant increase in DPA and EPA levels in the cell phospholipids and vice-versa. However, DPA supplementation did not increase DHA levels suggesting that a part of DPA gets converted to EPA but in these cells, it does not get metabolised to DHA. It was also observed that all the three fatty acids including DPA, led to a significant decrease in the expression levels of SREBP-1c, HMG CoA reductase, FASn, ChREBP and ACC, relative to the vehicle-treated cells. Western blot analysis of ACC and SREBP-1 protein also showed a decrease in expression of respective proteins in the cells treated with DPA. It was also confirmed in the study that the effect of DPA on these genes was due to DPA itself and not due to its conversion into EPA. Preliminary pilot studies from cellular triglyceride assay show that DPA may possess triglyceride lowering effects.

Conclusion

Thus DPA may be as effective as EPA and DHA in reducing lipogenesis in liver cells.

Concurrent Session 7: The Science Behind Weight Management

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Are high carbohydrate, high fibre diets the best choice for overweight women?

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Background

Prospective and experimental studies indicate that high carbohydrate diets rich in dietary fiber and wholegrains are protective against cardiovascular disease, diabetes and obesity. Numerous studies, however, have suggested that diets involving a reduction in carbohydrate intakes may be more beneficial for overweight and obese people and for those with features of the metabolic syndrome. However in most such studies the sources of dietary carbohydrate in the comparison diets may not have been appropriate.

Objective

To compare, in overweight women, two low-fat weight loss diets, one high in fibre-rich carbohydrate and the other high in protein and moderately reduced in carbohydrate.

Design

Eighty-three overweight or obese women, 18-65 yrs, were randomised to either a high fiber, high carbohydrate (55% carbohydrate, >35 g total dietary fiber, 20% protein) diet (HFib) or a moderately high protein (30% protein, 40% carbohydrate) diet (HP) or to for 8 weeks. Energy intakes were restricted.

Outcomes

Participants on both diets lost weight (HFib: -3.3 kg; 95% CI: -4.2, -2.4 kg and HP: - 4.5kg ; 95% CI: -3.7, -5.4 kg), and reduced body fat (measured by DEXA), total and LDL cholesterol, triglycerides, fasting plasma glucose and blood pressure. However participants on HP lost more body weight (-1.3 kg; 95% CI: -2.5, -0.1kg; P=0.039) and total body fat (-1.3 kg; 95% CI: -2.4, -0.1; P=0.029). Diastolic blood pressure decreased more on HP (-3.7 mm Hg; 95% CI: -6.2, -1.1; P=0.005).

Conclusions

A high protein weight-reducing diet was associated with several more favourable outcomes when compared with a high carbohydrate, high fibre diet in high risk overweight and obese women.

The effect of two different weight loss approaches on anthropometric and metabolic profiles of obese individuals

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Background

Obesity, in particular central adiposity, is associated with increased plasma levels of pro-inflammatory cytokines and acute phase proteins, as well as with increased risk of insulin resistance, type 2 diabetes, cardiovascular disease, and stroke. A weight loss of between 5-10% can substantially reduce this risk. However, losing weight is not easy as hunger and slow progress can reduce motivation to succeed.

Objective

To compare the effect of two different weight loss diets on anthropometric and blood lipid measurements, as well as on levels of inflammatory biomarkers.

Design

Obese individuals (BMI 30-40 kg/m^2) followed one of two weight loss diets. Diet 1 used a healthy eating weight loss plan of 5000 kJ/day for 12 weeks. Diet 2 used a very low kJ diet (VLkJD) of 3000 kJ/day, with meal replacements, for 4 weeks. Nutrition information was given to both groups. Fasting blood samples, anthropometric measurements, 3-day food diaries, and health surveys were collected at baseline and post intervention.

Outcomes

The mean weight loss for Study 1 participants (n=18) was 3.17 kg (3.37%) compared to Study 2 participants (n=14) with a loss of 6.54 kg (6.92%) (p=0.001). Compared with Study 1, participants in Study 2 also experienced significantly greater reductions in BMI (p=0.000), cholesterol (p=0.005), LDL (p=0.01), blood glucose, total body fat and waist circumference ($p \leq 0.05$). There was a greater reduction in hip measurements in participants in Study 2 compared to Study 1, but the difference was not significant. Compared with Study 1. participants in Study 2 had significantly reduced leptin levels (p=0.000). Reductions in TNFa, CRP, IL-6, and LTB4, although greater in participants in Study 2, were not significantly different.

Conclusion

Participants in Study 2, following the VLkJD, experienced a weight loss of between 5-10% and greater reductions in central adiposity and inflammatory biomarkers compared to participants in Study 1. It would appear that short-term,

quicker weight loss is more effective than a slower, longer term weight loss strategy for reduced health risks.

Predictors of retention rates in a 12-week commercial web-based weight loss program

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Background

On average, 20 percent of participants drop-out of behavioural weight loss interventions. The identification of participant characteristics that predict retention could inform approaches to support and engage those who are more likely to drop out.

Objective

To describe retention rates of a 12-week commercial web-based weight loss program, and determine baseline characteristics of participants that predict retention.

Design

De-identified data for 11173 participants who enrolled in The Biggest Loser Club between August 15th 2007 and May 31st 2008 were provided, including dates of enrolment and cessation. An enrolment survey captured self-reported anthropometric data, demographics, dietary and physical activity behaviours, and reasons for trying to lose weight. Cox Proportional Hazards was used to explore predictors of retention from baseline to 12 weeks.

Outcomes

At 12 weeks, 89.6% of participants were retained. Preliminary analysis with unadjusted hazard ratios suggests being: aged 45 to 54.9 years (HR 0.53 95% CI 0.42-0.67), Obese Class I (HR 0.77, 95% CI 0.62-0.95), Anglo-Saxon (HR 0.58, 95% CI 0.44, 0.76), of highest socio-economic status (HR 0.65 95%CI 0.52-0.81), as well as exercising for 2 or 3 days per week (HR 0.74 ,95% CI 0.65-0.85), eating breakfast (HR 0.77, 95% CI 0.68-0.87) and using low fat products (HR 0.77, 95% CI: 0.69-0.87) were the strongest predictors of retention. The strongest predictors of drop-out related to poor dietary behaviours: frying foods [HR 1.25, 95% CI 1.11-1.40], using butter in cooking [HR: 1.19, 95% CI: 1.06-1.34], drinking full sugar soft drinks [HR1.39, 95%CI 1.23-1.57], skipping meals [HR 1.23, 95% CI: 1.09-1.38], and eating vegetables 'hardly ever' [HR 1.48, 95% CI 1.09-2.03]

Conclusion

Retention rates in the web-based program are high. Positive dietary and physical activity habits, along with some demographic characteristics were predictive of retention. Further study is required to determine if providing additional support to participants with the characteristics identified as predicting drop-out, increases retention.

Dietary intake changes in men participating in the SHED-IT weight loss intervention

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Background

The prevalence of overweight and obesity in men is high, yet they are less likely than women to attempt weight loss. Of the few weight loss interventions that have targeted men, none have comprehensively reported change in dietary intake.

Objective

To describe dietary changes in 65 overweight/obese men participating in the Self-Help, Exercise and Diet using Information Technology (SHED-IT) study.

Design

An assessor blinded randomized controlled trial compared the SHED-IT Internet intervention [n = 34] to an information only control group [n = 31]. Assessments were conducted at baseline, 3-month and 6-months. Dietary intake was assessed using the Dietary Questionnaire for Epidemiological Studies (DQES) a food frequency questionnaire developed by the Cancer Council of Victoria. The DQES was analysed using NUTTAB95.

Outcomes

The average portion size factor decreased significantly over time [χ^2 = 20.9, df=5, P<0.001] with no differences between groups. While both groups reduced mean daily kilojoule intake [GLM χ^2 = 34.5, df=3, P<0.001] there was a trend towards a greater reduction in the Internet group [GLM χ^2 = 3.3, P = 0.07]. Both groups reduced percentage kilojoules from fat [P<0.05], saturated fat [P<0.001] and energy dense/nutrient poor items, P<0.05 with no change in dietary fibre or alcohol [P>0.05].

Conclusion

Although men reported some positive dietary changes resulting in weight loss, they did not increase vegetable intakes nor decrease alcohol consumption while saturated fat, fibre and sodium intakes still exceeded national targets. Future interventions for men should promote specific food-based guidelines aligned with improving diet-related risk factors for chronic disease.

A clinical trial comparing a low glycemic index and a low fat, high fibre diet in women with polycystic ovary syndrome

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Background

Lifestyle modification has been shown to significantly improve reproductive and metabolic outcomes in women with polycystic ovary syndrome (PCOS), yet there are no specific dietary recommendations for these women and few studies have explored the effect of diet composition.

Objective

to compare the effects of a low glycemic index (GI) and high fibre diet on reproductive and metabolic outcomes in women with PCOS.

Design

Overweight and obese pre-menopausal women with PCOS (n = 96) were assigned to either an *ad libitum* low fat low GI (LGI) diet or a low fat high fibre (HF) diet in alternate order and followed until they had lost 7% of their baseline body weight. Changes in insulin sensitivity, glucose tolerance, body composition, plasma lipids, reproductive hormones, health-related quality of life and menstrual cycle regularity were assessed.

Results

Of the 49 subjects who completed the study, only 55% achieved a 7% weight loss within 12 months. Average weight loss was 4.8%, with no significant difference between the two groups. Change in whole body insulin sensitivity (ISI_{OGTT}) was significantly greater on the LGI diet compared to the HF diet (2.2 ± 0.7 vs 0.7 ± 0.6; p = 0.03). Fibrinogen levels were reduced on the LGI diet but increased on the HF diet (-0.2 ± 0.2 vs 0.2 ± 0.1; p = 0.05). Subjects on the LGI diet experienced a greater improvement in menstrual cycle regularity than those in the HF group (95% vs 63% of those who has irregular cycles at baseline; p = 0.03).

Conclusion

A low GI diet leads to greater improvements in insulin sensitivity and menstrual cycle regularity than a standard low fat, high fibre diet.

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Dietary arabinoxylan increases short chain fatty acid concentration of digesta from pigs consuming a high risk "Western diet": a pilot study

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Background

It is known that soluble fibres such as β-glucan can have important health benefits, including fermentation in the large bowel. End-products of such fermentation are short chain fatty acids (SCFA) which are known to be, at least part of the mechanism by which the potential health benefits occur. These include: reduced gut pH, and reduced risks of insulin resistance and colon cancer. However, little is known about the benefits of arabinoxylan (AX), the principal soluble fibre found in wheat.

Objective

To determine the effects of a wheat arabinoxylan concentrate on gut fermentation, the profile and concentrations of SCFA in eight areas of the gastrointestinal tract (GIT) were studied in a porcine model.

Design

Two groups of pigs (n=5) were fed a highly-digestible Western-type diet containing barbecued red meat, with or without a wheat extract containing 27% AX. After four weeks on the diet, the animals were euthanased and samples taken from four areas of the small intestine, and four areas of the large intestine, including the caecum. SCFA were analysed in the digesta of all sections.

Outcomes

Consumption of the high risk diet containing the AX fraction led to significantly higher total SCFA concentration in the caecum, and in the proximal and medial colon ($P<0.05$). For the caecum, acetic, propionic and butyric acids, were all greater ($P<0.05$) for the AX diet, though butyric was not higher in other locations. There were no significant differences between diets for all sections of the small intestine.

Conclusions

This pilot study suggests that an arabinoxylan-enriched wheat fraction is highly fermentable in the caecum. As part of a balanced diet, it could play a positive role in the maintenance of gut health. Further work is in progress, to ascertain the effects of the AX fraction on a range of other biomarkers.

Effect of chilli intake on platelet aggregation

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Background

Hyper-aggregation of platelets is associated with increased risk of stroke and cardiovascular disease. *In vitro* studies have shown a concentration dependent effect of capsaicin (active ingredient of chilli) on inhibition of platelet aggregation.

Objectives

We investigated the dose effect of chilli intake on platelet aggregation under *ex vivo* conditions.

Design

Fourteen participants with mean (±SD) age of 44.0±12.7 years undertook five meal-challenge tests, each in a random order. All meals comprised of bread, meat patty and a glucose drink. Three of these five meals also contained 20, 30 and 40 g of chilli paste (62% chilli), respectively. One meal included capsules filled with dry chilli powder. The other meal was a control. The nutrient composition of these meals was ~1900 kJ energy, 70% carbohydrate, 16% protein and 15% fat. Platelet rich plasma separated from citrated plasma collected at fasting and 40 and 120 min postprandially was subjected to adenosine-*diphosphate* (2.5, 5 and 10 µmol/L) induced platelet aggregation.

Outcomes

Intake of all five meals resulted in a reduced maximum platelet aggregation at 120 min postprandially compared to the fasting samples (overall $P<0.001$). The three meals containing chilli paste resulted in further inhibition of platelet aggregation compared to the bland (control) and the chilli capsule meals.

Conclusion

Consumption of chilli inhibits platelet aggregation and may reduce the risk for cardiovascular disease.

Effects of different amounts of chilli consumption on postprandial glucose and insulin

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Background

Repetitive postprandial hyperglycemia and hyperinsulinema increases the risk of developing type-2 diabetes and cardiovascular disease. Consumption of chilli has been

shown to lower and improve postprandial plasma glucose and serum insulin responses in healthy humans.

Objective
To investigate and compare the acute effects of a bland meal and meals containing different amounts of chilli paste on postprandial plasma glucose and serum insulin concentrations.

Design
Nineteen participants aged 43.6 ± 12.3 years (mean ± SD), BMI 28.3 ± 3.8 kg/m^2, took part in a randomised, cross-over study where postprandial responses to four different meals were assessed at fasting and at regular intervals to two hours postprandially. The four meals were a bland (spice free) meal and 3 chilli containing meals, including amounts of 20 g, 30 g and 40 g of chilli paste (62% cayenne chilli). The nutrition composition of the meals was ~1900 kJ energy, 70% carbohydrate, 16% protein and 15% fat.

Outcomes
Fasting concentrations of plasma glucose and serum insulin were not significantly different between the four meals (all P>0.16). The meals containing chilli led to a reduced incremental area under the curve (iAUC) for serum insulin compared to the bland meal (P=0.07). The net area under the curve (net-AUC) for glucose was not significantly different between the four meals (overall P=0.22). Results of participants above the median BMI (27 kg/m^2), showed a larger reduction in iAUC for insulin with increasing amounts of chilli (P<0.01).

Conclusion
Chilli consumption dose-dependently reduces postprandial hyperinsulinemia especially in overweight and obese people.

The effects of cocoa antioxidants in a rat model of non-alcoholic steatohepatitis

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Background
Non-alcoholic steatohepatitis (NASH), found generally as a consequence of the metabolic syndrome, is a condition in which the liver becomes fat loaded and further damaged by oxidative stress.

Objectives
To investigate the antioxidant effect of a cocoa rich diet to treat and prevent the conditions associated with non-alcoholic steatohepatitis in rats fed a high fat methionine choline deficient (MCD) diet.

Design
12 week old female Sprague Dawley rats completed one of six feeding regimes: 1) high fat MCD diet for 7.5 weeks followed by a high fat MCD diet with cocoa treatment for 4 weeks; 2) high fat MCD diet for 7.5 weeks followed by a high fat MCD diet with cocoa treatment for 8 weeks; 3) high fat MCD diet with cocoa prevention for 11.5 weeks; 4) high fat MCD diet with cocoa prevention for 15.5 weeks. Additional controls were fed a high fat MCD diet or a high fat methionine choline sufficient (MCS) diet.

Outcomes
7.5 weeks on the MCD diet resulted in non-alcoholic fatty liver disease in rats and increased glutathione (GSH) and liver weight significantly when compared to MCS control. Treatment and prevention with cocoa decreased liver GSH. Treatment for 4 and 8 weeks significantly increased RBC GSH when compared to MCD, MCS and prevention groups. MCD diet caused a significant reduction in plasma triglyceride levels; this effect was further increased by the cocoa treatment and prevention diets. RT-PCR demonstrated an increase in L-FABP mRNA in rats treated with cocoa for 4 weeks and a decrease in NOX-1 mRNA in all groups compared to MCS control.

Conclusion
GSH increases in liver as a protective measure against oxidative damage, which is not displayed when exogenous antioxidants are introduced. Cocoa acts to increase fat transport by increasing levels of L-FABP. NOX-1 may act through a signalling mechanism as a protective measure in liver disease.

The glycaemic response of concept snack bars made from a vegetable starch in comparison to standard bars made from standard white flour and from wheat starch

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Background
There is a market for reduced blood glucose response foods with high consumer appeal and validated nutrition and health advantages.

Objectives
To determine the blood glucose response in humans of snack bars made with; a) a vegetable starch-gluten mixture and; b) a wheat starch-gluten mixture, in order to determine the effects of the different starches in a background of approximately equivalent protein, starch, lipid and fibre.

Design
Fifty gram serve sizes of snack bars developed at the New Zealand Institute for Plant & Food Research, were tested in 13 free-living individuals. Oral glucose loads of 5, 12.5, and 25g in 320 m carbonated water were also measured on 2 occasions. Standard blood glucose testing procedure was followed.

Outcomes

The vegetable starch-gluten bar had a lower blood glucose response than the standard white bar (13.0 glycaemic glucose equivalents (GGE) compared with 23.4 GGE, approximately a 44% reduction, p=0.035). The vegetable starch-gluten bars did not have a lower blood glucose response than the wheat starch-gluten bars made on an equal starch, protein and fibre basis (13.0 GGE *c.f.* 19.2 GGE, p=0.206)

Conclusion

The vegetable starch bars showed an ability to favourably modulate glycaemic response in this setting, however more work is needed to determine whether it is possible to show a significant difference in blood glucose response between a vegetable starch bar and a wheat starch bar of equivalent protein, starch, lipid and fibre.

In vitro digestibility of starches

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Background

Starch is the major contributor of energy in the human diet. The worldwide crisis of over-nutrition and associated problems like diabetes has prompted intense interest in the study of starch digestibility. The digestibility of starch is influenced by factors such as botanical source, particle size, processing methods etc.

Objective

The objective of the present study is to determine the effect of various physical treatments on the digestibility of granular starches and to identify different microstructural mechanisms of starch digestion. .

Design

Four commercial starches (from potato, normal maize and two from high amylose maizes namely, Gelose 50 and Gelose 80) were used as raw materials. Treatments such as cryo-milling, freezing, freeze-drying, air drying, solvent drying and size separation were carried out. Raw and treated starches were digested *in vitro* for various time intervals. Microscopic analyses of partially digested starches were used to study the digestion mechanism.

Outcomes

The digestion rates as well as patterns differ between starches. Normal maize starches were rapidly digested compared to potato and high amylose maize starches, irrespective of physical treatments. The pores and channels present on the surface of normal maize starch granules render them more liable to amylase attack. Microscopic studies showed that maize starch is evenly digested by amylase whereas heterogeneous and nonspecific hydrolysis patterns were observed for potato and high amylose maize starches. Cryo-milling and freeze drying significantly increased potato and high amylose maize starch digestibility whereas oven drying, air drying and solvent treatment had minor effects. Smaller granules of potato starches were more rapidly digested than larger granules with an approximate inverse square dependence of digestion rate on diameter, suggesting that surface area/size determines digestion rates.

Conclusion

The rate and pattern of *in vitro* granular starch digestion depends qualitatively on the source of starch. Milling and freeze drying increase digestibility but do not alter the slow digestion property of potato and high amylose maize starches, whereas maize starch granules are always rapidly digested due to the presence of surface pores allowing ready internal access of amylase. Granule size is a quantitative determinant of starch digestion rate.

Concurrent Session 9: Dietary Methodology

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Dietary methodology in clinical trials: changing dietary fat profiles

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Background

Animal model studies have shown that dietary fat affects insulin action and the amount and distribution of body fat. Clinical trials that test this effect in humans need to be able to present dietary data that show the shifts in dietary fat have actually occurred.

Objective

The aim of this study was to present dietary data from clinical trials to demonstrate the role of dietary methodology in reporting differences in dietary fat in clinical trials.

Design

Data from one 3-month and two 12-month randomised controlled parallel trials were examined. Each trial involved overweight adult subjects, one inclusive of type 2 diabetes mellitus. Data was collected using a diet history questionnaire and a 3 day food record.

Outcomes

The baseline dietary intakes of the current 12 mo study showed an energy intake of 9973±3447.34kJ, with 18.47 ± 3.31% protein, 34.25±6.02% fat, 41.90±6.35% carbohydrate and a Poly:Mono:Sat fat ratio of 6:13:13. This was consistent with the profile of intakes reported by participants in the previous 2 trials. In the 3mo dietary fat/energy balance trial the groups with the higher PUFA targets increased dietary PUFA significantly more than the control groups ($P<0.001$), and in the 12 mo food based trial the walnut group significantly increased dietary PUFA compared to the control ($P=0.035$).

Conclusion

Dietary methodology supported the characterisation of the study samples at baseline and provided the means to reporting achievement of experimental targets commensurate with the hypothesised effects of dietary fats being tested in the trial.

Funding sources

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Fast food definitions: an explanation for conflicting evidence?

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Background

Frequent fast food (FF) consumption has been suggested as a contributor to higher obesity rates among those with lower socio-economic characteristics. However, evidence of associations between socio-economic factors and FF purchasing remains inconsistent. A possible explanation is the way FF use has been measured (consumption, purchasing frequency, total expenditure). Further, perceptions of 'what is FF' vary between socio-economic groups thus misleading results may be obtained when FF is not clearly defined.

Objective

To investigate definitions of FF use on associations with socio-economic predictors.

Design

Two studies are used in this comparison. First, the cross-sectional 2003/04 Australian Bureau of Statistics (ABS) Household Expenditure Survey (HES) (n=6921) which includes a broadly defined 'FF and takeaway' category. Purchasing is expressed as the percent of total household food expenditure dedicated to FF. Second, the multi-level cross-sectional 2003 Victorian Lifestyle and Neighbourhood Environments Study (VicLANES) (n=2547) which includes a tightly defined FF measure based on five franchised FF chains (McDonalds, KFC, Pizza Hut, Hungry Jack's and Red Rooster). This study recorded the frequency of purchasing over one month. Analysis is undertaken on similarly defined predictors (education, occupation and income).

Outcomes

Results from the ABS HES showed that those with no post-school education, the unemployed and lower income household spent less on FF and takeaway than those with higher socio-economic characteristics while analysis of the VicLANES data suggested lower socio-economic groups purchased FF more often.

Conclusion

This study demonstrates the potential for inconsistent evidence in relation to socio-economic predictors of FF purchasing. A plausible explanation for this is the definition of FF used, particularly given that broadly defined measures would vary between individuals as perceptions of what constitutes FF vary by socio-economic characteristics.

Additionally, it can be demonstrated that a higher expenditure within the ABS HES may actually reflect the purchasing of healthier foods.

The Gastrointestinal Symptoms Evaluation for Nutritional Supplements (GSENS): Preliminary development and validation

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Background

Long-chain omega-3 polyunsaturated fatty acid (LCn-3PUFA) supplements are becoming increasingly used in clinical trials. Gastrointestinal disturbances are commonly reported following the consumption of these supplements, which may lead to poor compliance. There are currently no gastrointestinal symptom rating scales designed to specifically measure the frequency and severity of these symptoms.

Objective

To examine the frequency and severity of gastrointestinal symptoms in children and adolescents.

Design

Eighteen children and adolescents aged 8-18 yrs rated their mood, behaviour and the frequency and severity of 11 gastrointestinal symptoms commonly reported in clinical trials of nutritional supplements over the previous week. Gastrointestinal symptoms were grouped according to severity.

Outcomes

Principal Components Analysis indicated that gastrointestinal symptoms could be grouped according to three levels of severity: minor (reflux, bloating, cramping, flatulence and constipation), sub-acute (urgency to pass stool, nausea and loose stool) and acute (pain, diarrhoea and vomiting). Participant scores on the GSENS were positively related to anxiety ($r = 0.49$, $p = 0.040$) and depression ($r = 0.75$, $p < 0.001$) and negatively related to quality of life ($r = -0.53$, $p = 0.024$).

Conclusions

The frequency and severity of gastrointestinal symptoms were successfully recorded using the GSENS. Future trials need to monitor gastrointestinal symptoms in larger numbers of participants prior to and following the consumption of LCn-3PUFA supplements. Monitoring of gastrointestinal symptoms may lead to more accurate assessment of the long-term benefits of nutritional supplements, including LCn-3PUFA.

Dairy intakes in Australian children and adolescents: Nutrient contributions and comparison to dietary recommendations

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Background

The 2007 National Children's Nutrition and Physical Activity Survey (NCNPAS) provides data on the diets of young Australians, previously assessed in 1995, enabling assessment of current dairy intakes.

Objective

This secondary analysis aimed to quantify and assess the nutritional contribution of current dairy food consumption in Australian children and adolescents and compare this against national dietary guidelines to help identify potentially at risk groups and to inform evidence-based dietary interventions and messages.

Design

Dietary intake data from the 2007 NCNPAS was provided by the Commonwealth Department of Health and Ageing. Data from a single dietary recall for 4487 children aged two to 16 years were used to identify core versus non-core dairy, and to calculate serves of dairy consumed and dairy intake by fat type.

Outcomes

The reported consumption of milk products and dishes (MPD) declined with age, 97% of four to eight year olds consuming compared to 89% of 14-16 year olds. Males consistently consumed more MPD than females at all ages with the highest intakes reported by two to three year old children (median: 422 g males cf. 355 g females) and the lowest intakes reported by 14-16 year old females (median: 223 g). Dairy milk was the most common source of MPD (64-87% consuming) and also represented the largest volume of dairy consumed (median: males 237-309 g cf. females 62-258 g). Whole milk was the most commonly consumed milk type (40-72% of children). Dairy intake peaked between 0700 and 0900 hours corresponding with the peak in dairy milk consumption. MPD are the primary source of children's total intake of calcium (51-67%), iodine (51-70%) and saturated fat (30-48%). Inclusive of non-core sources, the median intake of dairy consumed was 2.0-2.6 serves (males) and 1.8-2.1 (females). Failure to meet minimum daily dairy serve recommendations decreased with age, 45-70% of males and 50-88% of females.

Conclusion

MPD provide a spectrum of macro and micronutrients essential for good health. Dairy milk is a primary source of calcium in the diets of young Australians. Most of this population, notably adolescent females, fail to meet daily dairy intake recommendations, understanding why will be a useful focus for future research.

A systematic review: validation of children's dietary fat intake using biomarkers

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Background

Accurate assessment of children's dietary fat intake is necessary to examine relationships between dietary intake and disease. Research in paediatric populations is limited as few validated dietary assessment tools exist. Blood fatty acids are nutritional biomarkers of dietary intake and can be used to objectively validate dietary assessment tools.

Objective

The aim of this study was to conduct a systematic review of studies that examined the association of dietary fat intake and fatty acid biomarkers in children.

Design

Relevant health databases were searched using key words for English language articles published from 1973 to March 2009. In addition reference lists of key articles were hand searched. Studies were assessed for quality and critically appraised using standardised tools prior to data extraction. Inclusion criteria were; children 0-18 years and dietary fat intake measured with a quantitative assessment tool compared to a blood fatty acid biomarker.

Outcomes

Of the 9821 articles identified; 74 were retrieved and 15 articles were reviewed. Limited studies assessed the relationship between fatty acid intake and biomarkers in children. Only one study analysed fatty acid biomarkers for the purpose of validating a dietary assessment tool. The most prevalent outcome measurements were from food records, and plasma phospholipids, triglycerides and cholesteryl esters. Many plasma fatty acids analysed showed a relationship to diet, the strongest were plasma fatty acids ∑PUFA (18:2n-6, 20:5n-3, 22:6n-3) ∑SFA, (14:0, 16:0 & 18:0.)

Conclusion

Plasma fatty acids in children are an objective measure for validation of dietary fat intake and show moderate to strong relationships to diet. Further studies should comprehensively measure and analyse dietary fatty acid intake in healthy children, using a longitudinal study design, food records and plasma triglycerides or cholesteryl esters.

Carbohydrate intake in gestational diabetes mellitus – should there be a minimum?

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Background

Moderation of carbohydrate (CHO) intake is part of the dietary management of gestational diabetes mellitus (GDM). Currently women with GDM at Royal Prince Alfred Hospital in Sydney are advised to include ~180–210 g of carbohydrate per day to ensure adequate intake of macro- and micronutrients. Due to the fear of post-meal hyperglycemia, which may lead to the initiation of insulin treatment, GDM women frequently restrict their CHO intake to below 180 g. The consequence of this 'over-restriction' on nutrient adequacy is unclear.

Objective

To investigate the effect of CHO intakes below 180 g on macro- and micronutrient intake, as well as on blood glucose control.

Design

Twenty-five women with GDM were asked to complete a three-day (including two weekdays and one weekend day) food record following their initial GDM nutrition education session. The women were provided with a 2D food model booklet to assist them with portion size estimation, and any ambiguous entries were clarified via discussion. Macro- and micro-nutrient intake, including total daily CHO intake and the distribution of CHO throughout the day were analysed using FoodWorks (Xyris Software). Intakes were compared to the Nutrient Reference Values for pregnant women. The women were also asked to measure their blood glucose level (BGL) four times a day (fasting, and one hour after each main meal) with a blood glucose meter.

Outcomes

Nearly half the women (48%) did not achieve the recommended daily CHO intake (mean CHO intake ± SD = 135 ± 37 g). Compared with those who achieved ≥ 180 g CHO per day (mean CHO intake ± SD = 205 ± 26 g), a higher proportion of CHO restrictors did not meet the recommended intake of fibre (92% vs 69%), and the estimated average requirement (EAR) of thiamine (50% vs 23%), vitamin A (25% vs 15%), vitamin C (17% vs 8%), magnesium (50% vs 15%) and calcium (33% vs 23%). There was no difference in the mean and maximum 1h postprandial BGL between groups.

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Comparative genomics of lactation; identifying new roles for milk

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Milk has a major function to provide nutrition to the suckled young. However, it has additional roles including programming and regulating development of the suckled young, and an autocrine impact on the mammary gland so that it functions appropriately during the lactation cycle. This central role of milk is best studied in animal models, such as marsupials that have evolved a different lactation strategy to eutherians and allow researchers to more easily identify regulatory mechanisms that are not as readily apparent in eutherian species. For example, the tammar wallaby (*Macropus eugenii)* has evolved with a unique reproductive strategy of a short gestation, birth of an altricial young and a relatively long lactation during which the mother progressively changes the composition of the major, and many of the minor components of milk. In contrast to eutherians, there is a far greater investment in development of the young during lactation and it is likely that many of the signals that regulate development of eutherian embryos *in utero* are delivered by the milk. Inappropriate timing of these signalling events may result in either limited or abnormal development of the young, and potentially a higher incidence of mature onset disease. Delivery of these signals in marsupial milk requires the co-ordinated development and function of the mammary gland and it is now clear that milk includes proteins that can impact on these processes. With the increasing availability of sequenced genomes, the use of functional genomics, proteomics and bioinformatics we can exploit these models to identify milk bioactives in livestock and humans.

Omega-3 fatty acids and human heath – more than just a fishy tale?

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Current intakes of very long chain omega-3 (ω-3) fatty acids eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) are low in most individuals living in Western countries. A good natural source of these fatty acids is seafood, especially oily fish. Fish oil capsules contain these fatty acids too. Very long chain ω-3 fatty acids are readily incorporated from capsules into transport (blood lipids), functional (cell and tissue) and storage (adipose) pools. This incorporation is dose-dependent and follows a kinetic pattern that is characteristic for each pool. At sufficient levels of incorporation EPA and DHA influence the physical nature of cell membranes and membrane protein-mediated responses, lipid mediator generation, cell signaling and gene expression in many different cell types. Through these mechanisms EPA and DHA influence cell and tissue physiology and the way cells and tissues respond to external signals. In most cases the effects seen are compatible with improvements in disease biomarker profiles or in health-related outcomes. As a result very long chain ω-3 fatty acids play a role in achieving optimal health and in protection against disease. Long chain ω-3 fatty acids not only protect against cardiovascular morbidity but also against mortality. In some situations, for example rheumatoid arthritis, they may be beneficial as therapeutic agents. On the basis of the recognized health improvements brought about by long chain ω-3 fatty acids, recommendations have been made to increase their intake. The plant ω-3 fatty acid, α-linolenic acid, can be converted to EPA but in humans conversion to DHA appears to be poor. Effects of α-linolenic acid on human health-related outcomes appear to be, at least in part, due to conversion to EPA.

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