PART ONE

FUNCTIONAL FOODS: CLINICAL STUDIES IN RELATION TO METABOLIC SYNDROME

A PILOT STUDY ON TREATMENT EFFECT OF TOMATO LYCOPENE AND SOY ISOFLAVONES ON INSULIN RESISTANCE

Wentong Pan, Himara Davila Arroyo, Manish Saraf, Manisha Chandalia, and Nicola Abate

The University of Texas Medical Branch, Galveston, TX, USA

Key Words: Tomato lycopene, insulin sensitivity, soy isoflavones

Background: Several biological antioxidants have been demonstrated to improve insulin sensitivity by stimulating the tyrosine phosphorylation of the insulin receptor and glucose uptake. As member of the antioxidant family, Lycopene/ Isoflavons derivative has been shown to increase insulin sensitivity in cell cultures of skeletal muscles and 3T3L1 adipocytes, and improve insulin resistance in animal models of type 2 diabetes of obese and lean Zucker rats. Data about the effect of oral administration of lycopene/Isoflavons on peripheral insulin sensitivity in human is still unknown.

Objective: The objective of the study is to assess the effect of 12 weeks treatment with tablets containing Tomato Lycopene, Soy Isoflavones or a combination of the two on insulin sensitivity in human subjects, as measured by changes in peripheral glucose disposal rate during euglycemic hyperinsulinemic clamp. The

hypothesis is that, a 12-week oral treatment with Lycopene derivative will increase peripheral insulin sensitivity in humans

Methods: This is a randomized pilot study in 20 patients, 18-40 years of age, non-diabetic with insulin resistance. The duration of the study is 16 weeks in its entirety. There are a total of 3 study visits and a required follow-up phone call to monitor for serious adverse experiences. Eligible patients are randomized to 1 of 3 treatments that they receive daily: Tomato Lycopene, Soy Isoflavones, or a combination of two. Insulin sensitivity is measured using a ―gold standard‖ method, a manual euglycemic hyperinsulinemic clamp technique. Results from clamps performed at baseline are compared with measurements at the end of treatment. Main outcome variable is the change in insulin-mediated total body glucose disposal rate (Rd-value).

Results: Preliminary data from 16 subjects who already completed the 12 weeks study shows daily lycopene/ isoflavones administration had no effect on food intake or body weight and no change was found in liver/ kidney function based on plasma ALT/AST and creatinine level. Lycopene/ Isoflavones showed 10-15% reduction in plasma LDL cholesterol level in 50% study subjects who has baseline LDL above 100 mg/dL. 8 subjects received combination treatment showed average of 11% improvement on their insulin-mediated total body glucose disposal rate with average post-treatment Rd of 4.7 ± 1.7 vs. baseline Rd of

4.2 ± 1.0 mg/dL/kg body weight, while there were variations among individual subject in the effect of oral administration of single formula of Lycopene or Isoflavones.

Conclusion: Oral administration of combination of Lycopene and Isoflavones showed moderate improvement on glucose and lipid metabolism in human subject. As a dietary supplement with minimized undesirable side effects, combination treatment with Lycopene and Isoflavones has demonstrated the potential to be a safe and effective non-pharmacological alternative treatment to improve insulin resistance and metabolic syndrome. This may provide a new approach for prevention of both diabetes and cardiovascular disease with resulting benefit to patients and to our population.

CAN NON-COMMUNICABLE DISEASES BE PREVENTED?

Marie Benton

New Zealand Tertiary College, New Zealand

Key Words: long finned eel, omega-3, non-communicable disease prevention

Background: The reason for the sudden onset of non-communicable diseases in Māori; when prior to their urbanization in the 1950s and 60s non-communicable diseases did not exist. This is not a cure for Type II diabetes mellitus and other non-communicable diseases; and I do not attempt to develop a pharmacotherapeutic formula for Type II diabetes mellitus prevention through dietary omega-3 intake. Therefore this proposition is not a pharmacological medical science approach focusing on the action of omega-3 ingestion or on the properties or characteristics of an omega-3 diet. Indeed pharmaceutical drugs do play an important role in treating diseases especially communicable infectious diseases, recent medical research suggests that chronic non-communicable diseases could be prevented through diet and lifestyle choices.

Methods: Ten year study to ascertain that regular consumption of the long finned eel prevented the development of non-

communicable diseases because of the essential fatty acid omega-3 and lethicin that they contain. Scientific chemical assay testing of the eel validated the presence of the unsaturated essential fatty acid omega-3. This assayed testing was repeated to assess the stability and quality of fresh and smoked eel. The significance of omega-3 initially has been established from a comprehensive literature review of Medical Clinical Trials specific to the omega-3 benefits that validated their benefits.

There was a ten year Interview Data Gathering Key Informant Study that initially commenced with fifty participants questioned with specific structured questions; that then divided into two smaller groups of nine participants. One group regularly gathered and ate the long finned eel and the other group never ate any native eel.

Results: The first group participants are all still living and aged over 80 years; but all the participants in the second group have all passed prematurely.

Conclusion: A ten year study of Waikato hapū supported the research finding in my doctoral thesis that regular consumption of the long finned eel and holistic practice through established lifestyle choices is associated with the prevention of Type II diabetes.

CLINICAL BENEFITS OF A PHYTOCHEMICAL-ENRICHED MEDICAL FOOD ON CARDIOMETABOLIC RISK FACTORS IN WOMEN WITH METABOLIC SYNDROME

Robert H Lerman, Mark McIntosh, Wadie Najm, Maria Luz Fernandez

Functional Medicine Research Center, Metagenics, Inc. Gig Harbor, WA, USA

Key Words: metabolic syndrome, cardiovascular disease, Mediterranean diet, low-glycemic- load, hops, rho iso-alpha acid, acacia, proanthocyanidin, soy, phytosterol, medical food.

Background: Lifestyle change is recognized as the first line of intervention in the management of metabolic syndrome (MetS). Whereas diet is acknowledged to be a key component, the most efficacious dietary pattern has not been determined. Research supports benefits of both a Mediterranean diet and a low-glycemic-load (LGL) diet. In two studies, we combined these dietary approaches using a LGL, Mediterranean-style diet. In one arm only, we added a soy and phytosterol-containing medical food to determine whether it would improve outcomes. The first of these studies examined overweight and obese postmenopausal women with high LDL cholesterol. Arm 1 was randomly assigned to consume a calorie-restricted, Mediterranean-style, LGL diet and a soy/phytosterol-containing medical food. Arm 2 consumed a similarly calorie-restricted, low-fat, American Heart Association

(AHA) diet. Both arms participated in equivalent aerobic exercise programs. Compared with those on the AHA diet, women on the Mediterranean-style, LGL diet with medical food lost more weight and had greater improvements in several cardiovascular disease (CVD) risk markers, including triglyceride/HDL-C ratio, suggesting a potential effect on MetS. It was unclear whether benefit was due to the diet or to the medical food. Prior to further human studies, extracts from natural products were tested in a lipogenesis 3T3-L1 cell model. Two candidate compounds, rho iso-alpha acids from extract of hops and proanthocyanidins from extract of Acacia nilotica bark and heartwood, were found to increase lipogenesis, glucose uptake, and adiponectin secretion, and to reduce IL-6 secretion. Subsequent studies indicated that these molecules inhibited protein kinases involved in insulin resistance. Studies in the db/db mouse model indicated that the hops and acacia molecules reduced serum insulin and glucose to the same order of magnitude as rosiglitazone or metformin. In a randomized trial of overweight and obese men and women with MetS and high LDL cholesterol, effects of a lifestyle program consisting of the Mediterranean-style, LGL diet with no caloric restriction and aerobic exercise (MED) was compared to that of the same lifestyle program with addition of a soy/phytosterol-containing medical food in combination with a hops and acacia extract supplement (PED). Results indicated a substantial weight loss despite lack of caloric restriction. Multiple cardiac risk markers improved in the PED arm compared with the MED arm.

Of note was a 43% net resolution of MetS in PED vs. 22% in MED. In a subset analysis, subjects at high risk with both MetS and LDL-C >160 mg/dL in the PED arm achieved lower CVD risk after 12 weeks of intervention whereas those in the MED arm experienced little benefit.

Objective: We initiated a multicentered clinical trial in overweight and obese women with MetS to investigate effects on CVD risk of a Mediterranean-style, LGL diet, with or without a soy protein-based medical food containing microencapsulated natural compounds derived from hops and acacia extracts.

Methods: Eighty-nine women recruited from 3 university study sites were instructed to consume the specified diet. Forty-four were randomly assigned to the control arm and 45 to the medical food arm. The medical food contained soy protein, soy-derived phytosterols, rho iso-alpha acids from hops, and proanthocyanidins from Acacia nilotica. No caloric restriction was imposed during the 12-week study, and exercise was maintained at baseline levels.

Results: At 12 weeks, both arms lost similar amounts of body weight (~ 1 lb/week) and experienced significant improvements from baseline in waist circumference, triglyceride levels, blood pressure, total cholesterol, LDL-C, non-HDL-C, and apolipoprotein B (apoB). Compared with control, the medical food arm had greater reductions in total cholesterol, LDL-C, non-HDL-C, apoB, and apoB/apoA-I (P = 0.019, 0.017, 0.016, 0.005,

and 0.041, respectively). At the conclusion of the intervention, 20 women (44.4%) in the medical food arm and 14 women (31.8%) in the control arm no longer met criteria for MetS.

Conclusion: A soy- and phytochemical-enriched medical food enhanced the benefits of a Mediterranean-style, low-glycemic-load diet leading to improvement in multiple cardiovascular disease risk factors in women with MetS.

GLYCEMIC INDEX, ANTHROPOMETRIC NUTRITIONAL STATE AND TOTAL CONSUMPTION OF ENERGY, MACRONUTRIENTS AND FIBERS BY ELDERYLY PEOPLE IN BRAZILE: A CASE STUDY

Dolinsky M., Mantovani H.H.R., Dores S.M.C. Azeredo V.B., Regent F., Moreira

Federal Fluminense University. College of Nutrition. Department of Nutrition and Dietetics, Rua Mário Santos Braga, Campus do Valonguinhos Niteroi, Rio de Janeiro, Brazil

Key Words: Elderly, Diabetes, Dietary Fiber.

Background: Populational aging has caused a gradual raise in cases of chronic non-communicable diseases.

Objective: Assess the glycemic index, the anthropometric nutritional state and total consumption of energy, macronutrients and fibers of elderly people in the city of Niteroi, Brazil.

Methods: The anthropometric measures (Body Mass Index - BMI and waist circumference) and food consumption behavior (24-hour diary) of 22 elderly subjects of both genders were analyzed. The glycemic measurement was carried out with an ACCU-CHEK Active glucose meter.

Results: The mean age of the subjects was 70.3 ± 5.4 years and mean BMI was 27 ± 5,4 Kg/m2, indicating that most elderly were

either overweight or obese (35% of the sample was overweight, and 30% obese). Inappropriate glycemic indexes and high values of waist circumference were observed (mean circumference of 92cm for women and 108cm for men). The mean fiber consumption was below the recommended daily value.

Conclusion: The results were consistent with the literature on the nutritional profile of the elderly population. Although the mean consumption of energy and macronutrients were within the recommended limits, the mean fiber intake was of only 15g/day — way below the recommended intake of 25 to 30g/day. This was a relevant finding, considering that an adequate consumption of fibers is considered to be an important aid to weight loss and treatment/control of overweight/obesity — a prevalent problem within the studied sample — besides being linked to the homeostasis of the glycemic index. The profiling of a target group may offer important subsidies towards a more effective nutritional intervention, and the assessment of the clinical and nutritional profiles of the elderly made in this study allowed a better understanding of this populational stratum. The sample analyzed (mostly women) presented a high rate of obesity, high concentration of abdominal adipose tissue, inadequate control of the glycemic index and low intake of fibers.

METABOLIC SYNDROME AND HISPANIC CHILDREN: AN EXPLORATORY CASE STUDY

Felix Barron, M. Bracey, J. Visser, Candace Schroer, Sara Gilbertson, Jerry Marsh, A. Wingate, Luke Shelton,Florencia Barron, NP.

Clemson University
Food Science and Human Nutrition, Clemson, SC, USA

Key Words: 24 hr food recall, Hispanic Children, metabolic syndrome, BMI, foods, eating habits

Background: Obesity and other metabolic syndrome symptoms are increasingly being cited as potential health problems in Hispanic children in the USA. Eating habits and physical activity are 2 major factors as causes.

Objective: To determine exploratory relationships among types of foods eaten, BMI, calorie intake, micronutrients and micronutrients.

Methods: 24hr food recall forms were designed and used to collect data from patients in a local clinic

Results: Data was collected regarding foods eaten in a particluar 24 hr period, the BMI of Hispanic Children between the ages of 7 and 14. Potential deficiency and excess in macronutrients, calories and micronutrients have been calculated.

Conclusion: At present, data analyzed indicates and confirm the expected direct relationship between calorie intake and amounts of food eaten. Other relationships between other parameters are in progress.

A COMBINATION OF L-ARABINOSE AND CHROMIUM LOWERS CIRCULATING GLUCOSE AND INSULIN LEVELS AFTER AN ACUTE ORAL SUCROSE CHALLENGE: A HUMAN CLINICAL TRIAL

Kaats GR[1], Leckie RB[2], Keith, SC[1], Keith PL[1], and Preuss HG[3]

[1]Integrative Health Technologies, Inc. San Antonio, TX, USA
[2]Business and Healthcare Consultants, San Antonio, TX, USA
[3]Georgetown University Medical Center, Departments of Physiology, Medicine and Pathology, Washington, DC, USA

Key Words: L-Arabinose, Glucose, Insulin, Dual Energy X-ray Absorptiometry

Background: To examine changes in capillary blood glucose and venous insulin levels after an oral sucrose challenge with and without simultaneous consumption of L-Arabinose and a patented version of chromium (LA-Cr).

Objective: The purpose of this study was to examine the effect that consuming an LA-Cr supplement would have on the capillary glucose and venous insulin responses to a sucrose challenge. The supplement contained both LA and a patented form of trivalent Cr.

Methods: On day-1, (Control) after a 10-hour overnight fast, baseline blood was drawn from 50 subjects who subsequently consumed 70

grams of sucrose and had their capillary blood glucose and venous insulin tested at predetermined time intervals. On day-2, (Treatment) the same procedure was repeated except that subjects also consumed LA-Cr in conjunction with the sucrose challenge to allow comparisons of changes from baseline for the two conditions. While there were no adverse effects reported during the acute phase, to assess safety over a longer time period, a pilot study was initiated in which 20 subjects completed baseline measurements and 10 randomly chosen subjects completed post-study measurements after consuming a daily serving of the supplement each day for four weeks.

Results:

Table 2: Changes from Baseline in Capillary Glucose and Venous Insulin Levels After a 70g Sucrose Challenge With and Without Simultaneous Consumption of LA-Cr for Pilot Study (N=20) and Clinical (N=50). *P* values are from repeated measures t-test and Area Under the Curve (AUC) as indicated.

Pilot Study N=20 (Glucose Only)					
Minutes from Baseline	**30**	**45**	**60**	**90**	**AUC**
% Difference Between Treatment vs Control	-22.3%	-26.0%	-43.2%	-43.5%	-31.4%
Significance Levels	P<0.007	P<0.001	P<0.001	P<0.031	P<0.0001
Clinical Study N=50 (Glucose Only)					
Minutes from Baseline (Glucose)	**30**	**45**	**60**	**90**	**AUC**
% Difference Between Treatment vs. Control	-19.1%	-26.1%	-24.8%	-27.1%	-18.4%
Significance Levels	P<0.01	P<0.001	P<0.01	P<0.05	P<0.0001
Clinical Study N=50 (Insulin Only)					
Minutes from Baseline (Insulin)	**30**		**60**		**AUC**
% Difference Between Treatment vs. Control	-11.9%		-28.3%		-28.3%
Significance Levels	NS		P<0.001		P<0.01

Compared to Control, changes from baseline glucose and insulin levels were significantly lower in the Treatment condition, averaging 18 to

28%. Dual Energy X-ray Absorptiometry- derived body fat measurements revealed a significant and negative relationship between glucose suppression and body fat, suggesting that increased amounts of LA-Cr may be required in subjects with high levels of body fat to obtain comparable glucose suppression.

Conclusions: Consumption of the LA-Cr supplement in conjunction with a 70-gram sucrose challenge led to a significant reduction of capillary blood glucose and venous insulin as compared to the sucrose challenge without the LA-Cr supplement. This finding may provide an important tool to reduce the adverse effects associated with elevated glucose and insulin levels.

Trial Registration: Clinical Trials.gov, NCT0110743

COMPARISON OF CHANGES IN BONE MINERAL DENSITY (BMD) IN WOMEN OVER-40 FOLLOWING THREE BONE HEALTH PLANS WITH A COMMON PLANT-SOURCED FORM OF CALCIUM AND VITAMIN D_3*

Kaats GR[1], Preuss HG[2], Croft HA[3], Keith SC[1], and Keith PL[1].

[1]Integrative Health Technologies, Inc., 4940 Broadway, San Antonio, Texas, USA; [2]Georgetown University Medical Center, Departments of Physiology, Medicine and Pathology, Wash., DC, USA; [3]Croft Research Group, San Antonio, TX, USA

Key Words: Bone mineral density, calcium, vitamin D_3, quality of life inventory

Background: The US Surgeon General's Report on Bone Health (SG) suggests America's bone-health is in jeopardy due to increasingly sedentary lifestyles, poor nutrition and inadequate health information. In response to this crisis, the SG issued a ―call to action‖ to develop bone-health plans incorporating components that: (1) improves nutrition, (2) increase health literacy and, (3) increase physical activity.

Objective: To conduct a Comparative Effectiveness Research study comparing changes in bone mineral density (BMD) in women over aged-40 with expected changes in two reference groups and those occurring in three different bone health plans, each containing a common plant-sourced calcium.

Table 1. Components of the three versions of bone-health plans

Ingredient or Component	Grp 1 N=35	Grp 2 N=102	Grp 3 N=35
Plant-souced Calcium (mg)	750	720	756
Magnesium (mg)*	65	72*	350
Vitamin D-3 (IUs of Cholecalciferol)	1,000	800	1,600
Vitamin K-2 as MK-4 (mg)	0	1.5	0
Vitamin K-7 as MK-7 (mcg)	0	0	100
Boron (mg)	0	0	3
Vitamin C (mg)	0	0	50
Strontium Citrate (mg)	No	680	680
Pedometer-based physicl activity plan	No	Yes	Yes
Health Literacy Information	No	Yes	Yes

***72 mg naturally occurring plus magnesium carbonate**

Methods: A total of 172 females over 40 years of age volunteered to participate in one of the three sequential open-label treatment groups summarized in Table 1. All groups completed the same baseline and ending DEXA tests of bone density, 43-chemistry blood test panels, and 84-item Quality of Life Inventory (QOL). Since Grp-1 took only the plant-sourced calcium and Vitamin D_3 for one year while Grps 2 and 3 followed their plans for 6 months, comparisons were made by converting all data to mean annualized percent change. Subjects in the three groups were classified as ―compliant‖ or ―partially compliant‖ based on their reports of product usage. Comparisons were also made between the treatment groups and two age-adjusted expected groups: a non-intervention group and a group derived from a review of previously published studies on non-plant sources of calcium.

Results: There were no significant differences between three

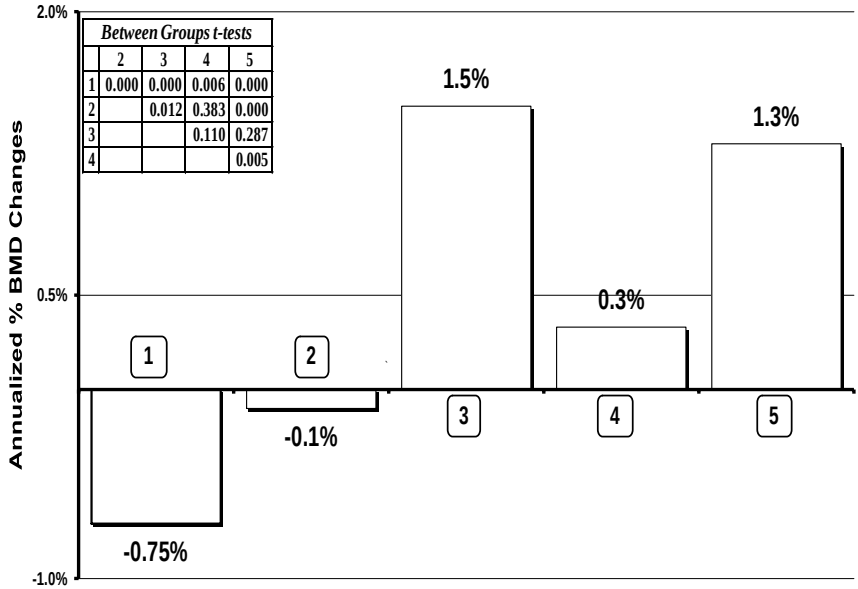
groups on baseline measurements of BMD, weight, age, or % body fat suggesting the treatment groups were statistically similar at baseline. Mean annualized changes in BMD are shown in Figure1 along with between-group t-test comparisons. All three treatment groups had significantly greater increases in BMD as compared expected changes. Groups 3 and 5 also had greater increases that the expected changes for non-plant sources of calcium reference group. Both groups. In all three groups, increases in BMD were greater in compliant, as opposed to partially compliant sub-groups. For all three groups, there were no statistically significant differences between baseline and ending blood chemistry tests or the total *Quality of Life* score derived from summing responses to all 84 inventory items. Nor were there any significant differences in baseline BMD between those who completed per protocol (PP) and those who were lost to attrition.

Conclusions: Taking either the plant-sourced calcium with Vitamin D3

by itself or in conjunction with either three-component bone health plan was associated with significant increases in BMD. Increased compliance was associated with greater increases in BMD as was modifying the bone health supplement with different amounts and types of nutrients, while holding all other components of the Plan constant. No adverse effects were reported in either group nor were there significant changes in blood chemistries or self-reported quality of life.

Trial Registration: ClinicalTrials.gov NCT01114685

Figure 1. Comparison of Annualized Age-adjusted Expected and Measured Annualized Changes in Bone Mineral Density (BMD) for Women Over 40-Years of Age Following Three Different Bone-Health Plans



1 = Age-adjusted expected change without treatment interventions
2 = Age-adjusted expected changes for non-plant sourced calcium/Vit D3 supplements (derived from literature review)
3* = Changes after taking only a plant-sourced form of calcium with Vitamin D3
4* = Changes for all subjects following the 3-component bone-health plan #1
5* = Changes for all subjects following the 3-component bone-health plan #2
**Components and ingredients are shown in Table 1.*

IMPACT OF DIETARY INTERVENTION WITH A FUNCTIONAL FOOD SUPPLEMENT TO COMBAT ANEMIA.,THE BLOOD IRON METABOLIC DISORDER AMONG THE COFFEE PLANTATION LABOURERS

Vasantha Esther Rani, Associate Prof and Head

PG Department of Home Science with food biotechnology,Fatima College,Madurai, S. India

Key Words: Dietary intervention, fumctional food, Spirulina, hemoglobin

Background: Indian economy greatly relies on agriculture. Agriculture is set to play a more dynamic role in the economy. The present study focuses on the nutritional status with special reference to the blood iron profile of manual coffee plantation labourers belonging to Thandikudi, Kodaikannal,

The outcome of this study on the dietary intervention based on nutritional picture of coffee plantation labourers will have a nation wide application because of the simplified , stable coffee plantation operations throughout India.

Objective: To design, implement and evaluate the impact of dietary intervention for the nutritional metabolic disorder which is directly related to productivity.

Methods: Experiments were carried out in three phases. In the first phase, the personal background of the labourers comprising of

the information on the socio economic background, their physical characteristics, anthropometry and physical fitness levels was assessed. In the second phase, the nutritional and health status of the labourers, through dietary survey comprising 24 Hour Food Recall record and weighment of cooked food consumed for three consecutive days, clinical and biochemical profile were studied. The blood iron status was estimated using the indices - Haemoglobin (Hb), Serum Iron, Transferrin Saturation, Serum Ferritin, Total Iron Binding Capacity (TIBC) and Unsaturated Iron Binding Capacity (UIBC). The serum iron was measured with the total iron binding capacity, from which the transferrin saturation was calculated. In the third phase, dietary intervention was implemented with a nutrient rich nutraceutical food supplement - Spirulina incorporated soup. The supplement! ation was extended for a period of 120 days. The subjects were grouped into Control and the Experimental group .The Control Group was given plain soup and the Experimental Group was administered with soup incorporated with Spirulina.

The impact of intervention on the biochemical, nutritional and physiological profile of the labourers was reassessed in similar working conditions.

Results: A deficit of 0.1 mg to 0.3 mg of iron intake per kg body weight in the male and female labourers when compared to their RDA The clinical pictures reveal that 94 per cent suffered from anaemia, 56 per cent showed anaemic signs of spooning of nails

(koilonychyia) , 36 per cent of the subjects had glossitis (mouth ulcer), 16 per cent suffered from bleeding gums, 4 per cent had cheilosis (ulceration of tongue) and 28 per cent had angular stomatitis.

The average haemoglobin level was 8.6 ±1.12g/dl for men and 8.1 ±1.01g/dl for women which is 28 and 37 per cent less than normal . Serum iron ranged from 40.3 to 55.0 mcg/dl in men and 37.2 to 49 mcg/dl in women. Serum ferritin level ranged from 9.4 to 15.5ng/ml (mcg/L) for male labourers and 8.3 ± 1.0 mcg/l for female labourers. The mean percentage of transferrin saturation was 9.8 per cent ± 1.7 and 7.6 per cent ± 1.7 The liver iron stores were in the range of 84 to140 mg and 63 to 99 mg for male and female respectively.

Dietary intervention 5g of Spirulina in 150 ml of soup had significantly improved the blood iron profile. Among the male labourers, 10 per cent who suffered from severe anaemia with 7.9 g.dl-1 haemoglobin before intervention were shifted to moderate levels of 9.4 g.dl-1, 30 per cent who suffered from mild anaemia were shifted to normal ones and among 60 per cent of male labourers who were moderately anaemic, 50 per cent reverted to normalcy.

Conclusion: In the present study it was observed that the habitual dietary intake showed deficiency in consumption of iron rich foods and the clinical picture revealed the signs and symptoms of anaemia The blood iron profile - Haemoglobin (Hb), Serum Iron,

Transferrin Saturation, Serum Ferritin, Total Iron Binding Capacity (TIBC) and Unsaturated Iron Binding Capacity (UIBC) were greatly influenced by dietary intervention with the functional food Spirulina incorporated soup The blood haemoglobin content increased by 21 per cent i.e from 10.9 to 13.3 g.dl-1 .Serum iron, serum ferritin and transferrin levels showed a consistent and pro¬gressive rise while TIBC and UIBC reduced resulting in a positive iron balance.

EFFECT OF OMEGA-3 FATTY ACID ON PLASMA LEVELS OXIDATIVE STRESS IN HEMODIALYSIS PATIENTS

Hamid Tayebi-Khosroshahi, Jalil Houshyar, Ali Tabrizi, Amir-Mansour Vatankhah, Neda Razzagi Zonouz, Reza Dehghan-Hesari

Department of Internal Medicine, Dialysis Center, Imam Reza Hospital, Tabriz University of Medical Sciences, Tabriz, Iran

Keywords: Oxidative stress, Omega 3 fatty acid, Hemodialysis

Objective: Oxidative stress in chronic renal failure patients and particularly in hemodialysis patients have been suggested major role in pathogenesis of atherosclerosis. We evaluated omega-3 fatty acids effects on oxidative and anti oxidants factors in hemodialysis patients.

Research Designs and Methods: In a clinical trial study, hemodialysis patients were randomly put and studied into case and control groups. Omega-3 capsule (1 gr) were given three times a day for case group and control group received placebo at the same shape of capsule and the same doses, both for 2 months. Blood samples were taken from patients before and at the end of the study and were examined about oxidative stress. Malondialdehyde (MDA) was determined using the colorimetric method, Glutathione peroxidase (GPx) and superoxide dismutase (SOD)

activity were assessed using the UV-Vis spectrophotometric technique Randox kit and Ferric Reducing Antioxidant Power (FRAP) was assessed on plasma used of school of life sciences methods**.**

Results: 75 hemodialysis patients including 37 patients in case and 38 in control group completed the study. Anti oxidants factors including Glutathione peroxidase, superoxide dismutase and Ferric Reducing Antioxidant Power were significantly increased in case group who received omega-3 fatty acids but in control group there was no significant difference. Malondialdehyde (MDA) levels were reduced with significantly difference in compared with control group.

Conclusions: the present study revealed that the supplementation with omega-3 fatty acids may result in better anti oxidation status in hemodialysis patients**.**

ASSOCIATION BETWEEN BODY MASS INDEX CLASSIFICATION AND DIABETES (TYPE 2) IN VETERANS

Angoorani P and Heidari M

Foundation of Martyrs and Veterans Affairs, Tehran, Iran

Key Words: Body mass index, Obesity, Diabetes, Veterans

Introduction: One of the most important factors for various diseases, such as diabetes type 2, is obesity, a nutritional burden and an independent factor which increases mortality. Veterans due to their life style and low level of physical activity, have more risk of obesity. The aim of this study was to assess the association between various degrees of obesity and diabetes type 2.

Materials and Methods: In a descriptive cross-sectional study, 108 over weight and obese veterans referred to clinic of diet therapy of Shahid Beheshti University during 2009 were studied. Weight, height and waist circumference, were measured and body mass index was calculated in patients. Subjects were divided into 4 groups according to their BMI. Demographic data and health and disease status such as diabetes were collected through a questionnaire. Statistical analyses were performed by using spss 17.

Results: Mean BMI was 36.3±7.4 kg/m² in patients. According to BMI classification, 16.7% of patients were overweight (25≤BMI<30) and the rest were obese which categorized to 3 grades: 35.1% grade 1 (30≤BMI<35), 16.7% grade 2 (35≤BMI<40) and 31.5% grade 3 (BMI≥40). 38 patients (35%) were diabetic (type 2). Diabetes type 2 were significantly higher in group 4 (BMI≥40) of patients. ($p<0.05$).

Conclusion: Extreme obesity (BMI≥40) and diabetes type 2 has a high prevalence in over weight and obese veterans. There should be more focus on balancing weight in this group. In this regard, such patients will benefit greatly from collaboration between nutritionists, endocrinologists and psychologists.

EFFECTS OF KOREAN RED GINSENG ON BISPHENOL A, AN ENVIRONMENTAL OBESOGEN

Mihi Yang

College of Pharmacy, Sookmyung Women's University, Seoul, Republic of Korea

Keywords: Korean red ginseng, obesity, diabetes, Bisphenol A

Background: Bisphenol A [2,2-bis(4-hydroxyphenyl) propane, BPA], an endocrine disrupting chemical, is suspected to induce endocrine and metabolic disorders, e.g. aging-related obesity and diabetes. Particularly, benzoquinone derivatives, metabolites of BPA, have been suspected to produce reactive oxygen species, which accelerate ageing. Among human population, young women are very sensitive to obesity or aging for beauty as well as health purposes. Thus, they look for anti-obesity or anti-aging materials and try to protect exposure to obesogens or oxidative stress. The Korean Red Ginseng (KRG) has been used as a main resource in Oriental medicine and recently showed some desirable effects on diabetes, atherosclerosis and oxidative stress.

Objectives: The goal of this study is to investigate whether KRG protects exposure to BPA, an obesogen.

Methods: We performed a clinical trial in Korean women (N=13; age, 23.50±1.79 yrs) for 2 weeks treatment of KRG (9 capsules

≈2.7g/day) and studied pharmacokinetic changes in levels of urinary BPA and malondialdehyde (MDA), an oxidative stress biomarker. One spot urine before breakfast was collected at 0, 4, 7, 14 days from each subject. We used reverse phase-HPLC/FLD and UVD methods for analyses of urinary BPA and MDA, respectively. Limit of detection (LOD) of BPA was 0.2 ng/ml. Urinary creatinine was also analyzed to adjust urine density with HPLC/UVD. Regression analyses were performed to study changes of BPA and MDA during the KRG intake period.

Results: Median of urinary BPA was 2.27 ug/g creatinine and their BPA exposure levels were similar to those, which were previously obtained in Korean adults. Urinary BPA levels were positively associated with MDA levels ($\beta = 0.16$, and $p = 0.08$). Thus, we confirmed BPA-induced oxidative stress. There were decreases in levels of MDA ($\beta = -0.06$, $p = 0.12$) rather than urinary BPA ($\beta = -0.04$, $p = 0.57$) during the KRG intake period.

Conclusion: Our pilot study showed some potential that KRG is chemopreventive to BPA via anti-oxidative mechanisms rather than pharmacokinetic effects (e.g. inhibition of BPA- absorption or -rapid detoxification) of KRG on BPA.

IMPLEMENTATION AND EFFECT EVALUATION OF A COMMUNITY-BASED GRANDPARENT AND GRANDCHILD INTERACTIVE NUTRITION EDUCATION PROGRAM

Mingzi Li, Lin Zhang

School of Nursing, Peking University, China

Key words: Grandparent and grandchild interactive, Nutrition education, Community, chronic disease prevention

Background: Dietary behavior is one of the most important factors that have impact on chronic disease. Family is the key place where people consume food and where dietary behavior forms and happens. Families with three generations are very common in Chinese urban communities, especially in those families which have preschool-age and school-age children. Middle-aged and elderly people in communities are usually the grandparents of these families, who are not only the food buyers and makers for the whole families, but also the main caregivers of the children. Their nutrition knowledge and behaviors impact not only their own health but also the whole family members'.

Objectives: To explore a community-based grandparent and grandchild interactive nutrition education program targeted to both middle-aged and elderly people and children to promote the knowledge and attitude of nutrition and healthy food, healthy

dietary behavior to prevent chronic diseases, and evaluate whether the program is more effective than standard nutrition education in improvement of residents on knowledge, attitude of nutrition and healthy food, dietary behavior and nutrition related health status.

Methods: A prospective experimental study design was adopted. On a voluntary basis, 5 primary care centers in Beijing had been recruited and randomised (cluster randomisation) with the result of 2 centers as intervention group and the other 3 centers as control group. 23 middle-aged and elderly people and 23 children were recruited in intervention group, and 78 middle-aged and elderly people and 87 children were recruited in control group. Based on *Knowledge-attitude-behavior model*, *Social Cognitive/Learning theory* and *Family theory*, comprehensive evidence-based reviews of studies and *Chinese Residents Diet Guide and Balenced Food Pyramid*, a community-based vivid and interactive nutrition education program had been developed and implimented in intervention groups during 6 months. Residents in control groups received standard nutrition education in primary care setting. Data of the subjects' nutrition knowledge, attitude and dietary behavior were collected via questionnaires and interview a pre and post intervention.

Results: 1. After 6-month education program, the scores of knowledge, attitude and KAP total of the adolescents in interactive education group were higher than the control group（P<0.01）; the

behavior score was higher than the control group but with no significant difference（P=0.077）. The scores of knowledge, attitude, dietary behavior and KAP total of the middle-aged and elderly people in interactive group were higher than the control group（P<0.01）.

2. After 6-month education program, the dietary behavior improved both in children and middle-aged and elderly people. Of the children, the intake rate of fried and puffed food, sweet, dried fruit and vegetables was lower than the control group（P<0.05）; the behaviors of ―often look up the nutritional label‖ and ―never buy food when hungry‖ increased（P<0.05）; the intake rate of tea drinks decreased（P<0.05）. Of the middle-aged and elderly people, the intake rate of milk products and legume products were higher than the control group（P<0.05）; the rate of ―never buy food when hungry‖ increased（P<0.05）.

3. There were no statistical significant differences between the two groups of children on the time of class, transportation, housework, sedentary, exercise and ourdoor activities（P>0.05）. The housework time median of middle-aged and elderly people in the interactive education group was higher than the control group at the baseline（z=-2.211, P=0.027）; there were no differences between the two groups on the housework and leisure time（P>0.05）.

4. There was no significant difference of body weight change between the intervention group and the control group（P >0.05）.

5. Five themes were induced through depth interview by using categorization: The community-based grandparent and grandchild interactive nutrition education program produced active changes among the residents; Multi-dimension of interactions; Enlightenments of the program; Effects of the program on the community nurses; The popularity of the program in communities.

Conclusions: The community-based grandparent and grandchild interactive nutrition education program had beneficial effects on both middle-age and elder people and children in urban community setting and could help the residents to improve their nutrition knowledge level, attitude and dietary behavior, and promote the healthy family dietary environment.

METABOLIC SYNDROME AMONG OBESE PATIENTS ATTENDING THE MEDICAL CLINICS OF THE THREE TEACHING HOSPITALS AT SANA'A CITY, YEMEN

Mohammed Ahmed Bamashmoos, Abdul Wahed, Abdul Jabar, Al Serouri

Community Health Department, Faculty of Medicine and Health Sciences, Sana'a University, Yemen

Key Words: Metabolic syndrome, obesity, cardiovascular diseases, diabetes mellitus, Yemen.

Background: Yemen faces major challenges in improving the health status of its population as itis entering an epidemiological transition with rising non-communicable diseases e.g. obesity, diabetes and cardiovascular diseases (CVDs).

Objective: We designed this study to find out the prevalence of Metabolic Syndrome (MS) and its components among obese Yemeni patients.

Methods: All obese (waist circumference >102 cm in male and >88 cm in female) attending the outpatients medical clinics at the three teaching hospitals in Sana'a city, were examined and their blood pressure (BP), fasting samples of plasma glucose, triglycerides, and HDL cholesterol were measured. The prevalence of MS obtained based on the Adult Treatment Panel III and

presence of at least 3 of the following: systolic BP ≥130 mm Hg and/or diastolic BP ≥ 85 mm Hg or on treatment for high BP, fasting glucose ≥110 mg/dl or on diabetes treatment, triglycerides ≥150 mg/dl, and HDL cholesterol <40 mg/dl in men and <50 mg/dl in women

Results: 200 obese were identified during study period with an overall MS prevalence of 46%. The metabolic co-morbidities were raised BP (68%), high triglycerides (66%), reduced high density lipoprotein (64%), and raised fasting blood glucose (40%).

Conclusion: Prevalence of MS is high among obese Yemeni patients and high BP was the commonest co-morbidity. These findings highlight an urgent need to develop strategies for prevention, detection, and treatment of MS that could contribute to decreasing the rising incidence of CVD and diabetes.

PHYSICAL EXERCISE IS MORE EFFECTIVE THAN HORMONE REPLACEMENT THERAPY IN IMPROVING LIPID PROFILE STATUS IN POSTMENOPAUSAL WOMEN

Sultana N, Ferdous T, Jahan N

Department of Physiology, Sir Salimullah Medical College, Mitford, Dhaka, Bangladesh

Key Words: Dyslipidemia, postmenopausal women, physical exercise, hormone replacement therapy

Background: Dyslipidemia in postmenopausal women is a health hazard all over the world. Physical exercise and hormone replacement therapy may play a significant role in the management of dyslipidemia among postmenopausal women. But physical exercise is the safest, easiest, less expensive and without having any complications for lipid lowering factor in postmenopausal women.

Objective: to observe the effects of physical exercise and hormone replacement therapy on serum lipid profile in postmenopausal women.

Methods: this study was done in the Department of Physiology Sir Salimullah Medical College (SSMC), Dhaka, Bangladesh from 1st January 2009 to 31st December 2009. Total 90 postmenopausal women, age ranged from 50-60 years and 30 premenopausal apparently healthy subjects, age ranged from 20-30 years was

included in the study. Among 90 postmenopausal women, 30 were performing regular physical exercise, 30 were taking hormone replacement therapy and another 30 were sedentary postmenopausal women. Lipid profile such as total cholesterol, HDL-C, LDL-C, TG and fasting blood sugar of all the subjects were estimated in Physiology laboratory, SSMC, Mitford, Dhaka, Bangladesh.

Results: mean serum TC, LDL-C and TG were significantly ($p<0.001$) higher and mean HDL-C was significantly ($p<0.001$) lower in sedentary postmenopausal women than those of premenopausal women, postmenopausal women with regular physical exercise and postmenopausal women with hormone replacement therapy. Again mean TC, LDL-C and TG were lower and mean HDL-C was higher in postmenopausal women with regular physical exercise than those of postmenopausal women with hormone replacement therapy. Moreover all the menopausal symptoms were comparatively lower in postmenopausal women with regular physical exercise than those of postmenopausal women with hormone replacement therapy.

Conclusion: physical exercise was more effective than that of hormone replacement therapy in lowering lipid levels as well as improving postmenopausal symptoms. Physical exercise is safe, easy to perform, less expensive and has no complications. So, regular physical exercise can be used as effective alternative to hormone replacement therapy and thereby reduces cerebro-vascular disease (CVD) risk and improve the daily lifestyle.

EFFECTS OF *DICHROSTACHYS GLOMERATA* SPICE ON CARDIOVASCULAR DISEASES RISK FACTORS IN NORMOGLYCEMIC AND TYPE 2 DIABETIC OBESE VOLUNTEERS

Dieudonne Kuate, Blanche C. Etoundi, Judith L. Ngondi, and Julius E. Oben

Laboratory of Nutrition and Nutritional Biochemistry, Department of Biochemistry, Faculty of Science, University of Yaoundé, Yaoundé, Cameroon.

Keywords: Dichrostachys glomerata, cardiovascular disease, diabetes

Background: Previous *in vivo* and *in vitro* experimental studies have shown Dichrostachys glomerata *(DG)*, a spice used in western Cameroon, to have potential antioxidant and hypoglycemic properties.

Objective: The purpose of the present study was to evaluate the effects of orally administered *DG* on various cardiovascular disease risk factors in obese normoglycemic and obese type 2 diabetic human subjects.

Methods: The study was an 8 week randomized, double-blind, placebo-controlled design with obese and obese/diabetic participants (20 males, 72 females, ages 25–65). The subjects were

randomly divided into four groups: 2 normoglycemic obese groups (active; placebo) and two type 2 diabetic obese groups (active; placebo). Capsules containing the active (400 mg. *DG*) or placebo formulation were administered 30–60 minutes before lunch and dinner throughout the study period. A total of 7 anthropometric and hemodynamic as well as 7 biochemical measurements were taken at the beginning of the study and after 4 and 8 weeks of treatment. All diabetic patients maintained their prior lifestyle intervention and dietary control for the duration of the study.

Results: Compared to the two placebo groups, the two active groups showed statistically significant differences on all 14 variables between Weeks 0 and 8. These included body weight, BMI, waist and hip circumference, body fat, blood pressure, blood cholesterol, triglycerides, glucose, and glycosylated hemoglobulin.

Conclusion: The results confirm the hypothesis that *DG* appears to reduce cardiovascular disease risk factors in obese normoglycemic and obese type 2 diabetic human subjects.

EFFECTS OF BROCCOLI SPROUT POWDER ON FASTING SERUM GLUCOSE AND LIPID PROFILES IN TYPE 2 DIABETIC PATIENTS

Z. Bahadoran, P. Mirmiran, F. Hoseinpanah, G. Asghari, M. Mehran, and F. Azizi

Research Institute for Endocrine Sciences, Shahid Beheshti Univercity of Medical Sciences, Tehran, Iran

Key Words: Type 2 diabetes, Broccoli sprout, Randomized clinical trial

Background: Broccoli sprout is a good source of health promoting compounds. Little is known about the direct effect of broccoli sprout on human health, but in vitro and in vivo data have provided some indication that young broccoli sprouts improves the abnormal metabolism of lipids in diabetic condition.

Objective: The aim of this study was to investigate the dose-effect of broccoli **sprouts powder (BSP) on fasting serum glucose (FBS) and lipid** profiles in type 2 diabetic patients. The present study was approved by the ethical committee of the Research Institute for Endocrine Sciences of the Shahid Beheshti University of Medical Sciences.

Methods: Eighty-one type 2 diabetic patients were randomized into three groups to receive one of the following: group A (BSP

10g/d, n=26), group B (BSP 5g/d, n=29) and group C (placebo, n=26) for 4 weeks. Serum levels of FBS, lipids and lipoproteins were measured before and 4 weeks after intervention. Seventy-tow subjects completed the follow up. No serious adverse events were reported. The mean energy and nutrient intakes remained constant during study period. Mean concentration of fasting serum glucose was significantly decreased in group A and B after 4 weeks as compared with baseline values (144±56 v.188±68 mg/dl, p=0.002 and 126±51 v. 146±56 mg/dl, p=0.006, respectively). Mean level of total cholesterol (TC) were significantly decreased in group A and B after 4 weeks as compared with baseline values (167±51 v.198±45 mg/dl, p=0.001 and 153±25 v. 176±31 mg/dl, p<0.001, respectively). Mean of LDL cholesterol were significantly decreased in group A and B after 4 weeks as compared with baseline values (98±35 v.115±38 mg/dl, p<0.001 and 85±24 v. 101±27 mg/dl, p<0.001, respectively). Mean level of triglyceride (TG) were significantly decreased in group A (141±82 v.166±87 mg/dl, p<0.05) but not in group B. The treatment effect was statistically significant increase in HDL cholesterol in the group A as compared with B and C (47.9±13.9 v. 40.2±7.8 v. 41.1±9.5, P=0.002). The treatment effect was statistically significant reduction in atherogenic index of plasma defined as log [TG /HDL-C] in the group A as compared with B and C (0.42±0.33 v. 0.50±0.20 v. 0.60±0.23, P=0.004). A linear regression model demonstrated dose response effect of BSP on mean differences of TG (β = -2.9, p<0.05), TC (β= -1.7, p<0.05) and HDL (β= 0.9,

p=0.006). There were no significant changes in fasting serum glucose and lipid profiles after 4 weeks of intervention in controls.

Conclusion: It is concluded that administration of broccoli sprout powder for 4 weeks had favorable effects on fasting serum glucose and lipid profiles in type 2 diabetic patients.

THE EFFECTS OF OMEGA-3 FATTY ACIDS SUPPLEMENTATION ON BLOOD LIPID PROFILE OF TYPE 2 DIABETIC PATIENTS

Pooya S[1], Guéant-Rodriguez[1], Jalali D[2], Toorang F.[2]

[1]Inserm U954, Medical faculty and CHU of Nancy, Nancy University, Nancy, France
[2]Department of Nutrition and Biochemistry, School of Health, Tehran University of Medical Sciences, Tehran, Iran

Key Words: Type II diabetes, Omega-3 fatty acids, Hemoglobin A1C, Lipid Profile, Apolipoprotein-A, Apolipoprotein-B100

Background and Objective: One of the most important mortality causes in diabetic patients is atherosclerosis. Omega-3 fatty acids decrease plasma triglyceride level, but there is uncertainty regarding their effect on total cholesterol level. In recent years, much research has been done to clarity roles of omega-3 fatty acids on the lipid profile of diabetic patients, but the results have not been consistent. The objective of this study was to determine the effect of omega-3 fatty acids on the HbA1c and lipid profiles.

Materials and Methods: A randomized double-blind placebo-controlled clinical trial was conducted on 81 type 2 diabetic patients. The patients were randomly assigned to either the case or the control group. Each subject received, per day, 3 omega-3 fatty acid capsules (EPA=1548 mg, DHA=828 mg and 338 mg other

omega-3 FAs) or a placebo for a period of 2 months. The serum fasting blood sugar (FBS), HbA1c, TG, HDL-c, LDL-c, total Cholesterol, malon-di-aldehyde (MDA), Apolipoproteins A and B levels were measured. Independent t-test for comparing differences between the treatment and control groups and paired t-test for comparing differences before and after intervention in the treatment and control groups was used. The body mass index (BMI) calculated and nutrient intakes were estimated using the 24h dietary recall.

Results: 90 persons including 49 women and 41 men with a mean age of 56.38 ±9.24 years old participated in this study. Average of body mass index (BMI) was 28.08 ±4.31 and food intake did not show any difference between the case and the control group before and after supplementation.

The 2-months supplementation led to statistically significant decreases in the serum levels of HbA1c (-8.2%), ApoB100 (-17.1%), triglyceride (-26.5%) and the TG: HDL ratio (-23%) in the case group with a p-value of ≤ 0.001. On the contrary, the level of HDL and the apoA:apoB ratio increased, but the changes were not statistically significant. There were no significant changes in other variables.

Conclusion: Consumption of omega-3 supplements (3gr/day) for 2 months can reduce the serum contents of HbA1c, Apo B100 and TG and the TG: HDL ratio in diabetic patients.

MONITORING PREMATURE INFANTS: NUTRITIONAL AND METABOLIC INDICATORS

Rebeca Monroy-Torres[1], Marisol López- López[1], Jaime Naves-Sánchez[2], Xochitl S. Ramírez-Gómez[1]

[1]Medicine and Nutrition Department, Division of Health Sciences. Campus Leon, University of Guanajuato, Mexico; [2]Maternal and Child Hospital SSG and UMAE, Mexico

Key Words: Premature infants, glucose, hemoglobin, weight gain

Background: Preterm infants have increased morbidity and mortality due to physiological immaturity and greater demands of growth. The nutritional intervention contributes to proper weight gain which translates into better growth and neurological development and prevent the onset of metabolic complications such as diabetes mellitus, hypertension and dyslipidemia.

Objective: To analyze and to monitor of nutritional and metabolic indicators in preterm infants.

Methods: We performed an analytical, transversal and comparative study in 100 infants, 30 to 36 weeks gestation, which had data on weight, length and head circumference at birth in Maternity Hospital de Leon, Guanajuato. They were evaluated from 5 to 9 months of age. The nutritional indicators were weight, length, head circumference and mid arm circumference and

metabolic indicators were: glucose, hemoglobin, cholesterol, triglycerides.

Results: The mean age was 7.3 ± 1.4 months. The average nutritional indicators were: weight of 6812 ± 1301 g, length 43.7 ± 4.6, head circumference of 45 ± 1.4 and mid arm circumference of 13.5 ± 2.1, and metabolic indicators were: glucose 100 ± 18 mg / dL, hemoglobin of 11.8 ± 1.2 g / dL, cholesterol 128 ± 32 mg / dL, triglycerides of 95.4
± 29.8. They had a little weight gain (60%), but with greater weight gain in the rest.

Conclusions: Premature infants showed a little weight gain as well as other anthropometric data; also in some of them, greater weight gain was observed according to the recommendations. Metabolic indicators likely reflect risk, so the monitoring is recommended in metabolic and anthropometric indicators, especially in the largest increases in weight, all this for prevent the presence of diseases such as diabetes and hypertension.

EFFECT OF DAILY INTAKE OF PREBIOTIC (FRUCTOOLIGOSACCHARIDE) ON WEIGHT GAIN AND REDUCTION OF ACUTE DIARRHEA AMONG CHILDREN IN A BANGLADESH URBAN SLUM

Shafiqul Alam Sarker, Sadako Nakamura, Mohammad Abdul Wahed, Yukiko Wagatsuma, Tsuneyuki Oku, and Kazuhiko Moji

International Centre for Diarrhoeal Diseases Research, Bangladesh (ICDDR,B), Mohakhali, Dhaka, Bangladesh and Shiebold University of Nagasaki, Japan

Key Words: Prebiotic, Fructooligosaccharide (FOS), weight gain, diarrhea, RCT

Background: Fructooligosaccharide (FOS) is a typical prebiotic agent. Feeding prebiotic agents have been shown to be useful in preventing certain disease conditions by selectively stimulating the growth of bifidobacteria and lactobacilli in the gut. There is currently insufficient evidence to support their use to prevent diarrhea in children

Objective: to evaluate the prebiotic effect of daily intake of an FOS-containing isotonic solution on body weight gain and reductions in diarrhea episodes in children living in an urban slum in Bangladesh

Methods: a randomized, double-masked, placebo-controlled study

on a total of 150 children aged 25-59 months. Sixty-four children in the FOS group received 50 ml of isotonic solution with 2 g of FOS added, and 69 children in the placebo group received a similar-looking solution with 1 g of added glucose once daily over six consecutive months. Body weights were measured on alternate days, and height and arm circumference were measured once every month, and the children's mothers were interviewed to obtain history of diarrhea, stool consistency and contents, other morbidities and antibiotic treatment.

Results: The number of diarrhea episodes was less in FOS group compared to the placebo group. However, the difference was not statistically significant. The total mean days with diarrhea as well as each episodes of diarrhea were significantly shorter in the FOS group (3.3 vs. 6.3 d, p=0.039 and 2.5 vs. 3.2 d, p=0.008, respectively). The body weight gain during the six-month period in the FOS group (0.86±0.55 kg) and the placebo group (0.89±0.48 kg) was not significantly different, and so were the height and the mid-arm circumference (table 1)

Conclusion: A daily intake of FOS shortens duration of diarrhea episodes, but is not useful in promoting weight gain or in preventing diarrhea. Further studies with optimizing the doses of FOS are needed to define better therapeutic effects in diarrhea in children

Table 1: Comparison of diarrhea episodes between FOS and Placebo group

	FOS (n=63)	Placebo(n=67)	p
Number of diarrhea episodes	1.3 ± 1.6	2.0 ±2.8	.098
Cumulative diarrhea days	3.3 ±4.9	6.3 ±10.8	.039
Duration of diarrhea days/episodes	2.5 ± 1.8	3.2 ±1.4	.008
Body weight gain (kg)	0.85 ±0.55	0.89 ±0.48	NS

PHYSICAL EXERCISE IS MORE EFFECTIVE THAN HORMONE REPLACEMENT THERAPY IN IMPROVING LIPID PROFILE STATUS IN POSTMENOPAUSAL WOMEN

Sultana N, Ferdous T, Jahan N

Department of Physiology, Sir Salimullah Medical College, Mitford, Dhaka, Bangladesh

Key Words: Dyslipidemia, postmenopausal women, physical exercise, hormone replacement therapy

Background: Dyslipidemia in postmenopausal women is a health hazard all over the world. Physical exercise and hormone replacement therapy may play a significant role in the management of dyslipidemia among postmenopausal women. But physical exercise is the safest, easiest, less expensive and without having any complications for lipid lowering factor in postmenopausal women.

Objective: to observe the effects of physical exercise and hormone replacement therapy on serum lipid profile in postmenopausal women.

Methods: this study was done in the Department of Physiology Sir Salimullah Medical College (SSMC), Dhaka, Bangladesh from 1st January 2009 to 31st December 2009. Total 90 postmenopausal women, age ranged from 50-60 years and 30 premenopausal apparently healthy subjects, age ranged from 20-30 years was

included in the study. Among 90 postmenopausal women, 30 were performing regular physical exercise, 30 were taking hormone replacement therapy and another 30 were sedentary postmenopausal women. Lipid profile such as total cholesterol, HDL-C, LDL-C, TG and fasting blood sugar of all the subjects were estimated in Physiology laboratory, SSMC, Mitford, Dhaka, Bangladesh.

Results: mean serum TC, LDL-C and TG were significantly ($p<0.001$) higher and mean HDL-C was significantly ($p<0.001$) lower in sedentary postmenopausal women than those of premenopausal women, postmenopausal women with regular physical exercise and postmenopausal women with hormone replacement therapy. Again mean TC, LDL-C and TG were lower and mean HDL-C was higher in postmenopausal women with regular physical exercise than those of postmenopausal women with hormone replacement therapy. Moreover all the menopausal symptoms were comparatively lower in postmenopausal women with regular physical exercise than those of postmenopausal women with hormone replacement therapy.

Conclusion: physical exercise was more effective than that of hormone replacement therapy in lowering lipid levels as well as improving postmenopausal symptoms. Physical exercise is safe, easy to perform, less expensive and has no complications. So, regular physical exercise can be used as effective alternative to hormone replacement therapy and thereby reduces cerebro-vascular disease (CVD) risk and improve the daily lifestyle.

INVESTIGATION ON THE CORRELATIVITY OF METABOLIC SYNDROME AND CHRONIC KIDNEY DISEASE AMONG POPULATION OLDER THAN 40 YEARS IN HENAN PROVINCE

Yan Shan

Medicine Department/Nursing College of Zheng Zhou University, Clinic Medicine Department of Nursing College in Zheng Zhou University, Zheng Zhou, China

Key Words: Kidney disease, Chronic, metabolic syndrome, prevalence, correlativity

Background: Risk factors for the initiation and the progression for CKD have been identified gradually, owing to the continuous epidemiological studies. Apart for diabetes and hypertension early known, the metabolic syndrome (MS) including obesity, hyperlipidemia, hyperuricemia and other factors, is also increasingly been confirmed to associate with CKD. Study shows that metabolic syndrome is a risk factor in the progression of CKD and the risk will increase with the number of MS. In addition, there is evidence that smoking is not conducive to the control of blood sugar in diabetic patients and may accelerate the progress of diabetic nephropathy. De Boer IH et al reported that there was a link between the concentration of cystitis C and the level of serum triglyceride and high density lipoprotein cholesterol. Mexico studies had demonstrated that low -education and low-income were related to the decline of creatinine clearance rate. Researchers have

also pay attention to the connection between CKD and the factors including alcohol, drugs, race, family history and level of education these years. Epidemiological data indicate that morbidity of hypertension has increased from 5.11% to 18.8% since 1950s and WHO survey of 2000 showed that 5.2 % of men and 5.3% of women suffered from diabetes In China .As increase of the two diseases mentioned above, the risk factors for CKD in China may similar to those in developed countries.

Objective: We initiated this study among population older than 40 years in Henan province, with the intention to determine the epidemiology of chronic kidney disease (CKD), investigate the correlativity of metabolic syndrome (MS) and CKD.improve the awareness and therapy rate of CKD. It will be helpful to health providers and medical professionals in controlling the pandemic of CKD effectively. And also important in evoking the establishing of effective prevention of CKD to control the pandemic of CKD.

Methods: 4156 residents were randomly selected by means of a stratified; cluster sampling. All subjects received a questionnaire. Each participant underwent weight、height、waist circumference、hip circumference and blood pressure measurements, using a calibrated scale.Indicators of kidney damage、blood suger and blood cholesterol were examined in our

laboratory. Data entry and managemant were performed by statistics.

Results: 1.4156 subjects were accepted to investigate,and Eligible data of 3981 subjects were enrolled in the study.Indicator of kidney disease was available for 95.70% of the participants examined. Age ranges from 40 to 90 years old and the sex ratio is 1.21 (male:female):1.The Prevalence of hypertension and diabetes mllitus in this population was 15.04%and 5.76%respectively.2.The crude prevalance of albuminuria,hematuria and reduced eGFR were 4.97%, 6.33%and 1.53%. After the adjustment of age and gender component, A1buminuria was detected in 4.51% of subjects, hematuria in 6.28%, reduced eGFR in 1.56%. Women had higher prevalence of albuminuria and hematuria than man(P <0.01), but adverse in reduced eGFR. Both prevalence of albuminuria and reduced eGFR are increased with age(P<0.01).3.The crude prevalence of CKD was 10.58%, while the standardized rate was 10.49%.it also increased with age($\chi 2$=16.107 P<0.01). 4. Both the prevalence of albuminuria and reduced eGFR were higher in the subjects with metabolic syndrome than those without(6.90%VS 4.65%, $\chi 2$=5.184 P<0.05 and 5.00%VS0.97%, $\chi 2$=51.148 P<0.01), and they all increased

with the numbers of MS combination($\chi 2$=211.638 P <0.0l).5.The awareness rate of CKD was only 9.50%,and just 8.31%in therapy.

Conclusion: 1.The prevalence of CKD was higher level among population older than 40 years in Henan province. 2. MS was a base risk factors of CKD,and the risk increased with the numbers of MS combination. 3. The awareness and therapy rate of CKD among adults in Henan province was far lower than that in western developed countries.

THE PREVALENCE OF METABOLIC SYNDROME AMONG 7-17 YEARS OLD CHILDREN AND ADOLESCENTS IN 8 PROVINCES IN CHINA

Yu Dongmei, Yang Xiaoguang, Yin Shian, Piao Jianhua, Zhao Liyun, Zhang Jian, Hu Xiaoqi, Meng Liping, Liu Aidong, Yu Wentao, Ma Guangsheng

National Institute for Nutrition and Food Safety, Chinese Center for Disease Control and Prevention, Beijing China, P.R.C

Key Words : metabolic syndrome, children , adolescents

Background: With the developing of society and economic in China, it has been shown in previous studies that the chronic diseases' risk factors are more and more clustered in children and adolescents. Early identification of metabolic syndrome in children and adolescents in later life is important.

Objective: To investigate the prevalence and distribution of metabolic syndrome among 7-17 years old children and adolescents in 8 provinces in China.

Methods: Analyses of cross-sectional data obtained from the Chinese Nutrition and Health Surveillance(2009) which conducted as a pilot study in 8 provinces in China in 2009. The prevalence and distribution of a metabolic syndrome among Chinese children and adolescents were defined using the criteria for IDF(International Diabetes Federation) (2007), Stephen Cook modified the criteria of

NCEP-ATPⅢ(National Cholesterol Education Program: Adult Treatment Panel III) (2003) for adolescents, and the criteria of NCEP-ATPⅢ modified the criterion of waist by waist circumference reference values for screening cardiovascular risk factors in children and adolescents (2009). The prevalence of metabolic syndrome was estimated overall, by sex, by region (urban or rural), by BMI. BMI was estimated using the criteria for overweight and obesity developed by WGOC (Working Group on Obesity in China). Anthropometric measure, laboratory assays, questionnaire survey and blood pressure were conducted by trained healthy technicians. Standardized rate was calculated using The fifth national population census of China in 2000.

Results: The study population was a sample of 2752 children and adolescents 7-17 years old from 8 provinces. The results of the prevalence and distribution of metabolic syndrome among 7-17 years old children and adolescents applying three criteria as following:

	IDF(2007) *		Stephen Cook modified the criteria of NCEP-ATPⅢ (2003)§		Modified the NCEP-ATPⅢ using Chinese reference values in children and adolescents (2009) #	
	N	% (Standardize	N	% (Standardize	N	% (Standardize

		d %)		d %)		d %)
Region						
Urban	14	1.5 (0.7)	55	3.5 (2.8)	149	9.6 (6.1)
Rural	14	2.0 (1.5)	43	3.6 (2.4)	106	8.9 (6.4)
Gender						
Male	18	2.2 (1.3)	53	3.8 (2.9)	131	9.4 (6.6)
Female	10	1.3 (1.2)	45	3.3 (2.2)	124	9.1 (6.0)
BMI(Chinese criteria)						
Normal	2	0.2 (0.02)	20	0.9 (0.3)	98	4.3 (2.4)
Overweight	4	2.5 (0.04)	20	8.0 (8.7)	67	26.8 (25.1)
Obesity	22	21.2 (27.9)	58	27.2 (29.1)	90	42.3 (39.7)
Waist(Chinese criteria)						
Non waist obesity	0	0	18	0.74*(0.3)	42	2.0 (1.2)
Waist obesity	28	15.0 (15.0)	80	25.72 (25.6)*	213	32.7 (24.7)
Total	28	1.7 (1.2)	98	3.6 (2.6)	255	9.3 (6.3)

**** IDF(2007) BMI(Chinese criteria) P<.0001, Waist(Chinese criteria) <.0001***

§ Stephen Cook modified the criteria of NCEP-ATPⅢ(2003): BMI(Chinese criteria) P<.0001

#Modified the NCEP-ATPⅢ using Chinese reference values in children and adolescents(2009): BMI(Chinese criteria) P <.0001, ,Waist(Chinese criteria) P <.0001

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§ Stephen Cook modified the criteria of NCEP-ATPⅢ(2003): BMI(Chinese criteria) P<.0001

#Modified the NCEP-ATPⅢ using Chinese reference values in children and adolescents(2009): BMI(Chinese criteria) P <.0001, ,Waist(Chinese criteria) P <.0001

Conclusions: The adult chronic diseases are now clustered in Chinese children and adolescents. As childhood metabolic syndrome likely tracks into adulthood, early identification is helpful to adopt effective interventions. In order to illuminate the importance of metabolic syndrome in Chinese children and adolescents, it's essential to establish the appropriate criteria.

THE PREVALENCE OF METABOLIC SYNDROME AMONG ADULTS IN 8 PROVINCES IN CHINA

Zhao Liyun, Yu Dongmei, Ma Guangsheng,et al
Zhao Liyun, Yu Dongmei, Yang Xiaoguang, Yin Shian, Piao Jianhua, Zhang Jian, Hu Xiaoqi, Meng Liping, Liu Aidong, Yu Wentao, Ma Guangsheng

National Institute for Nutrition and Food Safety, Chinese Center for Disease Control and Prevention, Beijing China, P.R.C

Key Words: metabolic syndrome, adult

Background: With the developing of society and economic in China, chronic diseases are increasing in Chinese adults over the past decades. Until recently, the metabolic syndrome has been regarded as a disease. The metabolic syndrome is associated with an increased risk of both diabetes and cardiovascular disease.

Objective: To investigate the prevalence and distribution of metabolic syndrome among adults in 8 provinces in China.

Methods: Analyses of cross-sectional data obtained from the Chinese Nutrition and Health Surveillance(2009) which conducted as a pilot study in 8 provinces in China in 2009. The prevalence and distribution of a metabolic syndrome among adults were defined using the criteria for CDS(Chinese Diabetes Society) (2007), IDF(International Diabetes Federation)(2004), and NCEP-

ATPⅢ(National Cholesterol Education Program: Adult Treatment Panel III)(2001). The criterion of waist was modified by waist circumference reference values for screening cardiovascular risk factors in Chinese adults. The prevalence of metabolic syndrome was estimated overall, by sex, by region (urban or rural), by age group, and by BMI. BMI was estimated using the criteria for WHO and for overweight and obesity developed by WGOC(Working Group on Obesity in China). Anthropometric measure, laboratory assays, questionnaire survey and blood pressure were conducted by trained healthy technicians. Standardized rate was calculated using The fifth national population census of China in 2000.

Results: The study population was a sample of 9182 adults from 8 provinces. The results of the prevalence and distribution of metabolic syndrome among Chinese adults applying three criteria as following:

	CDS (2007) *		IDF(2004)※		NCEP-ATPⅢ(2001)§	
	N	% (Standardized %)	N	% (Standardized %)	N	% (Standardized %)
Region						
Urban	694	12.7 (11.2)	1042	19.0 (17.2)	479	8.8 (7.8)
Rural	323	11.7 (11.4)	428	15.5 (12.4)	236	8.5 (5.7)
Gender						

Male	517	12.8 (11.1)	632	15.6 (11.9)	249	6.1 (4.)
Female	500	11.9 (11.7)	838	20.0 (16.6)	466	11.1 (9.1)
Age group						
18-	185	5.2 (7.5)	269	7.5 (8.3)	108	3.0 (3.0)
45-	448	16.3 (17.2)	623	22.7 (20.5)	312	11.4 (10.6)
60-	384	20.2 (15.1)	578	30.3 (22.9)	295	15.5 (11.2)
BMI						
WHO criteria						
Normal	75	1.4 (0.6)	306	5.9 (3.0)	145	2.8 (1.3)
Overweight	704	27.8 (26.4)	868	34.2 (29.1)	367	14.5 (10.4)
Obesity	238	45.7 (45.4)	296	56.8 (49.1)	203	39.0 (35.6)
China criteria						
Normal	45	1.0 (0.3)	169	3.9 (1.7)	94	2.2 (1.0)
Overweight	510	19.0 (19.3)	696	26.0 (21.2)	263	9.8 (6.0)
Obesity	462	38.5 (36.5)	605	50.4 (45.0)	358	29.8 (27.4)
Total	1017	12.3 (11.4)	1470	17.8 (13.9)	715	8.7 (6.4)

**** CDS (2007): Age group P <.0001, WHO criteria P <.0001, China criteria P <.0001***

※IDF(2004): Region P =0.0117; Gender P = 0.0272; Age group P <.0001; WHO criteria P<.0001; China criteria P <.0001

§NCEP-ATPⅢ(2001): Region P = 0.0284; Gender P <.0001; Age group P <.0001; WHO criteria P <.0001; China criteria P<.0001

Conclusions: The cardiovascular disease risk factors are co-occurring in Chinese adults, especially in obese ones. There is urgent need for strategies to prevent the metabolic syndrome.

PART TWO

FUNCTIONAL FOODS AND METABOLIC SYNDROME: SCIENTIFIC CONSEPTS

THE CHANGES IN IRON AND ZINC CONCENTRATION IN LUNG AND KIDNEY TISSUES OF RABBITS DURING THE PROGRESSION OF ATHEROSCLEROSIS

Mohamed Anwar K Abdelhalim

Department of Physics and Astronomy, College of Science, King Saud University, Riyadh, Saudi Arabia

Key Words: Iron; zinc; atomic absorption spectroscopy; high fat diet; atherosclerosis; rabbits.

Background: Hyperlipidemia or a high level of serum triacylglycerol and cholesterol is a risk factor for premature atherosclerosis. Today's challenge is to effectively incorporate risk-reduction strategies into daily clinical practice and reduce the personal and societal burden of cardiovascular disease. The severity of atherosclerosis can be markedly influenced by iron (Fe) and zinc (Zn) overload or deficiency in aorta tissues of the rabbits. Fe has participated in diverse pathological processes by catalyzing the formation of reactive oxygen free radicals. Zn has reduced oxidative damage and the risk of cardiovascular disease.

Objective: The changes of Fe and Zn in lung and kidney tissues of rabbits during the progression of atherosclerosis have not been documented before. In addition to the authors would like to convey an important relationship between iron and zinc concentrations in lung and kidney tissues of rabbits and the progression of

atherosclerosis. Thus, the aim of the present study was to estimate the changes of Fe and Zn in lung and kidney tissues of rabbits fed on high fat diet (HFD) for a feeding period of 12 weeks.

Methods: The HFD group was fed a normal rabbit chow supplemented with 1.0% cholesterol plus 1.0% olive oil for a feeding period of 12 weeks. Fe and Zn concentrations were measured in two types of tissue from control and HFD rabbits using atomic absorption spectroscopy (AAS).

Results: Comparing HFD rabbits to control rabbits, we found that the Fe concentration was significantly ($p < 0.05$) increased with percentage normalized changes of 95.29% in lung and 7.08% in kidney tissues of HFD rabbits compared with control rabbits; while the Zn concentration was decreased with percentage normalized change of 3.61% in lung and significantly ($p < 0.05$) decreased with percentage normalized change of 71.40% in kidney of HFD rabbits compared with control rabbits.

Conclusion: The findings of this study can be summarized as follows; the highest percentage normalized change of increase of Fe was 95.29% in lung tissue accompanied by the lowest percentage normalized change of decrease of Zn 3.61% in lung tissue; while the lowest percentage normalized change of increase of Fe was 7.08% in kidney tissue accompanied by the highest percentage normalized change of decrease of Zn 71.40% in kidney

tissue. This study indicates that high concentration of iron in lung is accompanied by low concentration of zinc while high concentration of zinc in kidney is accompanied by low concentration of iron. This study suggests that the increase in Fe concentrations in lung and kidney tissues may accelerate atherosclerosis through the production of free radicals; while the decrease in Zn concentration may delay or prevent atherosclerosis through reducing lesion Fe content thus it acts as a protective factor against atherosclerosis. This study also suggests that zinc may be highly excreted from the kidney which can be used as an important risk factor during the progression of atherosclerosis. Zinc may replace iron in the aortic tissue of rabbits, thus this study suggests that further supplemental zinc diet may retard and prevent the progression of atherosclerosis. Thus, the changes of Fe and Zn concentrations in lung and kidney tissues of rabbits are closely related to the progression of atherosclerosis.

ANTIOXIDENT STABILITY OF ANIGERIAN OKRA SEED FLOUR BY IN VITRO DIGESTION

Adelakun OE, Ade-Omowaye BIO[a], Adeyemi IA[a], Van de Venter M[b]

[a]Department of Food Science and Engineering, Ladoke Akintola University of Technology, Ogbomoso, Oyo State, Nigeria,
[b]Department of Biochemistry and Microbiology, Nelson Mandela Metropolitan University, Port Elizabeth, South Africa

Keywords: Okra seed, *in vitro* digestion, antioxidants activity and gastrointestinal tracts

Background: Antioxidant is any substance that reduces oxidative damage caused by free radicals which ultimately results into chronic diseases. Dried seeds of okra have been proposed as a new high protein crop for the temperate zone and the tropics. Studies have been conducted on okra seed oil, defatted meal, nutritional value of okra seed flour and its antioxidant property (Karakoltsidis and Constantinides, 1975; Oyelade *et al*., 2003, Araptisas, 2008, Adelakun *et al*., 2009). Since high antioxidant levels in foods don't necessarily translate into levels found in the body and the potential health benefits of these antioxidants ultimately depend on how they are absorbed and utilized in the body, this study was carried out to determine the stability of okra seed antioxidant under gastrointestinal tract by in vitro digestion.

Methods: Known and equal (100g) quantities of okra seeds were soaked in 300ml of water for 6, 12, 18, 24, 36 and 48 hours respectively. The soaked seeds were then washed and dried in a cabinet dryer at 45 ± 2^{o}C for 48 hours to moisture content of 10-13% (Adeyemi *et al*., 1988). The seeds were milled and sieved to obtain a flour fraction of less than 250 µm. Determination of the free radical scavenging activity (FRSA) in the 1,1-diphenyl-2-picrylhydrazil radical (DPPH) assay and *in vitro* digestion were determined as reported by Adelakun, *et al*., 2009.

Results: Digestion *in vitro* with enzymatic extracts mimicking conditions in the gastrointestinal tract showed that the amount of antioxidants released by the okra matrix into the human intestine was higher than that from measurements made on the usual aqueous-organic extracts for all samples and also higher than those released in the gastric phase (Fig 1). From the results, it could be inferred that most antioxidants would be available in the intestinal phase of the gastro-intestinal tracts. The acidic conditions in the stomach and enzymatic hydrolysis in the duodenum are reported to likely increase the solubility and activity of antioxidant (Perez-Jimenez and Saura-Calixto, 2005). Moreover, significant increasing level of antioxidant activity ($P< 0.05$) was observed in all analyses which reached a peak during the 18th hour of soaking after which a significant decrease ($P< 0.05$) occurred. Other important antioxidant fractions can also be released during fermentation (Fardet *et al*., 2008). Nagah and Seal (2005) also demonstrated the significant influence of *in vitro* gastrointestinal digestion on the increased antioxidant release from

whole-grain cereal foods. Digestion is believed to partially hydrolyse the hydrolysable phenolics. Similarly the digestion of starch and protein may also increase the release of polyphenols. These results are particularly important since they suggest that antioxidants exert their effect within the digestive tract, providing an environment that protects the intestinal epithelium from pro-oxidative compounds.

Conclusions: The health benefit of this seed was established due to its stability under gastro intestinal tracts. This work has also been able to widen the seeds' scope of utilization other than re-generational purposes which it is known for in Nigeria. We therefore conclude that the flour obtained from the seed could find useful application in food formulations.

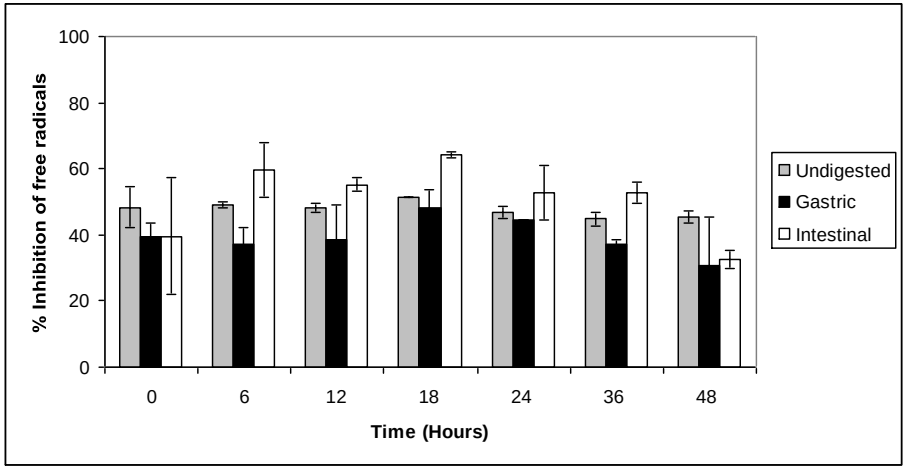

Fig 1. *In vitro* antioxidant activity of soaked okra seed Flour
Values are the means ± SD of triplicate measurements

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EFFECT OF HIGH FAT AND SUCROSE DIET INDUCED OBESITY ON RENAL AND OXIDATIVE MARKERS IN RATS: ROLE OF GARCINIA ADMINISTRATION

Amin K.A, Kamel H.H, Eltawab M.A

Biochemistry Department, Faculty of Veterinary Medicine, Beni-Suef University, Beni-Suef, Egypt and the Clinical Pathology Department, Faculty of Veterinary Medicine, Beni-Suef University, Beni-Suef, Egypt

Key Words: Sucrose, fat diet, renal, obesity oxidative markers, Garcinia.

Background: Obesity became a major health problem in the world and associated metabolic syndrome resulted from oxidative stress.

Objective: The objective of this study was to examine the effect of high sucrose (65%) and high fat (35%) diet to induce obesity on antioxidant defense system, biochemical changes in blood and tissue of control, non treated and treated groups by administration of Garcinia cambgia, and explore the mechanisms that link obesity with altered renal function.

Methods: Rats were fed a standard control for 10 wk or a diet containing 65% high sucrose (HSD) or 35% fat (HFD) for 8 wk, and then HFD group divided into two groups for the following 2 wks. One group was given Garcinia+HFD, the second only high fat, Also the HSD divided into two groups, 1st HSD+Garcinia and

2nd HSD. Blood and renal, visceral, prirenal, epidedimal adipose tissues were collect for biochemical measurements.

Results: HFD and HSD groups of rat showed significant increase in feed, energy intake, BW and BMI. Also significant increase weight of visceral, prirenal, epidedimal adipose tissue in HFD and HSD groups. HFD and HSD affect kidney by increasing serum urea, creatinine and uric acids levels and decreased level of nitric oxide (NO) and increased blood glucose, LDL, TG and TC and MDA. Moreover, renal catalase activities and MDA level were increased while NO was lowered. Glucose 6phosphate dehydrogenase (G6PDH) activities were significantly increased in HSD groups. These changes improved by Garcinia that decrease, oxidative stress and increased level of NO. There were a significant correlation between BMI and kidney function (Creatinine, urea and TG) and Oxidative marker (renal MDA and catalase).

Conclusion: Rats fed a diet with HFD or HSD showed, hypertriglyceridemia, increased LDL production, increased oxidative stress, renal alteration. Moreover, suggesting association between lipid peroxidation, obesity and nephropathy and Garcinia, ameliorate the damaging effects of the HFD or HSD and decrease feed intake, MDA level and decreased oxidative stress in renal tissues.

SCREENING OF ANTIBACTERIAL ACTIVITY ON SOME MEDICAL PLANT EXTRACTS

Belgasem A., Naffati K., Agha A., Enwige M, and Saad M

Department of Microbiology, Biotechnology Research Center, Twaisha, Libya

Key words: Staphylococcus aureus, artemisia, medicinal folklore

Background: the antimicrobial activity plant oils and extract has been recognized for many years ago, proximately 20% of plants found in the world have been submitted to pharmacological or biological test, Ethno botanical data have provide to be useful in the search for new antimicrobial agents and many of these compounds have been isolated from medical plants.

Objective: of the present study to find out if the plant extracts presented has ability to kill or inhibit the growing of the bacteria.

Methods: four local strains organisms namely; *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Salmonella Spp*., sterile metallic borer (6mm), DMSO, four crude extracts of *thymus vulgaris*, *peganum harmala*, *urtica dioica* and *artemisia*. Agar well diffusion method (cup-cut diffusion method), Standard bacteriological technique.

Results: the results based on zone of inhibition. *Artemisia*, was found active against *Salmonella Spp*. and *Escherichia coli*, Whereas, the *thymus vulgaris*, *peganum harmala*, *Artemisia*, is given good effective on *Staphylococcus aureus* only. On the other hand antibacterial activity of *urtica dioica* has no effective on all strains of bacteria which are presented.

CINNAMON EXTRACT INHIBITS THE OVERPRODUCTION OF THE POSTPRANDIAL APOLIPOPROTEIN B48-CONTAINING LIPOPROTEINS: EVIDENCE FROM EX VIVO AND IN VIVO STUDIES

Bolin Qin[1, 2], Richard A. Anderson[1]

[1]Diet, Genomics and Immunology Laboratory, Beltsville Human Nutrition Research Center, ARS, US Department of Agriculture, Beltsville, Maryland, USA
[2] Integrity Nutraceuticals International, Spring Hill, TN, USA

Key words: Cinnamon extract, insulin resistance, lipoproteins, enterocytes, insulin signaling

Background: Dyslipidemia, a component of the metabolic syndrome, is characterized by high levels of apolipoprotein B (apoB)-containing lipoproteins and ***plays an important role*** in ***metabolic syndrome development.*** There is accumulating evidence obtained from both *in vitro* and *in vivo* studies in animal models and humans, indicating that dyslipidemia associated with insulin resistant states results from overproduction of both intestinal and hepatic triglyceride-rich lipoprotein (TRL)s. Recent data also indicate that small intestine enterocytes, similar to the hepatocytes, overproduce lipoprotein particles in insulin resistance. Moreover, postprandial apoB48-containing particles not only enhance the production of hepatic lipoproteins through increased substrate delivery in the form of chylomicron remnants, but also delay the clearance of hepatic-TRLs. Cinnamon extract (CE) has been

shown to improve aspects of the metabolic syndrome in cell culture, animal and human studies. The pro-inflammatory cytokine, TNF-α, that is over expressed in obesity, insulin resistance, and the metabolic syndrome, stimulates the overproduction of intestinal apoB48-containing lipoproteins.

Methods and Results: *In vivo* oral feeding with a water extract of cinnamon (Cinnulin, 50mg per kg BW) inhibited the postprandial overproduction of apoB48-containing lipoproteins and serum triglyceride levels in rats and hamsters. In ex vivo ^{35}S labeling studies, CE (10 and 20μg/mL) inhibited the over-secretion of apoB48 induced by TNF-α treated enterocytes into the media. CE treatment decreased the mRNA expression of the inflammatory factors IL1□, IL6 and TNF-□, improved the mRNA expression of IR, IRS1, IRS2, PI3K and Akt1, inhibited CD36, microsomal triglyceride transfer protein (MTP), phosphatase and tensin homolog (PTEN) and enhanced the impaired sterol regulatory element-binding protein (SREBP)1c expression in TNF-□ treated enterocytes. Acute oral CE (50mg per kg BW) inhibited the increase in postprandial triglycerides and the overproduction of apoB48-containing lipoproteins in high fructose diet-fed insulin resistant rats. CE also inhibited the secretion of apoB48 in enterocytes isolated from fructose-fed hamsters, enhanced the impaired mRNA expression of intestinal insulin signaling, and down-regulated the over expression of MTP and SREBP1c mRNA levels. In Wistar rats fed a high-fructose diet to induce insulin

resistance, supplementation with an aqueous CE (50 mg/kg daily) for 8 weeks reduced blood glucose, plasma insulin, triglycerides and total cholesterol. Soluble CD36 and retinol binding protein 4 of plasma, which are reported as novel markers of insulin resistance, also were inhibited by CE consumption.

Conclusions: In summary, our data suggest that an aqueous extract of cinnamon effectively ameliorates circulating levels of apoB-containing particles partially mediated via regulation of the expression of multiple genes involved in inflammation, insulin sensitivity, and lipogenesis and may be beneficial in the control of dyslipidemia.

FRUCTOSE CONSUMPTION, OXIDATIVE STRESS, AND LIVER DAMAGE IN RATS

Botezelli JD[1], Dalia RA, Moura LP, Araújo MB, Ribeiro C, Ghezzi AC, Cambri LT, Voltarelli FA, and Mello MAR

São Paulo State University – UNESP, Department of Physical Education, Rio Claro – São Paulo, Brazil

Key words: oxidative-stress, fructose-rich diet, anti-oxidants

Background: Over the last three decades, the fructose consumption in developed countries raised 500% approximately. The timing of the increase in the prevalence of obesity coincides with the increased use of fructose in the diet. When consumed in great amounts, this carbohydrate can contribute to liver lipogenesis, thus triggering an increase in β-oxidation, and, consequently, generation of free radicals. Free radicals cause both cell damage and inflammation, which contribute to Non Alcoholic Fatty Liver Disease appearance (NAFLD). Earlier studies associated the NAFLD occurrence to metabolic syndrome. For this reason, it is important to investigate the implications of fructose-rich diet consumption on health.

Objective: This study aimed to evaluate the blood and liver levels of triglycerides and its effects on oxidative-stress, anti-oxidant responses and liver injury in rats fed on fructose-rich diet.

Methods: Sixteen weaned Wistar rats (28 days) were housed on a 12 h light/dark cycle at room temperature of 25ºC and fed on commercial pelletized diet until adulthood (120 days). At this point, they were distributed randomly into two groups with eight rats in each: Control (C) and Fructose (F). The C was composed by animals fed on balanced diet (AIN-93) whereas the F group by animals fed on a fructose rich diet (60% fructose) during 60 days. At the end of the experiment, the animals were killed by sodium thiopental administration in order to determine: a) serum triglyceride (TG) and TBARs concentrations, Aspartate aminotransferase (AST)/ Alanine aminotransferase (ALT) ratio, as well as the Catalase enzyme (CAT) activity, and b) liver triglyceride (TG) and TBARs concentrations and the CAT activity. The results were statistically analyzed by *student t-test* with significance levels established at 5%.

Results: The results are described in the table 1. As expected, the fructose-rich diet increased both liver and serum triglycerides concentrations. The increased liver lipid storage in the F group probably triggered β-oxidation that activated the lipid peroxidation, leading to liver injury. This hypothesis is supported by the high AST/ALT ratio observed in these animals. The anti-oxidant system of the F rats was down regulated at liver. This contributed to high TBARs levels and to liver injury. On the other hand, the serum CAT activity was increased after fructose-rich diet

administration; this finding may be a crossover response from other tissues to higher serum TBARs levels.

Table 1. Serum and liver parameters. Results are Mean± SD of 8 rats in each group

Groups	Serum				Liver		
	TG (mg/l)	TBARS (µM)	AST/ ALT	CAT (U/ml)	TG (nmol/mg)	TBARS (nM/mg)	CAT (mol/min.mg)
C	88.0± 19.6	16.4± 2.0	1.08± 0.4	42.2± 10.0	6.4± 1.9	0.3± 0.03	0.06± 0.01
F	268.3 ±35.*	22.2± 4.4*	2.8± 0.8*	68.6± 6.8*	16.2± 4.6*	0.35± 0.02*	0.02± 0.00*

C= Control; F= Fructose. *Significant difference compared to C group ($p \leq 0.05$ *student t-test*).

Conclusion: The results demonstrated that fructose-rich diet altered blood and liver levels of triglycerides. These alterations led to oxidative-stress, which was not fully counteracted by the antioxidant system. Further studies are required in order to better elucidate the actual mechanisms involved in liver damage of animals fed with a fructose-rich diet.

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INTERVENTION OF α-LIPOIC ACID AMELIORITES METHOTREXATE-INDUCED OXIDATIVE STRESS AND GENOTOXITY: A STUDY IN RAT INTESTINE

V.P. Dadhania, D.N. Tripathi, A. Vikramb, P. Ramarao, G.B. Jena

National Institute of Pharmaceutical Education and Research, Nagar, Punjab, India

Key Words: Methotrexate, α-Lipoic acid, Oxidative stress, Comet assay, Intestine, Rat

Background: Methotrexate (MTX) is an anti-metabolite, widely used in the cancer chemotherapy and rheumatoid arthritis. However, its long-term clinical use is restricted on account of its severe intestinal toxicity. The genotoxic effects of MTX have already been reported in both somatic and germinal cells employing chromosome aberration, micronucleus test and comet assay as the end points of evaluation. α-Lipoic acid (LA), a naturally occurring sulphydryl compound found in virtually all plants and animal species, is a potent antioxidant with high efficacy of chemo protection. It is also involved in the chelation of metal ions, regeneration of exogenous and endogenous antioxidants and repair of oxidized proteins. Further, it has been reported that LA increases the anti-apoptotic protein Bcl-2 in endothelial cells of male rat. Recently LA has gained considerable attention as a potential antioxidant in different pharmacological intervention studies. It appears that LA exerts its beneficial effects

in conditions where oxidative stress plays a critical role in the induction of cellular damage/target organ toxicity and hence it was considered for the present study and an attempt has been made to evaluate the possible protective effects of LA against MTX-induced GI toxicity.

Objective: The present study was aimed to investigate the intestinal toxicity of MTX and the possible protective effect of -lipoic acid (LA) on Sprague–Dawley rats.

Methods: MTX-induced intestinal toxicity was evaluated at the dose of 2.5 mg/kg for short-term (5 days treatment) and 1 mg/kg for long-term (5 days in a week for four consecutive weeks treatment) study. The possible protective effect of LA was evaluated in both short- as well as long-term study in a dose-dependent manner.

Results: MTX treatment induced diarrhea and mortality in rats, indicating its severe toxicity in the target organ of investigation, i.e., intestine. Further, the intestinal toxicity of MTX was assessed by evaluating different parameters of oxidative stress, DNA damage, cytotoxicity as well as histological changes. Immunostaining for p53 revealed higher genotoxic assault in the intestinal cells due to MTX treatment. Pretreatment of rats with LA led to significant decrease in the oxidative stress, DNA damage, cellular damage, inflammatory changes and apoptosis as determined by malondialdehyde level, glutathione level, comet

assay parameters, histological evaluation, immunostaining and terminal deoxynucleotidyl transferase-mediated dUTP nick end labeling (TUNEL) assay. Our results clearly indicate that LA treatment reduced MTX-induced oxidative stress and cytotoxicity. Further, LA treatment led to decreased DNA damage and cellular toxicity.

Conclusion: In the present investigation, we reported that LA pretreatment ameliorates MTX-induced intestinal toxicity in rat as evident from the protection against oxidative stress, decrease in DNA damage and protection of cellular morphology as well as improvement in the stool consistency and animal survival rate.

ANTIMICROBIAL ACTIVITY AND POLYPHENOL EXTRACT OF LIBYAN MEDICINAL PLANTS

Elfahem A., Elmeshri S., Abuzweda A., and Altomi A.

Biotechnology Research Centre, Tripoli, Libya

Key Words: Antibacterial activities, plant extracts, methicillin-resistant Staphylococcus aureus

Ethnopharmacological relevance: eight traditional Libyan plants were used for source of antibacterial against methicillin-resistant Staphylococcus aureus (MRSA).

Aim of the Study: To assess in vitro anti-MRSA activity of some Libyan herbal extracts.

Materials and Methods: The disc diffusion and minimum inhibitory concentration (MIC) techniques were performed to assess the antibacterial activity of methanolic extract.

Results: All the presented 8 plants showed anti-MRSA activity with MIC of 50mg/ml. - 25mg/ml. The most active antimicrobial plants were Cistus salvifolius Salvia officinalis, Pistacia atantica, Arbutus pavarii and Myrtus communis the other methanolic extract of Teucrium polium, Thymus capitellatus, and Euphorbia dendroides shows weak effect against MRSA.

Conclusions: Cistus salvifolius Salvia officinalis, Pistacia atantica, Arbutus pavarii and Myrtus communis can use as a source of antimicrobial against methicillin-resistant Staphylococcus aureus (MRSA). This finding warrants necessity of further investigation of this product of folk medicine.

HYPOLIPIDEMIC EFFECT AND ANTIOXIDANT PROPERTIES OF THE ETHANOLIC EXTRACTS OF *XYLOPIA AETHIOPICA* AND *OCIMUM GRATISSIMUM* IN TRITON WR-1339 INDUCED ACUTE HYPERLIPIDEMIC RATS

Etoundi Omgba CB, Kuaté D, Ngondi JL, Oben J.

Laboratory of Nutrition and Nutritional Biochemistry, Department of Biochemistry, BP 812, Faculty of Science, University of Yaoundé I, Cameroon.

Key Words: Xylopia aethiopica, Ocimum gratissimum, Hyperlipidemia, antioxidant activity.

Background: Hyperlipidemia, which represents a major risk for cardiovascular diseases, is on an increase worldwide. Though there are different medications prescribed to remedy this condition, the use of natural plant remedies is always preferred. *Xylopia aethiopica* and *Ocimum gratissimum* are spices commonly used in Cameroonian cuisine. This study evaluates the lipid lowering effect and antioxidant properties of the ethanolic extracts of these two spices on triton WR-1339-induced hyperlipidemic rats.

Methods: Hyperlipidemia was induced in three groups of rats (positive control and 2 tests groups) by a single intravenous injection of Triton WR-1339 (850mg/Kg). The test groups were treated three times per day with the extracts (400mg/Kg), 24 hours

after injection with triton. Free radical DPPH scavenging, ferric reducing antioxidant power assay (FRAP), folin Ciocalteu's phenol method, and ABTS (3-ethyl-benzothiazoline-6-sulfonic acid) assays of the extracts were also carried out.

Results: Triton induced a significant ($p<0.05$) rise in triglyceride levels, which was significantly decreased ($p<0.05$) by the extracts. These extracts also exhibited very strong antioxidant properties with all the four methods used.

Conclusion: Our results indicate that, ethanolic extracts of *Xylopia aethiopica* and *Ocimum gratissimum* extracts have hypolipidemic and antioxidant activities, which could play an important role in the management of metabolic syndrome.

INCIDENCE OF BRUCELLA SPECIESIN SLAUGHTERED FOOD ANIMALS AND ITS EDIBLE OFFALS IN BENI-SUEF, EGYPT

Fatma Ali, H.M. and *Mahdy, E.A.

Department of Food Hygiene, Faculty of Veterinary Medicine Beni-Suef University, Beni-Suef, Egypt
*Department of Pathology, Faculty of Veterinary Medicine Beni-Suef University, Beni-Suef, Egypt

Key Words: brucella melitesis, brucella abortus, liver, kidney, spleen, lung, heart, edible offal, Brucellosis.

Background: Edible offal is a delicacies food for people and also a good source of protein. Brucella is endemic disease in Egypt. At slaughter house regulations, condemned only spleen,lymph nodes and udder of serologically positive for brucella slaughtered animal. Other organs (liver,kidneys,lungs and hearts) pass for human consumption. According to food habits, these organs may be eaten raw or under cooked, so it constitutes a greater source of brucellosis in man through eating or handling in abattoir or at home.

Objective: This work was done to show the incidence of brucella in different slaughtered food animals (cattle, buffaloes, sheep and goats) at Beni-Suef city, Egypt. Also to show the incidence of brucella aborts and melitensis in edible offal as spleen, liver, kidney, lung, heart and lymph nodes as they may be responsible for a dangerous zoonotic disease in man brucellosis.The public health significance and control of brucellosis was discussed.

Methods: A total of 80 slaughtered animals,20 each of cattle, buffaloes ,sheep and goat.These animals were previously tested for brucella as they were positive and they were collected from Beni-suef abattoir. Each slaughtered animal was represented by liver, spleen, lung, kidney, heart and lymphnodes. Isolation and identification of brucella was carried out according to the technique recommended by Alton et al. (1975) as follows: Preparation of samples: Spleen, liver, kidney, lung , heart and lymph node of serologically positive brucella slaughtered animal were collected in sterile polyethylene bags separately at Beni-suef abattoir, then rapidly transferred to laboratory of food hygiene department in ice box with minimum of delay. Direct plating from the examined tissue after surface sterilization on two media 1- Oxoid blood agar base with 10% equine serum and 1% glucose added aseptically after autoclaving (Farrell's medium). Oxoid brucella selective supplement (BBS) is added to this base (Farrell, 1974).2-Trypticase soy agar with 5% sheep blood. The plates were incubated at 37C for 4 days in an aerobic atmosphere containing 10% CO2. The isolated organisms was then identified Histopathological sections of the examined tissues was done in department of pathology Immuuno fluorescence methods.

Results: The incidence of brucella melitensis was higher in cattle than other species of animals. The percentages of brucella melitensis was greater in lymph nodes,spleen and liver. The lowest percentages was in kidney,lung and heart respectively. Brucella aborts could be isolated with different percentages from edible offal in cattle,buffaloe,sheep and goats. The pathological examination

revealed presence of granulomatous reactions in lymphoreticular tissues and also in liver, spleen, kidneys and lungs.Epithelioid cell and giant cell granulomas were predominating in the lymph nodes and liver and tuberculous-like granulomas were found in kidneys and lungs. The handling of edible offal in abattoir or at home can cause brucellosis. Also insufficient cooking of spleen ,liver ,kidneys,lungs and hearts from sources of brucellosis.

The incidences of brucella melitensis in Liver, spleen, lung, kidney, heart and lymph nodes were 40%,50%,15%,20%,15% and 50% in cattle respectively, these percentages were 30%,40%,15%,15%,20% and 40% in buffaloes respectively, while in sheep it was 35%,40%,10%,15%,10% and 40% respectively, and the incidences in goats were 35%,40%,10%,15%,15% and 45% respectively.The incidences of brucella aborts in cattle were 10,15,0.0,5,5, and 15%in liver, spleen, lung, kidney, heart and lymph nodes respectively. While in buffaloes they were 15,15,5,10,0.0 and 15% respectively. Finally brucella aborts could not be detected in any examned organ.

Conclusion: From the present study it could be concluded that brucella melitensis and brucella aborts could be isolated from spleen, liver, kidneys, lungs, hearts and lymph nodes and may cause brucellosis in man. So for prevention of brucellosis, the edible offal of serologically brucella positive slaughtered animals should be condemned.

EVALUATION OF CRASS CARP (CTENOPHARYNGODON IDELLUS) SURIMI IN BURGER FORMULA IN EGYPT

Gehan M. A. Kassem

Food Hygiene and Control Departmentt, Cairo University,
Faculty of Veterinary Medicine, Cairo, Egypt

Key Words: Crass carp, Surimi, beef burger, frozen storage

Background: Although Surimi and its products are, vast growing industry in many countries, surimi and other fish products are nearly not known among Egyptian consumers. However, beef burger is one of most popular fast food especially for kids and youth, in spite of its high fat content and many hazards it involved. Grass carp is produced by a relatively high quantity in Egypt because of its wide production from aquaculture source and natural resources as it used in nationwide programme for biological weed control in the irrigation and drainage systems. However, the fish is low in price and has limited scope for consumption in the fresh form. With over increase concern of consumption of fish as healthy food and in order to maximize the utilization of grass carp surimi was produced and processed into fish burger in pure form and mixed with beef in different concentration (5-20%).

Objective: This study is aiming to explore the effects of beef

substitution by surimi in beef burger formulation and evaluate their shelf life during frozen storage.

Methods: Frozen surimi was tempered at room temperature for 2 hours. Surimi and meat were minced separately through meat grinder at 8mm. Meat, surimi, fat, seasoning and water were added in a bowl mixer and chopped at low speed for 5min. After mixing the batter was formed into discs of 100 gm by using manual patty former. six groups of burger were prepared 1) 100% surimi, 2) 100% beef burger, 3) 5% surimi & 95% beef burger formulation (BBF), 4)10% surimi & 90% BBF, 5) 15% surimi & 85% BBF, 6) 20% surimi & 80% BBF. Chemical composition (Protein, fat, ash and moisture), fatty acid profile were determined for processed burger. Physico- chemical characteristics (pH, cooking loss, color, Shear force, TVB-N, TBA-value), bacterial load (total aerobic plate count, coliforms count, S.aureus count), and sensory qualities also were performed at monthly interval during frozen storage at -18 °C for 3 months. The sensorial quality was determined by means of a panel test, which assessed odor, color, texture, and general appearance during the storage period.

Results: In term of sensory evaluation all groups were highly acceptable moreover, control group (beef burger 100%) proved higher scores than other groups. Protein, moisture content was significantly increased, while fat content was decreased with

increased surimi content in burger. Omega-3 fatty acids were detected in burger groups contains surimi only. Shear force was decreased with increase surimi content in the burger, while cooking loss was increased with increase the surimi content due to high moisture content of surimi. Aerobic plate count was reduced by more than 2 logs at the end of storage in all examined groups. Coliforms and S.aureus counts were detected only in burger containing beef. Coliforms countwere significantly reduced by frozen storage but S.aureus couldn't be detected at the end of storage time. Fat stability represented by TBA-value was significantly stable in burger containing surimi than beef only, but no significantly difference is noticed in TVB-N among groups during storage time.

Conclusion: This study outlines the scope for the development of new ready-to-cook surimi-beef mix burger with high consumers' acceptance and good quality for three months of frozen storage. In the other hand, surimi burger was accepted and show high quality during frozen storage but with some confusion of raw material used in its processing.

THE EFFECT OF THE DIETARY PROBIOTIC *BIFIDOBACTERIUM LACTIS* ON THE ONSET OF METABOLIC SYNDROME IN JUVENILE OSSABAW SWINE FED A HIGH FAT-HIGH FRUCTOSE-HIGH CHOLESTEROL (HF/HFR/HC) DIET

Gloria I Solano-Aguilar[1], Marta Restrepo[1], Sean Smith[2], Cecilia Coscaron[1], Alva Mitchell[3], Timothy Ramsay[3], Kiran Panickar[1], Josh Parker[4], Brittany Garr[1], William Hare[1], Harry Dawson[1], Patrick Gillevet[2], Joseph Urban[1]

[1] Diet Genomics and Immunology Laboratory, Beltsville Human Nutrition Center, Agricultural Research Service, United States Department of Agriculture, Beltsville, MD, USA
[2] Microbiome Analysis Center, George Mason University, Prince William Campus, Manassas, VA, USA
[3] Animal Biosciences and Biotechnology Laboratory, Animal and Natural Resources Institute, ARS-USDA, Beltsville, MD, USA
[4] Masonic Cancer Center Comparative Pathology Shared Resource, University of Minnesota, St Paul, MN, USA

Keywords: Metabolic syndrome, adipocyte, Ossabaw pigs, leptin, fat deposition

The prevalence of high body mass index (BMI) among children in the US at or above the 95th percentile ranges from approximately 10% for infants and toddlers, to approximately 18% for teenagers according to the latest CDC study. In obese children, most of their excess weight is gained before puberty. Therefore, intervention early in life may constitute an important strategy for the control of childhood obesity and related chronic disease later in life. Ossabaw pigs fed a high fat (HF), high fructose (HFr), high cholesterol (HC) diet develop all the components of the metabolic syndrome (MetS) including primary

insulin resistance, obesity with significant visceral adipose expansion, hypertriglyceridemia, and increased LDL: HDL cholesterol ratio. Thus, the Ossabaw pig model provides a unique animal resource to gain insight into complex factors involved in the development of obesity at an early age. To establish the kinetics and level of expression of MetS and to evaluate the effect of probiotics on a dietary induced obesity juvenile model, recently weaned Ossabaw pigs were maintained on a HF/HFr/HC diet supplemented daily with 2 x 10^{10} cfu of *Bifidobacterium lactis* (Bb12) (Group I, n=8), a HF/HFr/HC diet with probiotic-free vehicle placebo (Group II, n=8), or a control basal diet (Group III, n = 8). Extensive data analysis generated from this study, showed that the HF/HFr/HC diet induced increases in weight gain, abdominal circumference, glucose tolerance/insulin insensitivity, total body fat deposition and circulating serum levels of LDL:HDL cholesterol compared to pigs on the basal diet. Pyrosequencing-microbiota analysis revealed less bacterial species diversity in pigs with MetS. Notably, analysis of adipocyte morphometry from abdominal fat deposits of the HF/HFr/HC group given the probiotic-free placebo demonstrated a significant increase in adipocyte size with increased leptin gene expression. These effects were not seen in adipocytes isolated from pigs of the HF/HFr/HC group given Bb12. The nature of these probiotic induced metabolic and molecular changes needs further investigation. The juvenile Ossabaw pig fed a HF/HFr/HC diet provides a unique model of childhood obesity to evaluate the effects of feeding a probiotic or an anti-obesity bioactive dietary component.

A SCIENTIFIC VALIDATION OF ANTIHYPERGLYCEMIC AND ANTIHYPERLIPEMIC ATTRIBUTES OF *TRICHOSANTHES DIOICA*

Prashant Kumar Rai, Shikha Mehta, Surekha Shukla, Sanjukta Chaterjee, Deepak Yadav, Vijay Bahadur and Geeta Watal

Alternative Therapeutics Unit, Drug Discovery and Development Division, Medicinal Research Lab, Department of Chemistry, University of Allahabad, India

Keywords: Trichosanthes dioica, Antidiabetic, Hyperlipemia, and Wister rats.

Background: Trichosanthes dioica had been considered as an effective and safe ethnotherapeutic in Indian traditional system of medicine.

Methods: Effects of variable doses of lyophilized powder of its aqueous extract were studied on blood glucose level (BGL), hemoglobin (Hb), total protein (TP), and lipid profile of normal, mild and severely diabetic animals. Body weight (bw) and urine sugar (US) were additional parameters studied.

Results: Dose of 1000 mg/kg decreases BGL of normal and mild diabetic rats significantly ($P < 0.01$) during fasting blood glucose

(FBG) and glucose tolerance test (GTT) studies respectively. Four weeks long term study of severely diabetic rats with the same dose showed significant fall ($P < 0.001$) in FBG, postprandial glucose (PPG), total cholesterol (TC) and triglyceride (TG) levels. A significant increase ($P<0.01$) in high density lipoprotein (HDL) is of value addition. TP, Hb and bw also showed significant increase ($P<0.05$). Moreover, a significant reduction in US was observed from +4 to +1.

Conclusion: The scientific validation of ethnotherapeutics efficacy of T. dioica as an antidiabetic agent could be used for developing an oral drug managing diabetes and hyperlipidemia associated with it.

INVOLVEMENT OF HEME OXYGENASE-1 IN ATTENUATION OF THE CARDIOPROTECTIVE EFFECT OF ISCHEMIC PRECONDITIONING IN DIABETIC RAT HEART

Harlokesh N. Yadav, Ishan Gupta, and PL Sharma

Department of Pharmacology, I.S.F College of Pharmacy,
Moga, Punjab, India

Key Words: Ischemic preconditioning; Diabetic heart, Caveolin, Nitric oxide; Heme-oxygenase-1, Hemin, Znpp

Objective: It has been noted in our laboratory that upregulation of caveolin attenuate the cardioprotective effect of ischemic preconditioning during diabetes mellitus by inhibiting the release of nitric oxide (NO). Heme-Oxygenase-1 (HO-1) is an enzyme, which facilitates release of NO by disrupting association of caveolin with eNOS. However the expression and activity of HO-1 is reduced in diabetic rat heart. This study was designed to investigate the role of HO-1 in attenuation of cardioprotection induced by IPC in diabetic rat hearts.

Methods: Experimental diabetes was induced by single dose of streptozotocin (50mg/Kg, i.p,). Isolated hearts from normal and diabetic rats were mounted on Langendorff's apparatus and 30 min of ischemia, 120 min of reperfusion with or without IPC (four cycles of 5 min ischemia and 5 min of reperfusion) had been given. Myocardial infarct size was estimated by TTC staining and release

of LDH, CK-MB and nitrite in coronary effluent was also measured. Sodium nitrite was perfused for 30 min after stablization, and was used as positive control. Daidzein (DDZ) (0.2mg/Kg/s.c) was administered to diabetic rat for one week, 3 weeks after the administration of streptozotocin. Hemin (4mg/kg/i.p.) was injected 18 hr before isolation of heart. Zinc Protoporphyrin (50μg/kg/i.p.) was injected 6 hr before hemin treatment.

Results: The cardioprotective effect of IPC was significantly attenuated in diabetic rat heart. Perfusion of sodium nitrite, or pretreatment with DDZ or hemin or combination of DDZ and hemin significantly restored the cardioprotective effect of ischemic preconditioning in diabetic rat heart. However administration of Znpp in hemin pretreated diabetic rat significantly attenuated the restoration of cardioprotective effect of ischemic preconditioning.

Conclusion: It may be concluded that attenuation of cardioprotective effect of ischemic preconditioning is due to impairment of HO-1 induced release of nitric oxide in diabetic rat heart. Activation of heme-oxygenase-1 enzyme by administration of hemin restored the cardioprotective effect of ischemic preconditioning in diabetic rat heart which may be due to the disruption of caveolin-eNOS complex and enhanced release of NO.

PREPARATION OF SHEEP BONE COLLAGEN PAPAIN AND TRYPSIN HYDROLYSATE AND ITS IMMUNOMODULATORY FUNCTION

[1]Huo Nairui, [1]Ma Lizhen, [2]Kong Baohua, [2]Liu Yuhua

[1]Tianjin Agricultural College, PRC, China
[2]Northeastern Agricultural University, Harbin, PRC, China

Key Words: sheep bone; collagen peptides; immunocompetence

Background: Meat processing leaves millions of tons of bone each year worldwide, bone contains of various minerals such as calcium, phosphor, iron, zinc and others, as well as nitrogenous collagen. Collagen occupies 90% of the bone protein and can be hydrolyzed into amino acids and polypeptides which is bioactive and helps to exert every function of collagen to the full length. Our research is designed to hydrolyze sheep bone into immunocompetence peptide with a hope to make full use of the byproduct in meat processing and greatly increase its functional potency. Metabolic syndrome weaken the body's immunity. If the sheep bone collagen peptide prepared in this research shows the ability to enhance the immunity, it could be developed as an assistant and safe supplement in the treatment of the disease.

Objective: Optimize the hydrolyzing condition for papain and trypsin to prepare the sheep bone collagen peptides that can

enhance cell mediated immunity evaluated in vitro, and investigate the immunomodulatory function of the cryodesiccated hydrolysates powder to enhance the humoral immunity and innate immunity in vivo.

Methods: Sheep bone powder was sequentially hydrolyzed by papain and trypsin. After inactivation of trpsin, the hydrolysate was vacuum freeze dried into powder. Enzymatic hydrolysis conditions were optimized using a four elements twice general rotation design method and was analyzed by Design Expert 7.1.2 software. MTT assay was used to determine the effect of the hydrolysates on the cell-mediated immunity(CMI) in vitro in which the effect on the ConA induced proliferation of spleen T cells was investigated. The content of soluble peptide was measured by Folin-hydrozybebzebe method, and the relationship between soluble peptide content with immunocompetence was analyzed by SAS software. ICR mice were divided by body mass into control、low-dose、medium-dose and high-dose groups. Carbon expurgation test and quantitative SRBC hemolysis test were used to determine the impact of the hydrolysates on mice innate and humoral immunity respectively.

Results: The optimum condition for papain catalyzed hydrolysis is enzyme/substrate level of 1576U/g, 64.05 ℃ for 7.22h, and substrate concentration of 0.271Kg/L. After inactivation of papain, adding 0.3% trpsin, adjust pH to 8.0, incubation at 38 ℃ for 3 h.. It

demonstrated that the resulted hydrolysate powder at low dose（0.5 g/kg•d, ig, 14 d）significantly enhanced the carbon clearance ability of macrophagocytes when compared with the control group ($P<0.05$); medium dose (1 g/kg•d, ig, 14 d)and highdose（3g/kg•d, ig, 14 d）would not, but these two dosages remarkably increased the amount of antibody-producing cells in the spleen while low dosage would not.

Conclusion: CMI-enhancing sheep bone collagen papain and trypsin hydrolysates has the potential to enhance both the specific and the innate immunity of normal mice, but this kind of immunomodulatory activity shows no linear relationship with the dosage administrated. Low dosages enhance the innate immunity while medium or high dose has a tendency to improve the humoral immunity.

PYCNOGENOL® AMELIORATES HYPERLIPIDEMIA AND ANTIOXIDANT STATUS IN PANCREAS OF TYPE 1 DIABETIC RATS

Kehkashan Parveen[1], Tauheed Ishrat[2], Waseem A. Siddiqui[1]

[1]Department of Biochemistry, Lipid Metabolism Laboratory, Jamia Hamdard, Hamdard University, New Delhi, India
[2]Department of Emergency Medicine, Emory University, Atlanta, Georgia, USA

Key Words: Pycnogenol; Type 1 diabetes; Oxidative stress; Streptozotocin

Background: Type 1 diabetes mellitus (T1DM), or insulin dependent diabetes mellitus (IDDM), is a chronic metabolic disorder and a major worldwide health problem. T1DM affects 10-20% of diabetic people throughout the world. It is characterized by an absolute or relative deficiency in insulin secretion associated with chronic hyperglycemia often accompanied polyurea, polyphagia, polydipsia and weight loss. Hyperglycemia leads to long-term complications of diabetes, which are the major causes of morbidity and mortality in human populations. Several experimental and clinical studies have shown that the generation of reactive oxygen species (ROS) is increased in TIDM and that the onset of diabetes is closely associated with oxidative stress. Multiple factors contribute to increased oxidative stress in diabetes. Persistent hyperglycemia itself increases the production of ROS [e.g., superoxide radical (O2•), hydroxide radical ($OH^{\bullet}$), hydrogen peroxide (H_2O_2)] through glucose auto-oxidation and

non-enzymatic protein glycation. Furthermore, alteration of the GSH redox state by activation of NADPH-dependent aldose reductase (polyol pathway) and non-enzymatic glycosylation of protein have also been shown to be a source of ROS. Recently the use of antioxidants for diabetes has been advocated, as oxidative stress is known to play an important role in the onset as well as development of further secondary complications of diabetes. Pycnogenol (PYC), an extract of French maritime pine (Pinus maritima) bark, has a high antioxidant capacity. Major constituents of PYC are bioflavonoids, such as monomeric and oligomeric units of catechin, epicathechin, and taxifolin. PYC has been shown to attenuate a wide range of disorders, including cancer, atherosclerosis, diabetes, inflammation, asthma, hypertension, atherosclerosis, immune disease, and others The present study evaluated the role of PYC on hyperglycemia, hyperlipidemia, and oxidative damage in the pancreas of STZ-induced type 1 diabetic rats. Our results support the efficacy of PYC in reducing glucose and improving diabetic complications in a clinically relevant type 1 diabetic model.

Methods: Thirty two rats were divided separated into four groups of eight animals each: group I (control group) control rats were fed standard diet throughout the experiment; group II (control + PYC) rats were fed standard diet throughout the experiment and given PYC (10 mg/kg body wt., intraperitoneally (IP) ; in saline) for 4 weeks; group III (STZ group) rats given a single IP injection of STZ at a dose of 60 mg/kg body weight (fresh solution in 0.1 N citrate buffer, pH 7.5). The rats had free access to 5% of glucose water and basal diet ad

libitum during the next 24 hours. Group IV (STZ + PYC) rats were injected with STZ and then supplemented with PYC (10 mg/kg body wt.; IP; in saline) for 4 weeks. The development of hyperglycemia in rats was confirmed by fasting blood glucose (FBG) estimation after 4 days of STZ injection. The animals that maintained FBG higher than 140 mg/dl were considered diabetic and selected for studies. The PYC treatment was started after diabetes was confirmed. At the end of the experiment, blood was drawn and rats were then sacrificed and their liver (hepatic glycogen) and pancreas was dissected for biochemical and histopathological assays.

Results: The level of fasting blood glucose, glycated hemoglobin, total cholesterol, triglycerides, low density lipoprotein-cholesterol and very low density lipoprotein-cholesterol significantly increased while high density lipoprotein cholesterol and hepatic glycogen decreased in the STZ group. PYC treatment significantly augmented ($P < 0.05$) these effects in the STZ + PYC group. Moreover, treatment with PYC significantly ($P < 0.05$) ameliorated thiobarbituric reactive substances, malonaldehyde and protein carbonyl, and glutathione, glutathione-s-transferase and catalase in the pancreas of the STZ group.

Conclusion: The study suggests that PYC is effective in reducing hyperglycemia, hyperlipidemia and oxidative stress related to the risk of diabetes. Thus, it may have a therapeutic value for the treatment of Type 1 diabetes.

METABOLIC SYNDROME AND ITS ONSET DUE TO DEFICIENCY OF MICRONUTRIENTS DURING THE PRECONCEPTION, CONCEPTION AND POST NATAL DEVELOPMENTAL PERIOD

M.S. Mohamed Jaabir[1], J.F. Rosario[2], S. Senthil Kumar[3], P. Geraldine[1]

[1]Dept. of Animal Science, Bharathidasan University, Tiruchirappalli, Tamil Nadu, India
[2]Dept. of Obstetrics & Gynecology, University of Texas, Health Science Center, San Antonio, Texas, USA
[3]Dept. of Biotechnology, Jamal Mohamed College *(Autonomous)*, Tiruchirappalli, Tamil Nadu, India

Key Words: metabolic syndrome, micronutrient restriction, insulin synthesis, TNF - □, and leptin

Background: Micronutrients such as vitamins and minerals play essential roles in cellular metabolism, maintenance and growth throughout life. Several studies indicate that vitamins and minerals affect insulin sensitivity. Moreover, deficiencies of these vitamins can have profound and often persistent effects on many fetal tissues and organs, even in the absence of any clinical signs of deficiency in the mother. Furthermore, the consequences of vitamin imbalance during fetal development may not be apparent at the time of the nutritional insult, but may manifest during later development. Multiple vitamin deficiencies, particularly during pregnancy and/or Post natal Development, are common in the

developing world and maternal vitamin deficiencies are associated with low birth weights and increased rates of perinatal mortality and morbidity.

Objective: In the present study, it was hypothesized that maternal dietary micronutrient restriction would predispose the offspring to increased body adiposity and insulin resistance in later life. This would facilitate the development of strategies to improve the nutritional status of pregnant mothers, which shall lead to a decrease in the birth of babies with low birth weight and in turn the consequent metabolic syndrome (Insulin Resistance, Hypertension and Obesity) in their later life.

Methods: The study was conducted in mice to validate our hypothesis as an effect of restriction of micronutrients as a mixture. Female weanling mice received the control diet (AIN93G) and 50% micronutrient restricted (MR) diet and were mated with control males. Pups born to the dams on the restricted diet were weaned on to the restricted diet till postnatal day (PD) 360. The effect of maternal dietary micronutrient restriction during preconception, conception and postnatal days was studied based on the levels of insulin resistance, fat metabolism, adipocytokines, oxidative stress, mitochondrial damage of the skeletal muscle and systolic blood pressure in the offspring periodically till post natal days 360.

Results: At birth, pups from deficient dams had reduced birth weight and crown rump length. Increased fasting glucose, insulin, total cholesterol and triglycerides levels were observed in the offspring of MR group. At PD-120, MR restricted offspring had an elevated systolic blood pressure than controls. Compared with controls, total body electrical conductivity (TOBEC) measurements indicated significantly higher body fat percentage, lower lean body mass and fat-free mass in MR offspring besides elevated plasma triacylglycerols. Maternal micronutrition restriction per se resulted in an increased body fat and in plasma triglycerides, free fatty acids and total cholesterol concentrations in the offspring. These changes seem to predispose the offspring to insulin resistance and hypertension in later life. Mitochondrial DNA damage and apoptosis in the skeletal muscle cells were also observed in the restricted group, strongly suggesting the molecular mechanisms underpinning long-term programming and onset of adult life disorders.

Conclusion: The results indicate that maternal micronutrient restriction leads to early changes in the adiposity of the offspring. They stress the importance of maternal micronutrient status in regulating insulin synthesis / secretion in the offspring, their insulin response to a challenge of glucose and progressive development of hyperglycemia / insulin resistance in them. The increased body adiposity appears to be due to alterations in the expression of adipocytokines like TNF - □ and leptin, which in

turn seem to regulate the synthesis and transport of fatty acids. Maternal micronutrient restriction also appears to irreversibly decrease muscle mass as well as its basal glucose uptake. It thus appears from our results that the recent increase in the incidence of insulin resistance and associated diseases among the Indians and other developing countries could at least partly be due to the widely prevalent micronutrient restriction among the pregnant and lactating women.

REGULATION OF INSULIN SENSITIVITY BY HERBS AND DIETARY FIBERS

Jianping Ye

Antioxidant and Gene Regulation Lab, Pennington Biomedical Research Center, Louisiana State University, Baton Rouge, LA, USA

Key Words: Insulin Sensitivity, Dietary Fibers, Herbs, Metabolic Syndrome

Background: Herbs and dietary fibers are botanical products. They are used widely by public in health promotion and chronic disease care (1). The efficacy and action mechanism are two major concerns on the botanical products (2-3). Here, I will use two examples to address the concerns with regard to metabolic syndrome. Berberine is a botanical alkaloid of herb Huanglian and used to control blood glucose in type 2 diabetic patients in China for many years (4).

Results: We investigated its action mechanism with a focus on AMPK activation (5). The Berberine induced AMPK activation at 0.5 h in cell culture and the activity remained for 16 h. The AMPK activation was associated with persistent elevation in AMP/ATP ratio, reduction in oxygen consumption, and an increase in glycolysis. These results suggest that berberine inhibits

mitochondrial function to improve glucose metabolism through glycolysis and AMPK activation. Dietary fiber leads to production of sodium butyrate (SB) after fermentation in the large intestine. We investigated mechanism of SB action in mice through oral supplementation (6). SB prevented body weight gain on a high fat diet and attenuated insulin resistance. The mechanism is induction of energy expenditure by SB. The energy expenditure was associated with fatty acid oxidation without an increase in spontaneous physical activity. Mitochondrial function was enhanced by SB with elevated gene expression (PGC-1a, UCP-1, CPT-1b and COX-I). The data suggest that butyrate improves insulin sensitivity through induction of mitochondrial function. These data suggest that mitochondria are targets of botanical products in the regulation of metabolism.

Conclusion: Butyrate improves insulin sensitivity through induction of mitochondrial function and mitochondria are targets of botanical products in the regulation of metabolism.

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A COMBINATION OF NUTRIENTS IMPROVES MITOCHONDRIAL BIOGENESIS AND FUNCTIONIMMUNE FUNCTION IN TYPE 2 DIABETIC GOTO–KAKIZAKI RATS

Jiankang Liu, Weili Shen, Jiejie Hao

Institute of Mitochondrial Biology and Medicine, Xi'an Jiaotong University School of Life Science and Technology, Xi'an, China
Department of Hypertension, Ruijin Hospital, Shanghai Jiaotong University School of Medicine, Shanghai, China,
School of Medicine and Pharmacy, Ocean University of China, Qingdao, China

Key Words: Dietary supplement, mitochondrial biogenesis and function, immune function, skeletal muscle

Background: Evidence indicates that type 2 diabetes and insulin resistance may be related to reduce mitochondrial number and oxidation capacity as well as immune dysfunction. However, it is not known whether increasing mitochondrial number and function and immune function improves type 2 diabetes and insulin resistance.

Objective: In the present study, we investigated whether a combination of nutrients could have protective and therapeutic effects on insulin resistance and mitochondrial biogenesis/function in skeletal muscle and immune function of type 2 diabetic Goto–Kakizaki rats.

Methods: Goto–Kakizaki rats were treated with a combination of R-α-lipoic acid, acetyl-L-carnitine, nicotinamide, and biotin for 8 weeks and mitochondrial biogenesis and immune function were examined.

Results: We demonstrated that defect of glucose and lipid metabolism is associated with low mitochondrial content and reduced mitochondrial enzyme activity in skeletal muscle of the diabetic Goto-Kakizaki rats. The treatment of combination of R-α-lipoic acid, acetyl-L-carnitine, nicotinamide, and biotin effectively improved glucose tolerance, decreased the basal insulin secretion and the level of circulating free fatty acid, and prevented the reduction of mitochondrial biogenesis in skeletal muscle. The nutrients treatment also significantly increased mRNA levels of genes involved in lipid metabolism, including Pparα, Pparδ, and Mcpt-1 and activity of mitochondrial complex I and II in skeletal muscle. We also found that immune dysfunction in these animals is associated with increased oxidative damage and mitochondrial dysfunction and that the nutrient treatment effectively elevated immune function, decreased oxidative damage, enhanced mitochondrial function, and inhibited the elevation of apoptosis factors. All of these effects of mitochondrial nutrients are comparable to that of the antidiabetic drug, pioglitazone. In addition, the treatment with nutrients, unlike pioglitazone, did not cause body weight gain.

Conclusion: These data suggest that a combination of mitochondrial targeting nutrients may improve immune function and mitochondrial function by stimulating mitochondrial biogenesis and exert hypoglycemic effects, without causing weight gain.

ANTI-OBESITY OF GINGER IN RAT FED WITH HIGH FAT DIET

Jintanaporn Wattananthorn, Wipawee Boongvang, Supaporn Muchimapura, Terdthai Tongun, Panakporn Wannanond

Department of Physiology, Faculty of Medicine, Khon Kaen University, Khon Kaen, Thailand

Key Words: Ginger consumption, body weight gain, and phytotherapy

Background: Obesity occurs as a result of an energy imbalance caused by increased ratio of energy intake to energy expenditure. It is an important risk factor to metabolic syndrome and its prevalence is still continually increasing its importance. Therefore, treatment and prevention against this condition is important for achieving the healthy life. Phytotherapy has been long term used in traditional folklore including in Traditional Chinese Medicine (TCM). According to TCM, weight gain or obesity is caused by an unhealthy interaction between yin and yang. Therefore, phytotherapy in TCM is focused on yang herb in order to increase metabolism and sweating. Ginger is one of the spices commonly used. Moreover, it has been recognized as yang herb and previously claimed to possess hypolipidemia effect and benefit for obesity. Although the anti-obesity of ginger has been previously claimed, the possible underlying mechanism is not fully

understood and less supported scientific evidence about anti-obesity is available.

Objective: To investigate the effect of ginger on obesity and determine the possible underlying its effect in high fat diet induced obese rats.

Method: Male Wistar rats, weight about 200-220 g, were induced obesity using high fat diet containing calory 4370 Kcal/kg after acclimatization for 1 week. Then, they were divided into various groups as following, high fat diet with vehicle, high fat diet with orilstat (positive control) and high fat diet with ginger at doses of 2.5 and 5 mg/kg BW once daily at a period of 48 days. Then, they were determined food and water intake, weight gain, lipase activity, amount of fat pad, and metabolism using body temperature as indicator.

Results: Our data showed that ginger could significantly decrease weight gain while no significant changes of food and water intakes were observed. In addition, ginger also decreased fat pad especially at mesenteric and retroperitoneal regions but increased body temperature. Therefore, the current data pointed out that the metabolic rate was enhanced by ginger.

Conclusion: Ginger consumption significantly suppressed body weight gain and decreased body fat pads especially in mesenteric

and retroperitoneal areas. Its effect may occur via the enhanced lipase activity and metabolism. Ginger, a very popular yang herb, is the potential candidate to combat against obesity induced by high fat diet.

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MODELING TYPE II DIABETES IN *DROSOPHILA MELANOGASTER*

Johannes Bauer, Siti Nur Sarah Morris, and Bilal Fawaz

Department of Biological Sciences, Southern Methodist University, Dallas, TX, USA

Key words: *Drosophila melanogaster*, diabetes, and insulin

Background: Diabetes affects ~8% of the US population, at an estimated cost of $116 billion. Great strides have been made in research into the causes and treatments of diabetes, but the lack of a cost effective animal model system has hampered research efforts. Recently, the fruit fly *Drosophila melanogaster* has emerged as a promising alternative to expensive mammalian models. *Drosophila* contains orthologs of mammalian pancreatic alpha and beta cells. These cells are the corpora cardiaca and the insulin-producing cells, respectively. Interestingly, insulin-producing cells are situated in the fly brain, and extend their axons into the fly thorax to the corpora cardiaca, a gland that is near the fly heart. Insulin-producing cells are stimulated by the fly ortholog of mammalian neuropeptide Y, the *Drosophila* short neuropeptide F, in response to food conditions and secrete up to four different insulin-like peptides. Cells in the corpora cardiaca in turn secrete adipokinetic hormone, a functional equivalent to glucagon. In addition, intracellular signaling pathways, such as insulin receptor signaling or the related TOR-signaling pathway, are conserved in flies as well. With the basic cellular and molecular

architecture regulating growth, metabolism and the response to nutrients in place, *Drosophila melanogaster* is a promising model system to study metabolic diseases, such as Type II Diabetes. We thus propose to use *Drosophila* as a cost-effective and rapid alternative model system for diabetes research. Measurements of *Drosophila* health parameters in response to various feeding regimes will define the conditions under which flies develop diabetic phenotypes. With benchmarks established, the *Drosophila* system can provide a quick and inexpensive screening tool for pharmacologic and genetic interventions that modify the diabetic phenotype.

Methods: In order to define *Drosophila* diabetes, flies will be raised on different diets with varying nutrient content, ranging from diets that lead to undernutrition to diets that cause overfeeding. Each diet component will be varied independently to investigate the effects of the different micronutrients on fly health. At defined ages, flies will be analyzed for metabolic benchmarks (stored glucose, glycogen and fat levels, circulating glucose and trehalose levels, as well as levels of insulin and glucagon). Furthermore, the main metabolic organ of the fly, the fat body, will be assayed for insulin-signaling activity. In order to rescue the diabetic phenotype, flies with genetic mutations that are known to alter metabolism, or wild-type flies treated with known anti-diabetic compounds, will be exposed to conditions that lead to hallmarks of hyperglycemia or diabetes (eg elevated levels of circulating sugars, decreased insulin-signaling in the fat body) to assess the efficacy of those interventions. These experiments will lay

the basis for performing pharmacologic and genetic screens for novel compounds and genetic factors that modify diabetes.

Results: Our results shows that fly fat bodies lose responsiveness to insulin in early adulthood. This process is accelerated on high caloric diets and delayed by interventions that extend longevity. Furthermore, flies raised on higher caloric diets have lower levels of the non-reducing sugar trehalose. Interestingly, trehalose levels have recently been linked to increased longevity in another important model system, the nematode *C. elegans*. Treatment with the anti-diabetic compound metformin leads to heavier flies and increased levels of stored glucose (glycogen and trehalose) on all dietary regimes, suggesting that Metformin may act by diminishing circulating glucose. In addition, metformin prevented the age-dependent decline in trehalose levels.

Conclusions: These data show that diabetes-like phenotypes can be observed in fruit flies. Hallmarks of this phenotype are low trehalose levels and the loss of insulin-sensitivity in the fly fat body. Furthermore, our data demonstrate that pharmacologic interventions inhibit these phenotypes and thus suggest that the fly will be a useful screening tool for the discovery of novel anti-diabetic compounds.

EXERCISE AT THE MAXIMAL LACTATE STEADY STATE ALTERS MARKERS OF NON ALCOHOLIC FATTY LIVER DISEASE AND METABOLIC SYNDROME IN RATS FED ON FRUCTOSE RICH DIET

José Diego Botezelli[1], Rodrigo Ferreira de Moura, Leandro Ferreira de Moura, Rodrigo Augusto Dalia, Michel Barbosa Araújo, Carla Ribeiro, Ana Carolina Ghezzi, Lucieli Teresa Cambri, Fabrício Azevedo Voltarelli, Maria Alice Rostom de Mello

[1] São Paulo State University – UNESP, Department of Physical Education, Rio Claro, São Paulo, Brazil.

Key Words: Metabolic syndrome, insulin sensitivity, and fructose rich diet

Background: Metabolic syndrome, also known as syndrome X or insulin resistance syndrome encompasses a spectrum of disorders, being the impaired glucose tolerance one of the most important. These disorders include insulin resistance (with or without type 2 diabetes *mellitus*), hypertension, obesity, dyslipidemia and endothelial dysfunction. The typical signs of metabolic syndrome have been induced in rats by prolonged administration of diet containing high amounts of fructose. Non-alcoholic fatty liver disease (NAFLD) is emerging as an acknowledged component of metabolic syndrome.

Objective: This study aimed to analyze the effects of exercise at the aerobic/anaerobic transition on markers of non alcoholic fatty

liver disease (NAFLD), insulin sensitivity and blood chemistry of rats fed on a fructose rich diet.

Methods: Male **w**istar rats, were separated into two groups according to diet: control (balanced diet AIN-93 G) and fructose (diet with 60% fructose). These animals were tested for maximum lactate steady state (MLSS) in order to identify the aerobic / anaerobic metabolic transition during swimming exercise at 28 and 90 days of age. One third of the animals of each group was subjected to swimming training at intensity equivalent to the individual MLSS, 1 h / day, 5 days / week from 28 to 120 days (early protocol); another third was subjected to the training from 90 to 120 days (late protocol), composing thus, six groups: C= Control, CET= Control Early Trained, CLT= Control Late Trained, F= Fructose, FET= Fructose Early Trained and FLT= Fructose Late Trained. At the end of the experiment, the animals were killed by sodium thiopental administration in order to determine: serum glucose and triglycerides (TG) levels; serum glucose disappearance rate using the insulin tolerance test (kitt); and total liver lipid concentration. The data were analyzed by ANOVA *two-way*; the Bonferroni post hoc test was applied when necessary.

Results: The results are described in the table 1.

Conclusion: The late-training protocol employed was effective in restoring insulin sensitivity of the acceptable level in the animals. Both training protocols were successful in preventing the NAFLD

appearance. Further studies are required in order to elucidate which exercise protocol is better to prevent and/or treat the metabolic syndrome.

Table1: Parameters determined at the end of experiment. Results are Mean± SD of 8 rats in each group

	C	CET	CLT	F	FET	FLT
Serum Glucose (mg/dl)	112.9± 27.5	100.6± 36.5	96.3± 13.8	107.0± 2.46	88.1± 18.0	101.3± 16.6
Serum TG (mg/dl)	150.4± 35.1	179.6± 32.6	145.0± 40.3	173.0± 32.9	213.2± 40.2†	192.1± 36.3ffi
Kitt (%/min)	1.4± 0.3	1.1± 0.1	0.7± 0.01*	0.14± 0.06*	0.38± 0.03†§	1.01± 0.02ffi§
Liver Lipids (mg/100mg)	5.9± 1.2	4.6± 0.3	6.0± 1.8	7.7± 0.5*	5.5± 1.6§	5.9± 1.7§

C= Control, CET= Control Early Trained, CLT= Control Late Trained,
F= Fructose, FET= Fructose Early Trained and FLT= Fructose Late Trained
*Significant difference compared to C group (p≤0. 05 Anova two-way).
† Significant difference compared to C group (p≤0. 05 Anova two-way).
ffiSignificant difference compared to C group (p≤0. 05 Anova two-way).
§Significant difference compared to C group (p≤0. 05 Anova two-way).

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FOOD SAFETY MEANING FOR HUMAN HEALTH

Kakha Nadiradze and Nana Phirosmanashvili

BioNanotechnology Inc., Tbilisi, Republic of Georgia

Key Words: Food Safety, GMO's, ISO's, HACCP

Objective: Food Safety, Biosafety, Nutrition

Methods: R-T PCR, ISO', Chromatography

Every man, women and child has the right to adequate food. Adequate food means enough food in quantity and quality, safe and culturally acceptable. A right-based approach is a different way to address food and nutrition insecurity; it emphasizes the rights of human beings to food and it obligates governments to create adequate pre-conditions to feed oneself as well as the priority for politicians to protect directly the poor and hungry. Different levels of understanding of biological mechanisms and an insufficient diffusion of knowledge on the current state of research in safety assessment of genetically modified products might be responsible for the present situation in communication of biotechnology issues. Food acquisition, production, modification, and processing technologies form a continuum of allied biotechnologies, which trace back to the beginnings of agriculture.

These technologies have made use of naturally-occurring genetic variation in an effort to create variety and expand the reach of food sources. This work resulted in the development of tiered decision trees for determining food safety issues for foods derived from microorganisms, single chemicals and simple mixtures, and whole foods and other complex mixtures.

A SCIENTIFIC VALIDATION OF ANTIHYPERGLYCEMIC AND ANTIHYPERLIPEMIC ATTRIBUTES OF *TRICHOSANTHES DIOICA*

Prashant Kumar Rai, Shikha Mehta, Surekha Shukla, Sanjukta Chaterjee, Deepak Yadav, Vijay Bahadur and Geeta Watal[a]*

Alternative Therapeutics Unit, Drug Discovery and Development Division, Medicinal Research Lab, Department of Chemistry, University of Allahabad, Allahabad, India

Key Words: Trichosanthes dioica; Antidiabetic; Hyperlipemia; Wister rats.

Background*: Trichosanthes dioica* had been considered as an effective and safe ethnotherapeutic in Indian traditional system of medicine.

Methods: Effects of variable doses of lyophilized powder of its aqueous extract were studied on blood glucose level (BGL), hemoglobin (Hb), total protein (TP), and lipid profile of normal, mild and severely diabetic animals. Body weight (bw) and urine sugar (US) were additional parameters studied.

Results: Dose of 1000 mg/kg decreases BGL of normal and mild diabetic rats significantly ($P < 0.01$) during fasting blood glucose (FBG) and glucose tolerance test (GTT) studies respectively. Four weeks long term study of severely diabetic rats with the same dose

showed significant fall (P< 0.001) in FBG, postprandial glucose (PPG), total cholesterol (TC) and triglyceride (TG) levels. A significant increase (P<0.01) in high density lipoprotein (HDL) is of value addition. TP, Hb and bw also showed significant increase (P<0.05). Moreover, a significant reduction in US was observed from +4 to +1.

Conclusion: The scientific validation of ethnotherapeutics efficacy of *T. dioica* as an antidiabetic agent could be used for developing an oral drug managing diabetes and hyperlipidemia associated with it.

INDUCTION OF FATTY LIVER IN RATS ON HIGH FAT DIET

Leandro Pereira de Moura[1], José Diego Botezelli, Rodrigo Augusto Dalia, Michel Barbosa de Araújo, Amanda Christine da Silva, Carla Ribeiro, Fabrício Azevedo Voltarelli, and Maria Alice Rostom de Mello

Leandro Pereira de Moura São Paulo State University, Physical Education Departament. Rio Claro, SP, Brazil

Key Words: Fatty Liver, High Fat Diet, and insulin sensitivity

Background: High-fat diets consumption has increased in the last decades, concomitant to a sharp increase in obesity incidence around the world, including in Brazil. Obesity may lead to several deleterious conditions, as the non-alcoholic fatty liver disease (NAFLD). NAFLD occurs due lipid accumulation in the liver and may impair the organ function. Taking into account the limitation imposed in researches with human beings, animal models may be of great benefit for the study of NAFLD pathogenesis.

Objective: In this sense the present study aimed to induce the NAFLD in rats, through a high-fat diet administration, in order to analyze insulin sensitivity (serum glucose disappearance rate - KITT - during insulin tolerance test), serum free fatty acids (FFA) and total lipids and lipogenic rate in the liver.

Methods: For this purpose, twenty male Wistar rats (90 days of age) were divided into two groups, in accordance to the diet regimen:

Control (C): eutrophic rats fed a standard balanced rodent diet (Purina®) and high-fat diet (HFD): rats subjected to a high-fat diet containing 35% of fat.

Results: At the end of 15 days, the HFD group showed: a) lower KITT (% removal of serum glucose / min) (C: 0.176 ± 0.05; HFD: 0.089 ± 0.06), indicating insulin resistance; b) higher serum FFA concentrations (μEq/L) (C: 0.203 ± 0.06; HFD: 0.314 ± 0.03); c) higher liver total lipids concentration (mg/100mg) (C:3.82 ± 0.190; HFD: 7.19 ± 0.90), and d) increased lipogenic rates (umol 3H incorporated /h.g) (C: 15.03 ± 1.95; HFD: 21.13 ± 2.12) than the C group.

Conclusion: The results of the present study showed that a short-time administration of a high-fat diet was able to increase liver fat content in young rats, associated to a high lipogenic activity. Therefore, the procedures here employed proved adequate to induce NAFLD in rats.

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EFFECT OF DEUTERIUM OXIDE (D2O) CONTENT OF DRINKING WATER ON GLUCOSE METABOLISM IN STZ-INDUCED DIABETIC RATS

[1]Miklós Molnár, [1]Katalin Horváth, [1]Tamás Dankó, [2]Gábor Somlyai

[1]Semmelweis University Budapest, Institute of Pathophysiology, Budapest, Hungary; [2]HYD LLC for Cancer Research and Drug Development, Budapest, Hungary

Key Words: deuterium depleted water, GLUT-4 translocation, diabetes

Background: Deuterium, a stable isotope of hydrogen, binds to oxygen to form D2O. D2O exist in the environment at 1/6700 of H2O (150 ppm) and is expected to have some biological effects. Several lines of evidences suggest that D2O inhibits insulin release from pancreatic islets. Very little or no data is available on the action of lowering D2O content of the cellular environment. Some experimental and clinical observations suggest that depletion of D2O has anti-mitotic effect in various tumor cells. Some clinical observations also suggest that depletion D2O interfere with glucose metabolism in diabetic patients.

Objective: In our experiments we wanted to test the effect of removal of D2O on the glucose metabolism in sreptozotocin (STZ)-induced diabetic rat model.

Methods: Diabetes was induced by a single ip. injection of 60mg/kg body weight of STZ. After 2 weeks, animals were randomly

distributed into several groups to test the effect of D2O (25-150 ppm) on glucose metabolism in diabetic animals with or without 2x1 U/day insulin treatment. The following parameters were tested: serum glucose, -fructose amine, -HbAIC, -creatinine, -TBARS and -insulin; urine glucose, -creatinine and -protein. At the end of the experiments, 8 weeks of treatment, membrane associated GLUT-4 content was estimated by western-blot technique from m. soleus.

Results: Our results indicate that STZ treatment significantly increased serum glucose, fructose amine, HbAIC and TBARS concentration. Depletion of D2O did not influence any of the measured parameters in animals not received insulin. However the measured parameters were significantly lower in those animals received lower D2O containing drinking water and insulin treatment. The membrane associated GLUT-4 was significantly higher in these animals also.

Conclusion: These data suggest that D2O depletion enhance insulin effect on GLUT-4 translocation and potentiate glucose uptake in diabetic animals. The major characteristic feature of metabolic syndrome is the decreased insulin sensitivity. Insulin resistance/hyper-insulinemia is strongly associated with hyperlipidemia and hypertension, two major risk factors of coronary heart disease. Based on our experimental data, deuterium depleted water could be used to treat patients with MS by increasing the insulin sensitivity.further experiments to elucidate this question.

EFFECTS OF RUTIN ON LIPID PROFILE IN HYPERCHOLESTEROLAEMIC RATS

Marjan Nassiri-Asl, Amir Ziaee, Farzaneh Zamansoltani, and Esmail Abbasi

Qazvin Metabolic Disease Research Centre, Qazvin University of Medical Sciences, Qazvin, Iran

Key Words: Rutin, Flavonoid, LDL, Hypercholesterolaemic Rats

Background: Rutin (3, 3′, 4′, 5, 7-pentahydroxyflavone-3-rhamnoglucoside) is a flavonoid of the flavonol type. Rutin is found in many plants and is also an important dietary constituent of food and plant-based beverages. Rutin has several pharmacological properties including antioxidant and cardioprotective activities. Also, it was identified that rutin is the major low-density lipoprotein (LDL) antioxidant compound of mulberry in an *in vitro* study.

Methods: The effects of rutin were tested by using it as a supplement in a high-cholesterol diet. Male rats were fed a high-cholesterol diet (1 ml/100 g) for 4 weeks with rutin (10 or 100 mg/kg) or rutin 100 mg/kg and lovastatin supplementation to study the hypocholesterolaemic effects of rutin on plasma lipid levels, hepatic enzyme activity, and liver tissue.

Results: Feeding the animals a high-cholesterol diet resulted in marked hypercholesterolaemia and increased the serum level of LDL cholesterol (LDL-C). Rutin (at 100 mg/kg) alone or in combination with lovastatin significantly reduced the levels of total cholesterol, and LDL-C and also markedly decreased liver enzymes and weight in animals with a high-cholesterol diet. Our findings show that 100 mg/kg of rutin alone or with lovastatin supplementation lowered liver weight and enzymes as well as plasma total cholesterol and LDL. The hepatic histopathological results reflect the correlation of rutin and lovastatin combination with both liver weight and the levels of plasma total cholesterol and LDL-C.

Conclusion: These results indicate that rutin in combination with lovastatin has increased anti-hypercholesterolaemic effects in an animal model. It seems that several mechanisms may contribute to the hypercholesterolaemic effects of rutin.

INHIBITION OF ANGIOTENSIN CONVERTING ENZYME *IN VITRO* BY APPLE SKIN EXTRACT, SELECTED FLAVONOIDS AND THEIR METABOLITES

Nileeka Balasuriya and H.P. Vasantha Rupasinghe

Department of Environmental Sciences, Nova Scotia Agricultural College, Truro, Nova Scotia, Canada

Key Words: Hypertension, renin angiotensin system (RAS), angiotensin converting enzyme (ACE), flavonoids, and apple

Background: Angiotensin converting enzyme (ACE) is a key component in the renin angiotensin system (RAS) which regulates blood pressure. As the over expression of RAS is associated with vascular hypertension, ACE inhibition has become a major target control for hypertension. The research on potential ACE inhibitors is expanding broadly and most are focused on natural product derivatives like peptides, polyphenolics, terpenes etc. Plant polyphenolics are strong bioactive molecules with various beneficial pharmacological properties. The current study is focused on investigating the ACE inhibitory property of fruit flavonoids. An apple skin extract (ASE) rich in flavonoids, the major constituents of the extract and their selected metabolites were assessed for the ACE inhibitory property *in vitro*. It is important to investigate the metabolites along with the flavonoids as they are the constituents active inside the human body.

Objective: To investigate whether flavonoids, flavonoid rich apple extracts and their metabolites could inhibit ACE *in vitro.*

Method: The samples were incubated with sodium borate buffer (30 µL, pH 8.3), 150 µL of substrate (Hip-His-Liu) and ACE (30 µL) at 37 ^{o}C for 1 h. The reaction was stopped by addition of 150 µL of 0.3M NaOH. The enzyme cleaved substrate was detected by making a fluorimetric adduct by adding 100 µL of o-phthaladehyde for 10 min at room temperature. Reaction was stopped by adding 50 µL of 3M HCl. Fluorescence was measured by using a FluoStar Optima plate reader at excitation of 350 nm and emission of 500 nm.

Results: The extract and the compounds showed a concentration dependant enzyme inhibition. Increasing concentrations from 0.001 ppm to 100 ppm of ASE showed an increment of 29% to 64% ACE inhibition. The IC_{50} values of ASE, quercetin, quercetin-3-glucoside, quercetin-3-galactoside, cyanidin-3-galactoside were 49 µg/mL, 151 µM, 71 µM, 180 µM, 206 µM, respectively. The major constituents of the ASE that were tested separately showed effective ACE inhibition. From the three metabolites tested, only quercetin-3-glucuronic acid showed concentration dependant ACE inhibition. The ACE inhibition of 0.001 ppm to 100 ppm of quercetin-3-glucuronic was were 43% and 75%, respectively and the IC_{50} value was 27 µM.

Conclusion: The results demonstrated that flavonoids have a potential to inhibit ACE in vitro and the inhibitory property varies according to type of sugar moiety attached at C-3 position. The results also revealed that the major contributing compounds of ASE for ACE inhibition belong to flavonoids. Among the tested compounds, the lowest IC_{50} value is associated with the quercetin-3-glucuronic acid, a major in vivo metabolites of quercetin and its glycosides. The results suggest that certain dietary flavonoids may possess properties of blood pressure regulation.

BETAINE REDUCES THE EFFECTS OF INFLAMMATION CAUSED BY HYPOXIA IN HUMAN ADIPOCYTES

Olli K, Tiihonen K, Lahtinen S, Rautonen N

Danisco Health and Nutrition, Kantvik, Finland

Key Words: betaine, adipocyte, obesity, inflammation, hypoxia

Background: In obesity hypoxic conditions are developed in the adipose tissue partly as a result of fat mass expansion. Expression and secretion of inflammation-related adipokines is increased in the white adipose tissue suffering from the low oxygen supply. Betaine functions as an intracellular osmolyte and methyl donor decreasing plasma homocysteine, which is associated with increased risk for cardiovascular disease by enhancement of oxidative stress or inflammation.

Objective: We examined the expression of adipokines in human adipocytes in vitro challenged by 1% O2 hypoxia and tested the effects of betaine on the expression of inflammation-related adipokines.

Methods: Human omental preadipocytes were differentiated into mature adipocytes, which were then treated with betaine and cultivated under hypoxic conditions (1% O2). Betaine was used in three concentrations; 50, 250 and 500 µmol/L which corresponded

to the average concentration of betaine in human plasma, and the peak concentrations in serum after a single doses of 1 and 3 g of betaine, respectively. The expression of selected adipocyte-derived markers (e.g. IL-6, leptin) was measured by RT-qPCR.

Results: Our experiments showed that hypoxia increased the expression of leptin, and that betaine reduced this effect caused by the low oxygen conditions. Furthermore, in hypoxic conditions betaine reduced also the expression of the pro-inflammatory marker IL-6.

Conclusion: Hypoxia occurring in the enlarged white adipose tissue contributes to the development of the inflammatory status. Treatment of adipocytes with betaine in in vitro hypoxic conditions diminished the expression of IL-6 and leptin, suggesting that consumption of betaine may reduce the chronic inflammatory state in the adipose tissue in obese subjects. By reducing the systemic inflammation in adipose tissue betaine may also reduce risk factors for other obesity-related diseases.

ANTIDIABETIC EFFECTS OF TRIGONELLA FOENUM GRAECUM AND SODIUM ORTHOVANADATE ON METABOLIC PARAMETERS IN EXPERIMENTAL DIABETES

Pardeep Kumar, Asia Taha, R.K. Kale and Najma Zaheer Baquer

School of Life Sciences, Jawaharlal Nehru University, New Delhi, India

Key Words: Alloxan diabetes, lipid peroxidation, protein kinase C, antioxidant enzymes, sodium orthovanadate (SOV) and Trigonella foenum graecum seed powder

Background: Diabetes mellitus is a heterogeneous metabolic disorder characterized by hyperglycemia resulting in defective insulin secretion, resistance to insulin action, or both. The use of biguanides, sulfonylurea and other drugs are valuable in the treatment of diabetes mellitus, their use, however is restricted to their limited action, pharmaco kinetic properties, secondary failure rates and side effects. Trigonella foenum graecum, commonly known as fenugreek is a plant which has been extensively used as a source of antidiabetic compounds from its seeds, and leaf extracts, preliminary human trials and animal experiments suggest a possible hypoglycemic and anti hyperlipedemic properties of oral fenugreek seed powder.

Objective: In the present study, the effect of sodium orthovanadate (SOV) and Trigonella foenum graecum seed powder administration has been studied on blood glucose and insulin levels, antioxidant enzymes, lipid peroxidation, pyruvate kinase (PK), lactate dehydrogenate (LDH) and distribution of protein kinase C (PKC) in heart, muscle and brain tissues of the alloxan induced diabetic rats and to see whether the treatment with SOV and Trigonella is capable of reversing these effects.

Methods: Diabetes was induced by administration of alloxan monohydrate (15mg/100gm b.wt.) and rats were treated with 2IU insulin, 0.6mg/ml SOV, 5% Trigonella in the diet and a combination of 0.2mg/ml SOV with 5% Trigonella separately for 21 days.

Results: Present results show that the action of fenugreek in lowering blood glucose levels is almost comparable to insulin effect. Rats treated with combined dose of vanadate and Trigonella had glucose levels comparable to controls, similar results were obtained with the activities of PK, LDH, antioxidant enzymes and PKC in diabetic rats.

Conclusion: Our results showed that lower doses of vanadate (0.2mg/ml) could be used in combination with Trigonella to effectively counter diabetic alterations without any toxic side effects.

HEMATOLOGICAL AND HISTOPATHOLOGICAL STUDIES OF ENDOSPERM RICH FRACTION OF FLAXSEED IN CHICKS

J. Rajesha[1*], A. Ranga Rao[2] , M. Karunakumar[3] and G.A. Ravishankar[2]

[1]Department of Biochemistry, Yuvaraja's College, University of Mysore, Mysore, India
[2]Plant Cell Biotechnology Department, Central Food Technological Research Institute, Mysore, India
[3]Department of Studies in Biochemistry, University of Mysore, Mysore, India

Key Words: Flax seed, endosperm rich fraction (ERF), layers, hematology, histopathology, and fatty liver

Introduction: Flaxseed is obtained from *Linum usitatissimum* belonging to family linaceae, commonly known as linseed. It is an important oilseed crop grown around the world for its oil and fiber and has been consumed as an ingredient in various food formulations and currently has a high demand in food industries and nutraceutical applications. Flaxseed has been playing a major role in the field of diet and disease research due to its potential health benefits associated with □-linolenic acid (57%) and its rich phytoestrogens or lignans.

Background: The objective of the present investigation was to elucidate the hematological and histopathological effects of endosperm rich fraction (ERF), a byproduct of dehulling processing of flaxseed, in laying hens.

Methods: Eighty, 32 weeks old, Single Comb White Leghorn (SCWL) laying hens were subjected to 4 dietary treatments namely Control, 5% ERF, 10% ERF and 20% ERF. At the end of 4^{th} week, all the groups were examined for their hematological and histopathological studies.

Results: There was a linear relationship between feed consumption and body weigh decrease between these parameters and increase in ERF treatments. The weight of the birds decreased was directly proportional to concentration of ERF in the diet. Packed cell volume (PCV) and red blood cell (RBC) counts had negative significant ($p<0.05$) linear relationship with increased level of ERF. There was a unit decrease in PCV and RBC counts with increase in ERF in the diet. Grossly, the livers of birds of groups fed with 20% of ERF were enlarged, pale in colour, soft in consistency and were having petechial haemorrhages with fat and fibrin deposits. Histopathologically, livers of group 20% ERF showed fatty infiltration, haemorrhages and mass of eosinophilic materials. The vacuoles coalesced to create clear space that displaced the nucleus to the periphery of the cell.

Conclusions: It was concluded that addition of dietary constituents from ERF of layer chicks not only resulted in increase in MCH and PCV but also in marked macroscopic and microscopic changes in liver.

EFFECT OF AEGLE MARMALOSE ON THE ACTIVATION OF INSULIN-STIMULATED GLUCOSE TRANSPORT AND GLUT-4 PROTEIN AND INHIBITION OF TNF-A AND NITRIC OXIDE RADICALS IN VITRO AND IN VIVO

Sabu M[2], Kuttan R.[1], Priya T.T.[1] , Kuttan G.[1], Nazamid S.[2]

[1]Department of Biochemistry, Amala Cancer Research Center, Thrissur, Kerala, India
[2] Faculty of Food Science and Technology, Universiti Putra Malaysia, Sri Sredang, Selangor, Malaysia

Key Words: Aegle marmelos, Glucose transport, Tumour necrosis factor, Glut-4 protein

Background: Blood glucose homeostasis is regulated by appropriate insulin secretion and glucagon production. Insulin enhances glucose uptake by muscle cells and fat cells and thereby suppresses hepatic glucose production. Family of glucose transporters (GLUT) mediates glucose transport across the cell membrane, and the GLUT-4 is the insulin sensitive glucose transporter. Both animal and human studies have suggested that there is a decrease in insulin- mediated glucose transport in diabetes mellitus. TNF- α has been shown to be involved in the development of insulin resistance and has been shown to be higher in the diabetic patients. The increase in glucose transport activity that occurs in response to their prolonged exposure to insulin, and evaluate the roles of herbal extracts on GLUT-4 protein.

Objective: The purpose of the present study is to investigate the effect of plant extract of Aegle marmelos on glucose transport and glucose transporter protein (Glut-4). We have also investigated the effect of plant extracts on the inhibition of nitric oxide and TNF-α in vivo as well as in vitro in normal and diabetic animals.

Methods: Extract was given once daily for ten days and glucose uptake was studied by incubation of rat gastrocnemius muscle (GC-muscle) and diaphragm with D-[U-14C] glucose. It was found that treatment with A. marmelos significantly increased the glucose transport in the presence and absence of insulin. Different concentrations of the homogenate (10, 20, 40 and 80μg/ml protein) were used for the estimation of GLUT-4 protein using ELISA method. Bioassay is designed on the basis of the cytotoxicity of TNF-α, to L929 cell lines in cultures. For the in vivo study, the extract (100mg/kg) was given once daily for 10 days to the normal and diabetes rats. In the in vitro study, macrophages were cultured with or with out lipopolysaccharide (LPS) (inducers of TNF-a) in the presence and absence of A. marmelos.

Results: The concentration of the GLUT-4 protein was found to be significantly higher in the homogenate of GC-muscles of rats treated with A. marmelos compared to non-treated groups. The results revealed that A. marmelos treatment in diabetes may increase the expression of GLUT-4 protein and thereby increasing the glucose uptake. Treatment with A. marmelos inhibited the

production of TNF-α and NO by the LPS activated macrophages. Addition of extracts also reduced the cytotoxicity induced by TNF-α in the L929 cells and administration of these extracts reduced the levels of TNF-α, which were elevated by 2- fold in the streptozotocin diabetic mice.

Conclusion: These observations conclude that extracts of A. marmelos could activate glucose transport produced by GLUT-4 protein. Moreover it could down regulate the levels of TNF-α and nitric oxide indicating its role in reducing the insulin resistance.

RESVERATROL: A PANACEA?

Shazib Pervaiz

National University of Singapore, Singapore

Key Words: wine polyphenol, resveratrol, Na^+/H^+ exchanger, and carcinogenesis

The diverse biological properties of the wine polyphenol, Resveratrol (RSV), have been extensively reported over the past decade or so. It's presence in grapes and wines have also been linked to the *"French paradox"*-wine consumption associated with cardio-protection. One aspect of RSV biology that has generated tremendous interest is its reported cancer preventive activity at least in animal models of carcinogenesis. Along these lines, we reported apoptosis inducing activity of RSV in human tumor cells via mechanisms involving up-regulation of the death receptor CD95-CD95L signaling pathway. Furthermore, in an attempt to understand the molecular basis for the apoptosis inducing activity of RSV, we resorted to 2D proteomics approach and pulled out an increase in cleaved lamin A in lysates from RSV treated HCT116 colon carcinoma cells. Biochemical analysis showed this to be linked to RSV-mediated activation of caspase 6. More recently, we have identified a novel transcriptional target of RSV, downregulation of which is associated with increased sensitivity to apoptosis inducing agents. Treatment of cells with RSV resulted in downregulation of the promoter activity as well as protein

expression of the pH regulator Na^+/H^+ exchanger, increased expression and activity of which have been linked to various processes of carcinogenesis. Intriguingly, this effect of RSV was mediated by a significant increase in intracellular ROS production, in particular hydrogen peroxide. In a separate set of experiments we have linked the biological activity of RSV to inhibition of the sphingosine kinase lipid-signaling pathway, which has broader implications for its anti-inflammatory as well as anti-cancer properties. These data provide novel insights into the biology of this remarkable compound and indicates its ability to function both as a pro- and anti-oxidant, depending upon its concentration and the cell type.

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NOVEL USE OF GRAPE SEED EXTRACT AND (Δ) TOCOTRIENOL AS SUPPLEMENTS IN INDUCTION OF PRO-INFLAMMATORY CYTOKINES IN MICE LIVER

Shradha Sodhani*[1], Dibyendu Dutta[2], Owen Kelly[1], Shanil.Juma[1], Parakat Vijayagopal[1], Laura Hanson[2], Nathaniel. Mills[2], Victorine Imrhan[1]

[1]Department of Nutrition and Food Sciences, [2]Department of Biology, Texas Woman's University, Denton, Texas, USA

Key Words: Fatty liver, lipid homeostasis, inflammation, grape seed extract polyphenols and delta tocotrienol supplementation.

Background: Nonalcoholic fatty liver disease (NAFLD) is a clinicopathological condition consists of a disease spectrum ranging from simple triglyceride accumulation in hepatocytes -hepatic steatosis to hepatic steatosis with inflammation (steatohepatitis), fibrosis, and cirrhosis (Neuschwander-Tetri B. A et al, 2003). NAFLD is the most frequent cause of abnormal liver function tests in the US (Clark JM 2003, Angulo et al 2002) affecting approximately 30 million Americans. Metabolic syndrome is a complex condition with all the above conditions either in part or in entirety, calls for urgent attention for therapy. NAFLD exhibits many factors and symptom is not limited to oxidative stress and subsequent lipid peroxidation, proinflammatory cytokines, adipokines and mitochondrial dysfunction (Albano et al, 2005).Excessive central adiposity permissive to insulin resistance and may make a major contribution to fat accumulation in hepatocytes. Hepatocytes represent about two-thirds of the total cell population in the liver. The

remaining population of nonparenchymal cells is diverse and includes Kupffer cells, the sinusoidal endothelial cells, stellate cells (fat storing), liver resident dendritic cells and lymphocytes.

The inflammatory infiltrate usually consists of mixed neutrophils and lymphocytes and predominates in liver (Angulo et al, 2002). Studies have found that hepatic activation of NF-κB in mice, via overexpression of IκB kinase β (IKK-β) induces insulin resistance in the liver and signs of systemic inflammation (increase in serum IL-6) and insulin resistance in skeletal muscle (Cai. D et al, 2005). Similar changes can be induced by feeding a high fat diet to mice (Cai. D et al, 2005, Kotronen. A et al, 2008).Thus, in mice fed a high diet, hepatic insulin resistance can be induced without inducing inflammation in adipose tissue but whether these data have relevance for disease is unknown. At present, pharmacological agents are available for treatment; however side effects of these drugs emphasize the need for natural supplements.

Objective: To evaluate the novel use of bioactive agents like delta tocotrienols and polyphenols from grape seeds in various dosages as supplements in high fat diet separately and in combination to establish the effectiveness for reversing or reducing pro-inflammatory condition in mice liver.

Method: Sixty four C57/BL/6J male mice, aged 5 weeks old were obtained from Jackson Laboratories (Maine, US). Mice were housed with 4 animals per cage, with 8 mice were placed in 8 groups in the Texas Woman's University facility 20 weeks. All animals were cared for

in accordance to the IUCAC guidelines of Texas Woman's University (Denton, TX). A commercially available high-fat diet, found to mimic the actual Western diet most closely was chosen. It is tailor-made for mouse models susceptible to obesity and metabolic syndrome. The High-fat diet(HFD) (Harlan Teklad # TD88137) and the normal-fat diet for control group (Harlan Teklad #TD 09752) for mice were used. The body weights were recorded twice per week. All groups had access to *ad libitum* diet and water. The supplement diet were made, by addition of δ-tocotrienols (δT3) 0.025% and 0.05% DeltaGold® 70 (American River Nutrition, Inc) grape seed polyphenol (GSP) 1% and 2% (Leucoselect® purchased from Indena S.p.A), to the HFD . At the end of 20 weeks, all animals were fasted for 10 hours and euthanized. Two hundred milligrams samples of liver tissue were fixed in modified Davidson's fixative, were cut into six micrometer (μm) sections were later used for Imnmunohistochemistry. The primary antibodies used were anti-mouse, CD4 (2:500) goat polyclonal IgG (SantaCruz Biotechnology Inc.CA. USA) and CD8- alpha (2:500) (Millipore, CA.USA). Secondary antibodies tagged by DyLight 594 donkey anti mouse IgG (1:100) (Jackson Immuno Research Inc, PA, USA) for fluorescence detection under the confocal microscope (Nikon, Inc). All samples will be analyzed in duplicate and a One-way ANOVA ($p \leq .05$) will be considered as significant. (SPSS, version 15).

Results: At the end of 20 weeks animals were sacrificed and histopathological analyses were performed for biomarkers in liver typical of human NASH. HFD led to increased triglyceride accumulation in the liver and induced histopathological features of

human NASH including hepatic steatosis, ballooning, inflammation and fibrosis. Mice on HFD also reached high total body weight and relative liver weights. The adipose tissue weight for both mesenteric and peritoneal depots were also representative of visceral and omental adiposity in relation to adipose tissue weight that characterize NASH in humans. The microscopic immunofluorescent pictures of the mouse livers, in the 6 supplemented groups compared to the 2 control groups have differences in cells staining for CD4+ and CD8+ content, higher in HFD and lowest in grape seed extract groups.

Conclusion: Supplements including polyphenols from grape seed extract and delta tocotrienol both alone and in combination can ameliorate many of the histopathological features of human NASH in C57BL/6J mice. The various dosages of grape seed extract may be beneficial in reversing or reducing inflammation of liver steatosis.

TEA POLYPHENOLS ARE INHIBITORS OF ENERGY TRANSDUCING ENZYMES

Steven B. Vik

Southern Methodist University, Department of Biological Sciences, Dallas, TX

Key Words: Theaflavin, Superoxide, ATP synthase, Complex I, NDH-1, NDH-2, Oxidative phosphorylation

Background: During the production of black tea, theaflavins, a special class of polyphenols, are produced from catechins. The major theaflavins in black tea are theaflavin(TF1), theaflavin-3-gallate (TF2A) and theaflavin-3'-gallate (TF2B), theaflavin-3,3'-digallate (TF3). Possible benefits of the consumption of tea polyphenols have been analyzed in a variety of studies, including in vitro, in vivo, and epidemiological ones. In addition to their inhibition of various enzymes, tea polyphenols from both green and black tea are active chemically as anti-oxidants. In mammals, most superoxide is produced in the mitochondria, and principally from the electron transport chain. Many inhibitors of the electron transport chain cause an increase in the rate of superoxide production, and inhibotrs of the ATP synthase can have the same effect.

Several plant polyphenols, including tea catechins, have been shown to be inhibitors of the mitochondrial ATP synthase. In this paper we have extended previous work by showing that

polyphenols from black tea, the theaflavins, can also inhibit the ATP synthase from E. coli, and that they are more potent than is EGCG. In similar fashion, these compounds are also inhibitory towards Complex I (NDH-1) of the electron transport chain. However, the superoxide scavenging capacity appeared to compensate for any possible excess production of superoxide caused by these inhibitory actions.

Objective: To determine whether theaflavins were inhibitors of oxidative phosphorylation, and if so, whether this led to increased levels of superoxide production.

Methods: Theaflavins were isolated from black tea extracts. Membrane vesicles were prepared from E. coli cells. Rates of ATP hydrolysis, proton translocation, and NADH oxidation were measured in the presence or absence of the tea polyphenols. Inhibition constants were calculated from the data. The tea polyphenols were docked by computer onto molecular models of the F1 ATPase and Complex I using AutoDock 4.2.

Results: Four dietary polyphenols, theaflavin (TF1) and its three derivatives, theaflavin-3-gallate (TF2A), theaflavin-3′-gallate (TF2B) and theaflavin-3,3′-digallate (TF3), have been isolated from black tea, and their effects on oxidative phosphorylation and superoxide production in a model system (E. coli) have been examined. The esterified theaflavins were all potent inhibitors of

the membrane-bound ATP synthase. ATP-driven proton translocation was inhibited in a similar fashion, as was the purified F1-ATPase, indicating that the primary site of inhibition was in the F1 sector. Computer modeling studies supported this interpretation. All four theaflavins were also inhibitory towards the electron transport chain, whether through Complex I (NDH-1) or the alternative NADH dehydrogenase (NDH-2). Inhibition of NDH-1 by TF3 appeared to be competitive with respect to NADH, and this was supported by computer modeling studies. Rates of superoxide production during NADH oxidation were measured. Superoxide production was completely eliminated by the addition of about 15 μM TF3.

Conclusion: There appears to be a widespread inhibitory effect of tea polyphenols towards the oxidative phosphorylation machinery. Unlike many such inhibitors, this inhibition is not associated with an increase in superoxide production from the electron transport chain. Any potential increase in superoxide production appears to be compensated by the free radical scavenging ability of these polyphenols.

BITTER MELON EXTRACT ENHANCES INSULIN SENSITIVITY BY MODULATING FGF21 SIGNALING IN HIGH-FAT DIET FED MICE

Zhong Q. Wang, Yongmei Yu, Xian H. Zhang, Huating Li[1], Jianhua, Qin, William T Cefalu

Nutrition and Diabetic Research laboratory,
Pennington Biomedical research Center, LSU system. Baton Rouge, LA
[1]Department of Endocrinology and Metabolism, Shanghai Jiao Tong University Affiliated Sixth Peoples Hospital, Shanghai, China

Key words: Bitter melon, *Momordica charantia*, and insulin sensitivity

Background: Bitter melon (BM), *Momordica charantia*, has been used primarily as an alternative therapy for diabetes in Asia, Africa, and Latin America, because of its hypoglycemic activity. Metabolic and hypoglycemic effects of BM extracts have been demonstrated in cell culture, animal, and human studies, but the precise mechanism of BM extract action of improving glucose metabolism is largely unknown. Recently study showed that fibroblast growth factor 21 (FGF21) is an atypical member of the FGF family that functions as an endocrine hormone. Pharmacologic studies show that FGF21 has broad metabolic actions in obese rodents and primates such as enhancing insulin sensitivity, decreasing triglyceride concentrations, and body weight.

Objective: We sought to investigate the effect of bitter melon extract (BM) on glucose metabolism, insulin sensitivity, and FGF21 signaling in high-fat diet fed mice.

Methods: Thirty male C57BL/6J mice were randomly divided into high-fat diet group (HFD) and two bitter melon extract groups (BM-V and BM-SB). Food intake and body weight were recorded weekly. Fasting plasma FGF21, HOMA-IR, and body composition were determined at week 12. Liver FGF21 and related gene expression were measured by real time RT-PCR assay.

Results: Body weight was significantly lower in BM-V mice than in BM-SB and HFD mice. Food intake was much higher in BM-SB group than in HFD and BM-V groups, but there was no difference between HFD and BM-V animals. Plasma FGF21, glucose, insulin, HOMA levels, IPGTT, and IPITT were significantly lower in BM-V mice than in HFD mice ($P<0.05$), these parameters did not have any significant difference between BM-SB and HFD groups besides IPITT. Moreover, there were closely correlations among body weight, visceral fat, fat mass and insulin concentration ($r = 0.57 – 0.80$, $P < 0.001$), but not among FGF21 levels, body weight, HOMA-IR and fat mass. FGF21 levels in plasma and liver as well as liver triglyceride content were significantly lower in BM-V mice than in HFD mice ($P<0.05$), In additional, gene expression of FGF21 in the liver was significantly down-regulated, FGFR1 and

FGFR3 were greatly up-regulated in both BM groups than in HFD group ($P < 0.05$ and $P < 0.01$).

Conclusion: High-fat diet feeding results in insulin and FGF21 resistance in mice. BM extracts significantly enhancing insulin sensitivity may be mediated by modulating FGF21 signaling in the HFD mice.

PART THREE

ROLE OF FUNCTIONAL FOODS IN METABOLIC SYNDROME AND RELATED DISORDERS

ENHANCED GREEN TEA POLYPHENOLS IMPROVE SKIN PROPERTIES OF WOMEN

Carolyn Moore[1] , Ulrike Heinrich[2], Hagen Tronnier[2], Wilhelm Stahl[3]

Texas Woman's University, Houston, Texas
[2]Institute for Experimental Dermatology, Witten-Herdecke University, Witten, Germany
[3]Institute for Biochemistry and Molecular Biology, Heinrich-Heine-University Düsseldorf, Düsseldorf, Germany

Key Words: green tea polyphenols, skin

Background: Nutritive parameters influence skin condition and an optimal supply of macro- and micronutrients contributes to skin health. Skin structure, texture, thickness, density, hydration, color, and shielding properties vary depending on endogenous and exogenous factors. In-vitro and animal studies indicate that tea flavanols, when applied orally or topically, ameliorate skin reactions following UV exposure, including skin damage, erythema and lipid peroxidation. Few randomized controlled trials, however, have been conducted utilizing orally ingested green tea polyphenols to examine potential photoprotection and other skin health benefits.

Objective: The present study was designed to investigate the effects of repetitive intake of a beverage enriched with green tea

polyphenols on skin structure, skin properties, and sensitivity towards UV exposure.

Methods: Sixty females (40 – 65 years) were randomized to consume either a drink with green tea polyphenols (GT) or a control beverage (C) for 12 weeks. The GT group received 1432 mg of total polyphenols primarily distributed as 100 mg epicatechin (EC), 980 mg epigallocatechingallate (EGCG), and 240 mg epicatechingallate (ECG) per day. Photoprotection, skin structure, and skin function were measured at baseline (wk 0) and wk 12. Hydration measurements were carried out by corneometry, transepidermal water loss (TEWL) with the TEWA-Meter, skin surface with the Visioscan, UV-induced erythema by chromametry (a-values), and skin elasticity with the Cutometer. Analysis of skin thickness and density were made by ultrasound measurements. The O_2C-system was used to measure peripheral blood flow and oxygen saturation of hemoglobin. Blood samples were analyzed by HPLC for selected polyphenols. Descriptive statistics and pre-post differences were calculated and compared using the Wilcoxon signed-rank test. Percentage changes and p-values were determined ($p<0.05$ as significant).

Results: EGCG was the main flavonoid in the GT beverage and serum levels of EGCG were significantly elevated in the GT group after 12 weeks demonstrating absorption and systemic availability of GT polyphenols. Table 1 summarizes changes in skin

parameters over the 12 week study. Following exposure of the skin areas to 1.25 minimal erythemal dose (MED) of radiation from a solar simulator, UV-induced erythema was significantly decreased in the GT group.

Table 1. Percent Changes of Mean Skin Parameters from Baseline to 12 Weeks

Skin Parameter	**Greet Tea (GT) Beverage**	**Control (C) Beverage**
UV Protection: Minolta-a value	-25% * ¥	+ 9% *
Hydration, au	+ 17% * ¥	+ 5%
Transepidermal Water Loss (TEWL) g h-1 m2	-12% * ¥	-0.9%
Density mm2	+ 8% * ¥	+ 0.6 %
Capillary Blood Flow at 1 mm depth, au	+ 29% * ¥	+ 1%
Oxygen Saturation, %	+ 34% * ¥	-3%

* Significantly different from wk 0, $p < 0.05$

¥ Comparison between GT and C treatment groups significantly different, $p < 0.05$

au = arbitrary units

Skin hydration was significantly elevated following 12 wk of supplementation with GT extract and TEWL was significantly decreased which pointed to an increased barrier function of the

skin ($p<0.05$). Skin density significantly increased although skin thickness was not affected (data not shown). Capillary blood flow and blood oxygen saturation significantly increased by approximately 30% and 34%, respectively in the GT group.

Conclusion: Consumption of a beverage rich in tea flavanols contributed photoprotection and helped maintain skin structure and function. The beneficial effect of UV protection and blood flow were correlated with plasma levels of epicatechin. Thus, the underlying mechanism for the observed skin improvements with GT beverage consumption maybe related to increased microcirculation and the erythemal protection related to UV-light absorption by GT flavoids deposited in skin.

METABOLOMIC MARKERS OF LUPUS

Chandra Mohan

UT Southwestern Medical Center, Dallas, TX

Key Words: Metabolomic, Lupus, Essential Fatty Acids, Vitamins, Methyl, donors

Background: Systemic lupus erythematosus (SLE or lupus) is a systemic autoimmune disease where the body's B-lymphocytes and T-lymphocytes aggressively attack self tissues. As a consequence of the renal disease that ensues, morbidity and mortality in this disease is high. Our research over the past decade has focused on unraveling the molecular pathways and genetic triggers that cause lupus, using mouse models.

Objective: The overall objective of the research is to understand the pathogenic mechanisms underlying this disease. One recent tool we adopted to study this disease is a Metabolomic scan.

Methods: We undertook a metabolomic scan of sera from lupus patients (N = 20), comparing them to healthy control sera (N = 10), using a combined GC/MS and LC/MS based approach to identify small molecules after removing serum proteins.

Results: A total of 124 small molecules were noted to be aberrantly expressed (at p-values less than 0.05) in lupus serum,

clustered around a couple of distinct pathways. Overall, we noted a significant reduction in the serum levels of methyl group donors (choline, methionine), certain vitamins, essential fatty acids (including n3-PUFA and n6-PUFA), and all long chain fatty acids in lupus. In contrast, lupus sera exhibited significantly increased levels of medium chain fatty acids (mostly of dietary origin), lipid peroxidation products, and a couple of pro-thrombotic and inflammatory mediators.

Conclusion: These findings suggest that several of the metabolomic changes seen in lupus may be tackled nutritionally, including diets supplemented with essential fatty acids, methyl group donors, anti-oxidants, and selected combinations of vitamins. In addition to testing these predictions, a couple of the metabolites are being examined as potential markers of disease severity. Collectively, these studies may help identify a novel class of biomarkers in SLE, and pave the way towards nutritional modulation of the disease.

ROLE OF DIETARY COMPONENT CURCUMIN IN CANCER PREVENTION

Manjula Vinayak

Biochemistry and Molecular Biology Laboratory, Department of Zoology, Banaras Hindu University, Varanasi, India

Key Words: Curcumin, IL-1a, Expression, Promoter site, Antioxidant enzymes, Cancer

Background: Turmeric belongs to spices and is used essentially during cooking of almost all Indians foods. It has got importance in Indian ethics and culture as use of turmeric is prohibited during cooking of food for a few days after death of a family member. This indicates the importance of turmeric in health. Curcumin is the active component of turmeric and is known to causes anti-inflammatory and antioxidant activities.

Objective: The present work is aimed to test the role of dietary component curcumin in cancer prevention.

Methods: Dalton's Lymphoma bearing AKR mice (DL mice) were used for the study. Five different groups of mice included control DL mice, vehicle treated DL mice and DL mice receiving three doses of curcumin in three groups. Curcumin was given intraperitoneally for nine consecutive days starting from the next day of DL implantation to healthy adult male mice. Morphological

changes and life span of mice were recorded. Further, the effect of curcumin administration on biochemical parameters was checked after sacrifice of mice on day eighteen of tumor implantation. Cell proliferation and cell survival was checked as a parameter of tumor growth.

Results: Curcumin administration to DL mice leads to decrease in accumulation of ascite fluid and causes increase in the life span of mice in a dose dependent manner. The activity of LDH as an anaerobic metabolic enzyme was found to decrease in DL mice receiving curcumin treatment. This activity is taken as a marker of tumor progression. Further, a dose dependent suppression of tumor maintenance was noticed by decrease in cell proliferation and cell survival in curcumin treated DL mice. Curcumin enhances the antioxidant defense system of the organism in a dose dependent manner as, observed by increase in expression and activities of antioxidant enzymes: SOD, CAT, GPX. The oxidative stress of DL mice was reduced as measured by lipid peroxidation and protein carbonylation capacity. Proinflammatory cytokines are known to induce as well as maintain tumor progression by inducing genetic instability due to synergistic interaction of various mediators of inflammation, leading to ROS generation. In the present study the expression and secretion of proinflammatory cytokine IL-1a was reduced in DL mice after receiving curcumin treatment which contributes to down regulate tumor progression. The analysis of the signaling pathway showed that the regulation of expression of

IL-1a was mediated via NF-kB binding as well as via IL-1a specific AP1 binding to respective promoter sites as curcumin administration to DL mice was able to decreases the binding of transcription factors to both the promoter sites.

Conclusion: The results demonstrate that dietary component curcumin prevents cancer progression by preventing genetic instability by enhancing organism's antioxidant defense system and by suppression of proinflammatory cytokine IL-1a via both NF-kB as well as AP1 dependent pathways. The present work highlights the importance of curcumin in prevention and management of cancer.

EFFECTS OF KOREAN PEARS, *PYRUS PYRIFOLIA CV. SHINGO*, ON ALCOHOL DETOXIFICATION

Mihi Yang[1], Jong Y. Park[2]

[1]College of Pharmacy, Sookmyung Women's University, Seoul, Republic of Korea
[2]Division of Cancer Prevention and Control, H. Lee Moffitt Cancer Center and Research Institute, Tampa, FL, USA

Key Words: metabolic syndromes, Korean pears, and alcohol consumption

Background: Chronic alcohol consumption is a well known risk factor for various metabolic syndromes, even cancer. Therefore, safe and efficient agents for alcohol detoxification have been demanded for many years. In addition, about half of northeastern Asians genetically lack activity of ALDH (Acetaldehyde Dehydrogenase), an enzyme involved in alcohol metabolism. People without ALDH gene often experience metabolic syndromes even after drinking a small dose of alcohol. Korean pears (*Pyrus pyrifolia cv. Shingo*) have been used as a traditional medicine for alleviating syndromes from alcohol hangover. However, their mechanisms for alcohol detoxification are not clearly determined.

Objective: The goal of this study is to investigate how Korean pears involved in alcohol detoxification.

Methods: To investigate a role of Korean pears in ADH (alcohol dehydrogenase) and ALDH activities, we performed *in vitro* and *in*

vivo studies using a gene alternated animal model. For pharmacokinetic (PK) analyses, 10 ml/kg bw (≈ one pear/60 kg of human) of the pears was administered to ***male C57BL/6*** *(Aldh*2 +/+) and *Aldh*2 -/- mice. Ethanol (1 g or 2 g /kg bw, ≈ average alcohol drinking levels in adult Koreans) was administrated to the mice via gavage. Korean pear impacts on alcohol consumption were assessed by level of alcohol or acetaldehyde in blood.

Results: Results from *in vitro* study suggested that Korean pears stimulated both of ADH and ALDH activities, e.g. 2-3 and 1.3 folds, respectively: The ADH activity was increased in a dose-dependent manner by the pear doses (2-8 v/v%: $p<0.05$). In animal PK studies, we observed that levels of blood ethanol in all mice were decreased after Korean pear intake, compared to non treated mice. However, only *Aldh2* deficient mice showed that Korean pears increased by 40 % of acetaldehyde. Thus, we found significantly different response to the pears intake between *Aldh2* normal and deficient genotypes.

Conclusion: Our data demonstrate that the Korean pears stimulate both of ADH and ALDH activities through *in vitro* and *in vivo* studies. Due to increase of toxic acetaldehyde in *Aldh2* deficient mice by pear intake, we recommend that the Korean pears may be useful in individuals with *ALDH2* normal types but not ones without *ALDH2* for human application.

ACACIA GUM AN EMERGING FUNCTIONAL FOOD IN PATIENTS WITH SYMPTOMATIC UREMIA

Aamir Jalal Al Mosawi

Department of Pediatrics and CME Center,
University Hospital in Al Kadhimiyia, Baghdad, Iraq

Keywords: Acacia gum and uremia

Background: Few papers reported a beneficial effect of acacia gum (AG) supplementation of low protein diet in symptomatic uremia .AG supplementation of a low-protein diet (LPD) provided patients treated with intermittent peritoneal dialysis (IPD) a long period of dialysis freedom and improved wellbeing .AG, the dried gummy exudates of the stem and branches of acacia trees (Senegal family leguminous), is a complex polysaccharide that is generally recognized as safe by the US FDA and widely used in the production of foods. AG is known to increase fecal nitrogen excretion and lower serum concentrations of urea and other retained metabolites in patients with chronic renal failure (CRF), animals, normal human subjects and patients with cirrhosis . There are no limitations to the use of AG as a food additive as the experimental evidence of safety demanded by international food safety committees has already been met. The aim of this study the function of AG in lowering urea elevations in patients with CRF and symptomatic uremia who were reluctant to undergo dialysis

and to achieve improved wellbeing without dialysis by the use of AG supplementation and a LPD.

Methods: From December 2005 to October 2009, 9 patients 5 males and 4 females, with various renal disorders (Biopsy-proven focal segmental glomerulosclerosis (FSGS), biopsy proven membranous glomerulonephritis, diabetic nephropathy, cystinosis, bilateral renal stone disease, and infantile polycystic disease, chronic GN including familial GN) and symptomatic uremia were referred for treatment due to their unwillingness to undergo dialysis. Ages ranged from 3.5 year to 72 years (mean 28.8).All patients considered dialysis in their circumstance to be associated with an unacceptable degree of discomfort and Suffering. Five patients have severe uremic symptoms. All of them were unable to walk independently; one of them (patient-1) has undergone one session of intermittent peritoneal dialysis and was referred 3 days after dialysis, Four patients (2, 3 and 4) refused dialysis session and were enrolled at the stage when dialysis was definitely indicated to maintain health and both patients had marked weakness. Patients (5) aged 72 years with chronic ischemic heart disease (on oral and sublingual nitrates) was considered by the dialysis team as a terminal patient and warned the family of the possibility of death during or after dialysis. Patient (2) with diabetic nephropathy was unable to stand and walk unaided. Four patients had less severe uremic symptoms. None of patients had with a life threatening uremic complication (e.g. gastrointestinal

bleeding, encephalopathy) on referral. They were followed for a period ranging from 8 weeks to 4 years. Dietary proteins were restricted to 1 g/kg/day for patients under the age of 10 years, and 0.5 g/kg for patients over the age of 10 years with at least 50% of the total intake given as egg albumin. Protein and phosphorus restriction was primarily achieved by restricting meat, poultry, fish, milk, cheese and yogurt. Additional restrictions were made on potassium-rich foods during elevation of serum potassium above 5 mmol/l. Powder AG 1 g/kg/day (maximum 75 g) was supplied in 2–3 divided doses (diluted with the desired amount of water with or without the addition of sugar and juices) by a pharmacist.

Results: All patients enrolled experienced amelioration of symptoms of uremia with improved general wellbeing in association with lowering of urea levels and creatinine during the period of therapy. Mean blood urea level before the start of therapy was 137.45 mg/dl (Mean BUN 61 mg/dl).After 2-4 weeks of therapy mean blood urea level was 78.5 mg/dl (Mean BUN 34.5 mg/dl). After 5-10 weeks of therapy mean blood urea level was 60 mg/dl (Mean BUN 25.7 mg/dl).Figure-1 shows the mean blood urea and BUN during therapy. It was possible to follow patient 7, 8 and 9 for more than 4 years on this therapy. They continued to experience low urea levels and didn't develop any uremic symptoms.

Conclusion: Previous attempts to treat the symptomatic uremia by a way other than RRT relied entirely on reducing nitrogen intake by restricting dietary proteins. The possibility of lowering serum urea levels by an approach relying on increasing fecal nitrogen excretion by the use of fermentable dietary fibers (Functional food) has been suggested. The need for an oral urea lowering agent to control symptomatic uremia is relatively straight forward. Acacia gum is a potential functional oral urea lowering agent) food (for use in patients with symptomatic uremia is associated with an important urea lowering effect. A controlled trial investigating the urea lowering effect of AG in symptomatic uremia is highly recommended.

STUDY ON THE HEPATOPROTECTIVE EFFECT OF PLEUROTUS FLORIDA (OYSTER MUSHROOM) AGAINST PARACETAMOL INDUCED LIVER DAMAGE IN WISTAR ALBINO RATS

Sultana N, Sumy A K, Jahan N

Department of Physiology, Sir Salimullah Medical College, Mitford, Dhaka, Bangladesh

Key Words: Oyster mushroom, Hepatoprotective, Liver marker enzymes

Backgroud: Liver is an important metabolic organ. Its damage can be occurred due to prolong use of higher doses of some drugs, exposure to some chemicals or infectious agents. But liver protective drugs are not available in modern medicine. Many hepatoprotective herbal medicines are often used in the treatment of liver damage.

Objectives: This experimental study was carried out to observe the hepatoprotective effect of Oyster mushroom (Pleurotus florida) against paracetamol induced liver damage in Wistar albino rats.

Method: This experimental study was carried out in the Department of Physiology, Sir Salimullah Medical College (SSMC), Dhaka, Bangladesh from 1st July 2009 to 30th June 2010.

A total number of 34 Wistar albino rats, age ranged from 90 to 120 days, weighing between 150 to 210 grams were selected for the study. After acclimatization for 14 days, they were divided into two groups, control group (Group A_1) and experimental group (Group B- mushroom pretreated and paracetamol treated group). Control group again subdivided into group A_1 (baseline control) and group A_2 (paracetamol treated group). All groups of animals received basal diet for 30 consecutive days. Group A_1 consisted of 10 rats, received propylene glycol (2 ml/kg bw, po) only on 30^{th} day. Group A_2 consisted of 14 rats, received single dose of paracetamol suspension (750 mg/ kg bw, po) only on 30^{th} day. Group B consisted of 10 rats, received mushroom extract (200 mg/ kg bw, po) for 30 consecutive days and paracetamol suspension (750 mg/ kg bw, po) only on 30^{th} day. All the animals were sacrificed on 31^{st} day. Then blood and liver sample were collected. Measurement of liver marker enzymes and total bilirubin level in serum, assessment of malondialdehyde concentration in liver tissue homogenate were done by using standard laboratory kits. Histology of liver was also done by using standard laboratory procedure. The statistical analysis was done by one way ANOVA and Bonferroni test as applicable.

Result: The mean serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin and in the liver tissue homogenate malondialdehyde (MDA) concentration were significantly ($p< 0.001$) higher in paracetamol treated group in

comparison to those of baseline control group. Again, the mean serum AST ($p<0.05$), ALT ($p<0.05$), total bilirubin ($p<0.001$) and in the liver tissue homogenate MDA concentration ($p<0.001$) were significantly lower in mushroom pretreated and paracetamol treated group (experimental group) when compared to those of paracetamol treated group (control). In this study, histological examination of liver revealed abnormal histological findings in 100% of rats in paracetamol treated rats. Again, 80% of rats in mushroom pretreated and paracetamol treated group showed almost normal structure and 20% showed mild histological changes in liver.

Conclusion: The present study revealed the hepatoprotective effect of Oyster mushroom (Pleurotus florida) against paracetamol induced liver damage in Wistar albino rats.

A COMPARATIVE STUDY OF THE ANTIPROLIFERATIVE EFFECT OF KOHLRABI AND GREEN CABBAGE ON COLORECTAL CANCER CELL LINES

Sandra Rizk and Valia Zarzour

Department of Natural Sciences, Lebanese American University, Beirut, Lebanon

Key Words: colorectal cancer, kohlrabi, cabbage, antitumor

Background: Brassica oleracea of Capitata group and Brassica oleracea of Gongylodes group known as green cabbage and kohlrabi respectively belong to the same cruciferea family. Varieties of Brassica Oleracea are rich sources of glucosinolates which hydrolyze into isothiocyanates. Isothiocyantes have reported their anticancer activity in vitro and in vivo and have shown growth-inhibiting and apoptosis-inducing activities in several cancer cell lines.

Objective: The aim of this study is to compare the effect of green cabbage and kohlrabi ethanolic extracts on colorectal cancer cell lines HT-29 and Caco2.

Methods: Kohlrabi and Green Cabbage plants were cultivated in Zahleh, blended and incubated in 90% ethanol. The extract was then concentrated and filtrated. Ht-29 and Caco-2 cells were

incubated with different concentrations of plants extracts (10, 20 and 40µg/µl), for 24 and 48hrs. Proliferation of the cells was assayed using WST-1, apoptosis induction was assessed using cell death Elisa kit, and the cytotoxicity of the extracts was evaluated using Trypan blue exclusion method.

Results: After 48 hours, proliferation of both cell lines was reduced to 8% upon the addition of 40µg/µl of kohlrabi extract; however it decreased to 1% upon the addition of cabbage extract. This concentration of kohlrabi resulted in 10% cytotoxicity on HT-29 and Caco2 while that of cabbage extract recorded 17% on both cell lines. 40µg/µl of kohlrabi extract caused a 2 times fold increase in apoptosis in HT-29, while a 1.7 fold increase in Caco2 cells. On the other hand, 40µg/µl of cabbage extract caused a 1.9 fold increase in apoptosis in HT-29 compared to 1.5 fold increase in Caco2.

Conclusion: Kohlrabi similar to green cabbage is efficient in inhibiting proliferation and inducing apoptosis without significant necrotic activity in colorectal cancer cell lines HT-29 and Caco2.

ASSOCIATION OF PLASMA CYSTEINE AND HOMOCYSTEINE CONCENTRATIONS WITH INSULIN RESISTANCE IN LOW SOCIO-ECONOMIC PREGNANT WOMEN

Sujatha P, Hemalatha R, Balakrishna N, and Ramalakshmi B

Lipid Chemistry Division, National Institute of Nutrition, Indian Council of Medical Research, Hyderabad, Andhra Pradesh, India

Key Words: Plasma cysteine, homocysteine, glutathione, high risk and Insulin resistance

Background: Several studies revealed that women with moderately elevated plasma homocysteine concentrations are associated with pregnancy complications including preeclampsia and may have enhanced risk for the development of cardiovascular related problems in later life. Furthermore, thiols such as intracellular glutathione and extracellular cysteine may also play a vital role in antioxidant cellular defense. In addition to plasma thiols, Insulin resistance during pregnancy was associated with several metabolic complications and it is one of the important components in metabolic syndrome. Though there are reports on the association of plasma homocysteine with Insulin resistance but limited data is available on the circulating levels of cysteine and glutathione in pregnant women, where these thiols were interrelated and may also play a major role in oxidative stress.

Objective: To evaluate the status of plasma thiols in different gestational ages including high risk pregnancy and to examine the relationship between amino thiols and Insulin resistance in low socioeconomic pregnant women.

Methods: The study was approved by the Institutes ethical committee, 117 subjects were enrolled cross sectionally from low socioeconomic status pregnant women, who were attended to the antenatal clinic at Niloufer hospital (Hyderabad) during the year 2008-2009. Women were recruited in three gestational periods; 21 women in 1st trimester (<12 weeks) 38 subjects in second trimester (20-28 weeks), and 58 women in third trimester (36-38 weeks). Among 117 pregnant women, 35 pregnant women were at high risk in the gestation period of >20 weeks.

Methods: Plasma thiols such as cysteine, cysteinyl-glycine, homocysteine and glutathione concentrations were measured using High Performance Liquid chromatography (HPLC) with fluorescence detection. Insulin resistance was calculated using fasting Insulin (µU/ml) and fasting glucose (mmol/L) the homeostasis model assessment (HOMA-IR). Because of wide variations in the concentrations of thiols, the values were transformed to log values and tested (t test) by using the software SPSS 14 version. Values are used (mean ±S.D). P value <0.05 was considered as significant. Pearson correlations were calculated for the means to study the relationship.

Results: There was a statistically significant difference in mean levels of plasma cysteine, homocysteine, and glutathione between 1st trimester (103.44±16.1, 6.62±2.0 and 2.57±0.47□moles/L respectively) and 2nd trimester (146.34±42.2, 9.48 ±2.79 and 3.25±0.91□moles/Lrespectively), whereas plasma cysteinyl-glycine did not show any significant difference between 1st and 2nd trimester. Mean levels of four thiols were significantly different between normal and high risk pregnancy; plasma cysteine and homocysteine levels were significantly ($P<0.001$, $P<0.01$ respectively) lower in normal pregnancy (113.05±21.9, 7.35±2.1□moles/L) compared to high risk (197.51±5, 8.49.02±3.2□moles/L). In contrast to cysteine and homocysteine, plasma cysteinyl glycine and glutathione levels were significantly ($P<0.05$) higher in normal pregnant women (22.54±2.5, 4.03±0.54□moles/L) compared to high risk (17.38±3.2, 2.56±0.77□moles/L). Among the four thiols, there was a posit! ive correlation between plasma cysteine and homocysteine ($r= 0.575$, $P<0.001$) and also between cysteinyl glycine and glutathione ($r=0.451$, $P<0.001$). With respect to insulin resistance, women with >2 HOMA-IR had significantly higher plasma cysteine 148.28±52.5□moles/L ($P<0.001$) and homocysteine 9.86±2.9□moles/L ($P<0.05$) levels than the women with <2 HOMA-IR (119.56±24.3, 7.64±2.4 □moles/L), whereas cysteinyl glycine and glutathione concentrations were not significantly different between >2 and <2HOMA-IR. With high prevalence of

Insulin resistance (45%), we observed a positive correlation of Insulin resistance with cysteine ($r=0.472$, $P<0.001$) and homocysteine ($r=0.333$, $P<0.01$) in these women.

Conclusion: The present study results indicate that women with higher cysteine and lower glutathione concentrations in pregnancy may be at higher risk for pregnancy associated complications. These results also show high prevalence of insulin resistance and HOMA-IR was positively associated with plasma cysteine and homocysteine.

STUDY ON THE HEPATOPROTECTIVE EFFECT OF PLEUROTUS FLORIDA (OYSTER MUSHROOM) AGAINST PARACETAMOL INDUCED LIVER DAMAGE IN WISTAR ALBINO RATS

Sultana N, Sumy A K, Jahan N

Department of Physiology, Sir Salimullah Medical College, Mitford, Dhaka, Bangladesh

Key Words: Oyster mushroom, Hepatoprotective, Liver marker enzymes

Background: Liver is an important metabolic organ. Its damage can be occurred due to prolong use of higher doses of some drugs, exposure to some chemicals or infectious agents. But liver protective drugs are not available in modern medicine. Many hepatoprotective herbal medicines are often used in the treatment of liver damage.

Objectives: This experimental study was carried out to observe the hepatoprotective effect of Oyster mushroom (Pleurotus florida) against paracetamol induced liver damage in Wistar albino rats.

Method: This experimental study was carried out in the Department of Physiology, Sir Salimullah Medical College (SSMC), Dhaka, Bangladesh from 1st July 2009 to 30th June 2010. A total number of 34 Wistar albino rats, age ranged from 90 to 120

days, weighing between 150 to 210 grams were selected for the study. After acclimatization for 14 days, they were divided into two groups, control group (Group A_1) and experimental group (Group B- mushroom pretreated and paracetamol treated group). Control group again subdivided into group A_1 (baseline control) and group A_2 (paracetamol treated group). All groups of animals received basal diet for 30 consecutive days. Group A_1 consisted of 10 rats, received propylene glycol (2 ml/kg bw, po) only on 30th day. Group A_2 consisted of 14 rats, received single dose of paracetamol suspension (750 mg/ kg bw, po) only on 30th day. Group B consisted of 10 rats, received mushroom extract (200 mg/ kg bw, po) for 30 consecutive days and paracetamol suspension (750 mg/ kg bw, po) only on 30th day. All the animals were sacrificed on 31st day. Then blood and liver sample were collected. Measurement of liver marker enzymes and total bilirubin level in serum, assessment of malondialdehyde concentration in liver tissue homogenate were done by using standard laboratory kits. Histology of liver was also done by using standard laboratory procedure. The statistical analysis was done by one way ANOVA and Bonferroni test as applicable.

Result: The mean serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), total bilirubin and in the liver tissue homogenate malondialdehyde (MDA) concentration were significantly ($p< 0.001$) higher in paracetamol treated group in comparison to those of baseline control group. Again, the mean

serum AST ($p<0.05$), ALT ($p<0.05$), total bilirubin ($p<0.001$) and in the liver tissue homogenate MDA concentration ($p<0.001$) were significantly lower in mushroom pretreated and paracetamol treated group (experimental group) when compared to those of paracetamol treated group (control). In this study, histological examination of liver revealed abnormal histological findings in 100% of rats in paracetamol treated rats. Again, 80% of rats in mushroom pretreated and paracetamol treated group showed almost normal structure and 20% showed mild histological changes in liver.

Conclusion: The present study revealed the hepatoprotective effect of Oyster mushroom (Pleurotus florida) against paracetamol induced liver damage in Wistar albino rats.

A NOVEL NANO-FORMULATION FOR ANTI-CANCER ORAL DELIVERY WITH BOVINE IRON SATURATED LACTOFERRIN FOR COLON CANCER

Rupinder K Kanwar, Ganesh Mahidhara and Jagat Kanwar

Laboratory of Immunology and Molecular Biology (LIMBR), Institute for Technology Research and Innovation (ITRI), Deakin University, Geelong, Victoria, Australia.

Key Words: anti cancer bovine lactoferrin, cancer, and neurodegenerative disorders

At nano scale, the fundamental and vital properties of matter can be changed, which can be used for daunting task such as oral administration of bio-macromolecules to be able to achieve sustained delivery, controlled release, target specific delivery and combinatorial therapy. In our study, we demonstrate the formulation of a novel alginate enclosed, chitosan coated ceramic anti cancer nano particles (ACSC NP). These NP were loaded with multi-functional anti cancer bovine lactoferrin (bLf), a natural milk based protein, for improvement of intestinal absorption, in order to develop a novel platform to carry anti cancer protein and/or peptides for oral therapy. Size, morphology, internalization and release profiles of the ACSC NP under varying pH were determined. Furthermore, uptake of these NP *in vitro* in colon cancer cell lines was analyzed, by measuring the endocytosis and transcytosis.

The results show the NP were having size range of 200nm and increased up to 400nm with addition of alginate layer and obtain a spherical morphology. SDS PAGE followed by western blotting, using specific antibodies against bLf confirms the structural integrity of the protein after the nano formulation; confocal microscopy and flow cytometry qualitatively and quantitatively determines the internalization of rhodamine labeled NP, upon treating them on to Caco-2 cell lines. Transcytosis studies indicate the transcytosis of the NP, with minimal damage to the Caco-2 cell monolayer. In conclusion, these NP can be used for future targeted protein/peptide or nucleic acid based drug delivery to treat fiddly diseases such as cancer and neurodegenerative disorders.

ENGINEERING FLIES FOR LONGEVITY

William Orr

Department of Biological Sciences, Southern Methodist University, Dallas, TX, (USA)

Key Words: antioxidant system, glutathione, oxidative damage, redox balance, longevity

The antioxidative system is highly complex with various overlapping and interconnected mechanisms for optimizing the cellular oxidant load. Critical components of the antioxidant system that may affect the aging process remain to be identified.

Glutathione (GSH) is a versatile antioxidant, which can react directly with radicals or donate electrons in enzyme-mediated elimination of reactive intermediates, whereas NAD(P)H provides the reducing equivalents for the regeneration of GSH from its oxidized form, GSSG, as well as for the elimination of hydrogen and organic peroxides. Thus, GSH and NAD(P)H act in concert to maintain the redox balance of cells. Together, they also provide the bulk of the reducing power in cells. Since the level of oxidative stress tends to increase with age, it is hypothesized that enhancement of the efficiency of antioxidant defenses, provided by the combined functions of GSH and NAD(P)H, would have a stabilizing effect on cellular redox state during the aging process.

GSH synthesis is determined by the activity of the rate-limiting enzyme -glutamate-cysteine ligase (GCL), which is composed of two subunits, the catalytic, GCLc, and the modulatory, GClm. NAD(P)H is mainly supplied by the pentose phosphate shunt, whose rate-limiting enzyme is glucose-6-phosphate dehydrogenase (G6PD). In my talk I will describe our efforts to manipulate these specific components of the antioxidative system in Drosophila transgenics in order to impact the aging process. The results of our work form the basis for a modified oxidative stress hypothesis of aging that takes into account not only the age related accumulation in oxidative damage but also the age-associated shifts in redox state that cause deterioration in signaling mechanisms.

ALLEVIATION OF NON-ALCOHOLIC FATTY LIVER BY BETAINE SUPPLEMENTATION IN RATS

Young C. Kim, Young S. Jung, Do Y. Kwon, and Sun J. Kim

College of Pharmacy, Seoul National University, Kwanak-Ku, Seoul, Korea

Key Words: non-alcoholic fatty liver (NAFL), non-alcoholic steatohepatitis (NASH), oxidative stress, Total Oxyradical Scavenging Capacity (TOSC), transsulfuration reactions, S-adenosylmethionine (SAM), glutathione (GSH).

Background: Non-alcoholic fatty liver (NAFL) is involved in the development of non-alcoholic steatohepatitis (NASH) and chronic liver injury. Previous studies revealed that induction of both acute and chronic liver injury is associated with dysregulation of hepatic transsulfuration reactions. The impaired metabolomics of sulfur-containing substances in alcoholic liver injury has been the subject of numerous studies, however, its role in non-alcoholic fatty liver disease (NAFLD) remains unknown.

Objective: The objective of this study was to evaluate the changes in hepatic transsulfuration reactions and their significance in the early stages of NAFLD. The effect of betaine on the induction of fatty liver and alterations in the metabolomics of sulfur-containing substances was also investigated.

Methods: Adult male rats were provided with a standard liquid diet or a high-fat (HF) liquid diet for 3 week. An additional group of rats was fed the HF liquid diet supplemented with betaine (1 %). Development of fatty liver and alterations in the metabolomics of sulfur-containing substances were determined after sacrifice. Antioxidant capacity of liver against hydroxyl, peroxyl radicals and peroxynitrite was measured using the Total Oxyradical Scavenging Capacity (TOSC) assay.

Results: The HF diet intake did not affect the food consumption, body or liver weight changes in the rats, but elevated hepatic triglyceride levels and serum tumor necrosis factor-alpha (TNFα) concentrations. The TOSC values of liver cytosol against hydroxyl and peroxyl radicals, but not peroxynitrite, were reduced significantly. Hepatic S-adenosylmethionine (SAM) and glutathione (GSH) were decreased, but hypotaurine and taurine concentrations increased. Methionine adenosyltransferase (MAT) enzyme content was not changed, but its activity was depressed while both activities and contents of cysteine dioxygenase (CDO) and GSH S-transferase (GST) were elevated in the rats fed the HF diet. Cystathionine gamma-lyase, but not cystathionine beta-synthase, was also increased by the HF diet intake. Betaine supplementation inhibited hepatic fat accumulation and serum TNFα elevation in the rats fed the HF diet. The decrease in cytosolic TOSC against hydroxyl and peroxyl radicals was also prevented. Both MAT activity and its content were induced

significantly by betaine supplementation. Hepatic SAM and GSH concentrations were increased, and elevation of hypotaurine and taurine was depressed.

Conclusion: The results indicate that hepatic transsulfuration reactions are disturbed significantly in the rats fed the HF diet, suggesting a causal role of the impaired metabolomics of sulfur-containing substances in the development of NAFLD. Imbalance between prooxidants and antioxidant capacity appears to be responsible for the disturbance of hepatic transsulfuration reactions, which further aggravates oxidative stress associated with the HF diet intake. Betaine supplementation in the HF diet protects the liver from fat accumulation and oxidative stress most probably via its effects on the transsulfuration reactions.

THE EFFECTS OF TRADITIONAL CHINESE MEDICINE HERBAL EXTRACT, CHI02, ON BIOCHEMICAL MARKERS OF BONE LOSS AND ARTICULAR CARTILAGE DEGRADATION IN OVARIECTOMIZED RATS

Qinlong Zheng[1*], Morten Karsdal[2], Per Qvist[2], Yujia Ding[1], Kim Henriksen[2]

[1]Nordic Bioscience (Beijing), Beijing, P. R. China,
[2]Nordic Bioscience, Herlev, Denmark

Key Words: Traditional Chinese medicine (TCM), osteoporosis, osteoarthritis, neutraceutical.

Background: The edible traditional Chinese medicine (TCM) herbs have been used as food or medicine for many years with the aim of preventing or improving general health, but also diseases such as osteoporosis and osteoarthritis. Recent data have indicated that some herbal extracts may possess chondro or bone protective effects in animal models. This raises the possibility that herbal extracts from traditional Chinese medicine may be used as a neutraceutical for improving cartilage and/or bone health.

By using highly specific biochemical markers of cartilage and bone degradation, we screened in excess of 50 herbal ingredients based on traditional Chinese medicine in the ovariectomized rat. This is a well-known animal model for postmenopausal bone loss, and in addition it has been

demonstrated that depletion of endogenous estrogen induces an accelerated loss of articular cartilage in this model.

Objective: To prepare a library of herbal extracts based on traditional Chinese medicine and to screen the extracts in the ovariectomized rat model for beneficial effects on bone and/or cartilage.

Methods: The raw materials were obtained from six different suppliers in China and subsequently formulated by a series of extraction and purification procedures. Subsequently, each dose of the TCM extracts was given by oral gavage, to 10 ovariectomized (OVX) 6-month old Sprague-Dawley rats for 6 consecutive weeks. For comparison, groups of rats were sham operated, or ovariectomised and given vehicle or oestrogen. Serum samples were collected from each rat for biochemical marker analysis at baseline, 3 week and 6 week after operation. Bone resorption and cartilage degradation were assessed by measurement of the serum samples in the CTX-I ELISA and CTX-II ELISA (IDS Nordic, Denmark).

Results: Ovariectomy (OVX) induced significant body weight gain in the OVX animals, reaching 18% in the OVX vehicle control groups. And the OVX rats treated with TCM-based herbal extract designated CHI02 at the dose of 750mg/Kg showed a significant reduction of body weight gain compared to the vehicle

control group ($p<0.05$). Bone resorption and cartilage degradation were quantified in all rats by measurement in serum CTX-I and CTX-II, respectively. Ovariectomy resulted in a marked increase in serum levels of CTX-1 and CTX-II by approximately 100% after three weeks in OVX rats compared with the sham-operated group, which is in accordance with the expected effect of OVX induced estrogen deficiency on bone turnover and cartilage degradation. The herbal extract CHI02 at a daily dose of 750mg/Kg for three weeks has significant effects on the reduction of CTX-I and CTX-II levels by 46.6% and 60.1% respectively in OVX rats, as compared with the vehicle control group ($p<0.05$).

Conclusion: The results of this screening study clearly demonstrated that Chinese herbal extract CHI02, has strong, short-term beneficial effects on both skeletal and cartilage compartments. Therefore this ingredient has potential for use as a neutraceutical for improving cartilage and/or bone health. However, further studies are needed to determine its utility in humans.

ORAL NANODELIVERY OF SMALL INTERFERING RNA AND MICRO RNA GENE THERAPY IN FOR BREAST AND COLON CANCERS

Jagat R. Kanwar[1], Rupinder K. Kanwar[1] and Chun Hei Antonio Cheung[2]

[1]Laboratory of Immunology and Molecular Biomedical Research (LIMBR), Center for Biotechnology and Interdisciplinary Biosciences (BioDeakin), Institute for Technology and Research Innovation (ITRI), Geelong Technology Precinct (GTP), Deakin University, Pigdons Road, Waurn Ponds, Geelong, Victoria, Australia.
[2]National Institute of Cancer Research, National Health Research Institutes (NHRI), Tainan, Taiwan R.O.C.

Key Words: cancer gene therapy, breast cancer, colon cancer, and RNA

The present presentation focuses more on the cancer gene therapy to silence the survivin gene with nanotechnological delivery systems. Various techniques have been developed to prepare nanoparticles for the delivery of drugs. More recently we were able to load cell permeable dominant negative survivin R9 (DNSurR9) and survivin and HSP-90 antagonists, ―shepherdin‖ on alginate gel-encapsulated, chitosan ceramic nanocores nanocarriers (ACNC-NPs) and able to induce apoptosis and disintegrate the mitochondria of colon and breast cancer cell lines (but not normal control cells) more efficiently in in vitro cell based assays. MicroRNAs (miRNAs) play important regulatory roles in targeting mRNAs for cleavage or translational repression. miRNAs may function as oncogenes or tumor suppressors. In the present study we loaded siRNA to survivin and

oncogenic antisense microRNA-27a (as-miR-27a) on ACNC-NPs and transferred to human breast cancer MDA-MB-231 and MCF-7 cell lines by endocytosis. Our results show that as-miR-27a loaded ACNC-NPs exhibits oncogenic activity. Suppression of miR-27a inhibits breast cancer cell growth and invasion. Simultaneous siRNA to survivin and oncogenic as-miR-27a loaded ACNC-NPs results in down expression of genes that are important for cell survival and angiogenesis faster and more efficiently than the monotherapy. In addition, these responses were accompanied by decreased expression of survivin, and angiogenic genes, including survivin, vascular endothelial growth factor (VEGF), and VEGF receptor 1(VEGFR1). We also demonstrated the downregulation of survivin expression in Western blot in the siRNA to survivin and oncogenic as-miR-27a loaded ACNC-NPs treated cells. TUNEL assay, caspase activity assay and changes in mitochondrial membrane potential reveal that cell death was mainly through intrinsic apoptosis pathway. Oral delivery of siRNA to survivin and oncogenic as-miR-27a loaded ACNC-NPs induced apoptosis, necrosis and cytotoxicity in the xenograft breast cancer model. Oral administration of siRNA to survivin and oncogenic as-miR-27a ACNC-NPs in combination regress tumour growth faster and inhibits angiogenesis in the xenograft breast cancer mouse model as compared to monotherapy. We also compared our results with the doxorubicin and taxol loaded ACNC-NPs. Taken together, our results are highly encouraging for the development of combination nano-therapeutic strategies that combine gene silencing and drug delivery to provide more potent and targeted therapeutic, especially in late and metastatic breast cancer.

PART FOUR

FUNCTIONAL FOODS AND METABOLIC SYNDROME: THEORETICAL APPROACHES

DIABETES AND CARDIOVASCULAR DISEASE: WHAT DO WE REALLY KNOW?

Anand Prasad

Interventional Cardiology, University of California, San Diego, USA

Key Words: Diabetes, myocardial infarction, and glycemic control

Diabetes is well established risk factor for early onset and aggressive atherosclerotic vascular disease. Macrovascular disease leading to myocardial infarction and stroke represent the greatest contributors to the mortality of diabetics. Despite the strong causal relationship between diabetes and atherosclerosis, strategies to specifically reduce cardiovascular events by strict glycemic control have yielded mixed results. In this presentation we will review the pathophysiology underlying atherosclerosis in diabetic patients with a specific focus on atheroma morphology and vascular distribution. Furthermore, the disconnect between micro and macro vascular complication reduction with glycemic control will be explored. The impetus to employ tight glycemic control based on data from the Diabetes Control and Complication Trial and the UKPDS studies will be reviewed. These data will be placed into perspective by the more recent results from the ACCORD, ADVANCE, and ADDITION studies which when taken together raise questions about the benefit of intensive glucose control. Additional strategies for primary and secondary risk reduction in diabetics will be discussed.

KEY ROLE OF DEFORMATION HARDENING AND FATIGUE SOFTENING OF BIOLOGICAL TISSUES IN EVOLUTION, AGING, CANCER AND DISEASES

Valery P. Kisel

Institute of Solid State Physics, Russian Academy of Sciences, Chernogolovka, Russia

Key Words: physiological stress, mitochondria,

It was shown that all types of phase transformations (PT) are determined by the deformation mechanisms at phase interfaces first of all (including biological tissues, BT)1-2. There are a lot of experimental evidences that the liquid (gas, plasma) shearing is determined by the so-called dynamic viscosity, η, which is closely related to the standard dislocation work hardening (DWH) and softening (DWS) in solids[1]. Two main deformation processes determine BT fatigue (aging): the chain of PT from DNA up to cells under body metabolism (including nutrition) and physiological stress, and the confining effects of micro-nano-environments[1]. The metabolism under electron fatigue deformation of BT in the electron-transport-chains of mitochondria is determined by the reverse DWS flow of electric currents. The softening due to the Bauschinger effect determines deformation durability of crystals and the longevity of BT[1]. The data of work[3] strictly confirms this by the facts that the life span of 12 species (birds and mammals) the less the more the rate of H2O2-defects nucleation at the reverse electron transfer in mitochondria of

cardiomyocytes. This means that it is the lower reverse deformation stress that makes the higher longevity of BT, and it is supported by the absence of high life span at the direct electron transfer (the direct stage of the Bauschinger effect)[3]. This is in line with the mechanism of charged particle superconductivity in solids, superfluidity in liquids, superdiffusion in severe deformed (up to nanostructures) crystals, enhanced dissolving or catalysis, etc., which is always concerned with abrupt rise in deformation hardening of materials[2]. Since the mechanisms of deformation in solids, liquids, biological tissues (BT), gases, plasma are the same[1], we have a well-founded expectation to see the same superfluid-hardening PT to malignant cells due to their abrupt hardening and high rate of proliferation[1]. This makes the cancer cell Young's modulus softer than in the neighbored healthy cells[4] and immortal like the long-living currents in superconductivity or continuous slipping as superfluid films of liquid HeII on the hard walls. This means that the drag of cell elements plays the key role in their PT. The data of work on fruit flies (Drosophila)[5] confirm this: the aging of embryonic stem cells (ESC) had made their asymmetric fission, and tissue homeostasis had been weakened due to wrong orientation of stem cells and thus delaying the cell cycle. This means that the cell aging - DWH of their intercellular liquid - localizes its deformation mainly in the fixed directions like in solids under DWH[1-2], thus preventing ESC from the right orientation. Recent data confirms the possibility of human ESC subpopulations of the same origin to develop into the malignant

ones with small changes in chromosome structure[6] like the wide spectrum of DWH of dislocations from the same source in solids[1]. This is in line with partial or complete spontaneous reversible deformation as in weak DWH-solids[7] and spontaneous involution of cancerous tumors (in the low DWH small or young BT) under physical and chemical stress[1].

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ANIMAL MODELS OF METABOLIC SYNDROME: RELEVANCE TO HUMAN CONDITION

Parakat Vijayagopal and Chandan Prasad

Department of Nutrition and Food Sciences,
Texas Woman's University, Denton, TX

Key Words: animal model, metabolic syndrome, and high calorie diet

The metabolic syndrome (MS) as the name suggests is not a single disease state but a cluster of metabolic abnormalities that often occur together which can lead to cardiovascular disease. There has been a fair degree of controversy with respect to contributions of different metabolic abnormalities in defining MS. Consequently, so far there have been four different definitions proposed by agencies such as WHO, EGIR, NCEP ATP III and IDF. Irrespective of who defines MS, the common metabolic abnormalities include, hypertension, central obesity, insulin resistance and dyslipidemia. Some of the other observed abnormalities in human that could be secondary to MS include, microalbuminuria, hyperuricemia and gout, impaired fibrinolysis, elevated coagulability, elevated CRP, endothelial dysfunction, low cardiorespiratory fitness, fatty liver disease, polycystic ovary syndrome, and decreased adiponectin.

Like many defined disease states such as Parkinson's disease, lupus and type I diabetes, availability of animal models have greatly enhanced our ability to acquire a better understanding

of the disease process. However, considering the nature of MS as outlined above, creating an animal model of the condition will be a Herculean task, if not impossible. Over the years, a number of large (primates, swine and dogs) and small (rabbits, rats, mice, hamsters and sand rat) animals species have been used to create model for MS largely by forcing these animals to consume high calorie diet. The purpose of this review is to critically analyze relevance some select animal models to human condition.

METABOLIC SINDROME AND IMMUNE CHRONIC DISEASES

Valery P. Kisel

Institute of Solid State Physics, RAS, Chernogolovka, Moscow District, Russia

Key Words: metabolic syndrome, chronic disease, and inflammation

Cell metabolism concerns the intracellular chemical reactions and physical effects (stress, light radiation and electromagnetic fields, etc) that convert nutrients, endogenous molecules and external fields into the energy and matter (nucleic acids, proteins and lipids) that drive life processes in biological tissues (BT). Adenosine triphosphate (ATP) is the key molecule that sustains all energy-dependent processes in cells and mainly generated by two metabolic pathways – glycolysis and oxidative phosphorylation.

Our previous works have shown that the production of reactive oxygen species (ROS) was the result of various phase transitions in BT: chemical reactions under metabolism, physical stress and mechanical deformation of BT.

A local inflammation of BT is a result of the impact of excessive ROS under severe hardening of tissues, and their long-term influence is in the grounds of various chronic diseases (cancer, cardio-vascular disease, arthritis, obesity, diabetes, aging, etc.).

Recent work[1] has shown the physical similarity between stiffening of BT and their diseases, and the ultimate mechanical work-hardening of solids, liquids, BT sustains the dragless superplasticity, superdiffusion, extradeep penetration, superconductivity and the so-called superfluidity in solids and liquids, superconductivity, cancer proliferation and metastasis in BT. There are a lot of only physical methods to destroy the above effects.

First, I intend to discuss in detail the new mechanisms of diseases and regeneration, the new physical methods to soften and regenerate the stiffened-aged BT.

Second, in the frames of the new science – epigenetics, the molecular processes that control a potential of new genes to act. Evidence now suggest that epigenetics can lead to inherited forms of obesity and cancer[2]. I suppose to discuss the reverse (recoverable) epigenetic effect – BT softening after diseases – under intermittent low physical or physiological stress, special nutrition and Russian peoples medicine.

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AYURVEDIC HERBS AND DIETARY SUPPLEMENTS IN THE MANAGEMENT OF METABOLIC SYNDROME

Ala Narayana

National Institute of Indian Medical Heritage
Osmania Medical College Building Koti, Hyderabad, India

Key Words: Metabolic syndrome**,** overweight, sedentary life styles

Background: Metabolic Syndrome (MS) is a combination of medical disorders that increases the risk of developing cardiovascular disease and diabetes. Ayurveda, the Indian system of medicine, described metabolic syndrome conditions like *Medoroga* (Obesity), *Prameha* (Diabetes), *Raktagatavata* (Hypertension) as separate disease entities. Ayurveda vividly focuses the main etiological factors of MS are Stress, overweight, sedentary life styles and lay down the treatment for diabetes, controlling the hypertension, maintenance of ideal body weight correction of dyslipidaemia & prevention of early atherosclerotic changes by using dietary supplements, single drugs, poly herbal formulations, minerals/metalloid-mineral formulations, specialized bio-purificatory measures and life style.

Objective: To validate Herbs and dietary supplements, which were described in Ayurveda in the management of Metabolic Syndrome are based on the knowledge of contemporary pharmacological

activities, therapeutic uses and clinical & experimental research outcome.

Methods: Traditional Indian diets are functional because they contain rich amounts of Dietary fiber (Whole grains and Vegetables), Antioxidants (Fruits, Vegetables and Spices), Probiotics (Curds and Fermented batter products). The following Ayurvedic drugs and dietary supplements for the management of MS described in ancient Ayurvedic classics will be studied in the light of contemporary medical science for validation.

Dietary supplements:

- Bengal gram (Cicer arietinum Linn) - Hypocholesterolemic, antiatherogenic, anti-stress, antihyperlipidemic, antihypertensive, cardiovascular.
- Lentil (Lens culinaris) – Hypocholesterolemic.
- Kokam butter (Garsinia indica Choisy) - weight reduction property due to 3- hydroxy citric acid.
- Java plum (Syzygium cumini (L) Skeels) - Hypoglycemic.
- Figs (Ficus glomerata Roxb) - Cardiac depressant, hypoglycemic, hypotensive.
- Bitter guard (Momordica charantia Linn) - Hypoglycemic, antidiabetic, mild hypotensive, hypolipidemic.

- Fenugreek (Foeniculum vulgare Mill) - Hypocholesterolemic, hypolipidemic, cardiovascular, hypoglycemic.
- Horse gram (Dolichos biflorus Linn) - Hypocholesterolemic, hypolipidemic, antistress, hypoglycemic, hypotensive.
- Indian gooseberry (Emblica officinalis Gaertn) - antioxidant, hypoglycemic, hypolipidemic.
- Garlic (Allium sativum Linn) - Hypocholesterolemic, hypolipidemic, antioxidant, antiageing, cardioprotective, hypoglycemic, hypotensive, fibrinolytic.
- Onion (Allium cepa Linn) - antioxidant, antidiabetic, hypoglycemic, hypocholesterolemic, antiatherogenic, antihyperlipidemic, antihypertensive, antiarterosclerotic.

Herbs:

- *Vacha* (Acorus calamus Linn) - Hypotensive, moderate depression of BP, tranquilizing.
- *Karanja* (Pongamia pinnata pierre) - Hypoglycemic, hypotensive, cardiac depressant, anti- cholinergic.
- *Aswagandha* (Withania somnifera Dunal) - Antioxidant, hypotensive, antistress, cardioprotective, anticoagulant.
- *Guduchi* (Tinospora cordifolia (willd) Miers) - Hypoglycemic, antihyperglycemic, hypotensive, antidiabetic, antistress, antioxidant.

- *Vata* (Ficus bengalensis Linn) - Hypoglycemic, hypotensive, antidiabetic, hypocholesteremic, antioxidant.
- *Ardraka* (Zingiber officinale Roscoe) - Hypolipidemic, antiatherosclerotic, cardiovascular, antioxidant, antistress, hypoglycemic.
- *Arjuna* (Terminalia arjuna W & A) - Cardioprotective, antianginal.
- Upakunchika (Nigclla sativa Linn) - Hypoglycemic, hypotensive, cardiovascular, antidiabetic, cardiac depressant.
- Meshasringi (Gymnema sylvestre R. Br) - Antidiabetic, hypoglycemic, cardiovascular, antiarterosclerotic.

Conclusion: The study endow with the wide range of Ayurvedic Herbs and dietary supplements, which proved greater effect on ailments like improved gastrointestinal health, enhanced immune function, weight management, reduced blood cholesterol, better management of diabetes, reduced risk of CVD, inflammatory diseases.

SIDDHA HERBS FOR DIABETES

J. Raamachandran

Murugan Siddha Marunthakam, "Murugan Illam",
Valasarawakkam, Chennai, India

Key Words: Siddha, Cassia auriculata, aavaarai, Murraya Koenigii, Kariveppilai, diabetes, insulin, retinopathy, skinproblem

Background: The siddha medicinal system developed by Tamil Siddhars is the oldest system in India. It has developed medicines in the most scientific way. It has medicines for several chronic diseases. In to-day's world the so called disease diabetes occupies a significant place since many people are affected by it. It is to be mentioned that Siddha sysytem does not treat diabetes as a disease but only a symptom and hence be cured if properly noticed at the earliest.

Objective: The Siddha system has its own method of finding and curing diabetes. It is quite common that people with diabetes may experience one of these symptoms, viz. excessive urination, especially at night, excessive thirst, itchy and dry skin, polyphagia, weight loss, blurred vision etc. But, how to identify whether these symptoms are really due to diabetes? There were no clinical labs in those days, the type we have to-day. So how to determine and then how to cure it? This paper will describe the ancient and still practiced way of Tamil siddhars.

Methods: It is to be mentioned that Tamil siddhars were the first to give to the world the idiom ―Food is medicine‖. For many diseases, a simple modification of food and food habits will do good. As far as diabetes is concerned, several herbs are quite useful which can be included in the daily food. One such herb is Cassia Auriculata(―aavaarai‖ in Tamil) which can either be used as medicine or as food. The flower of this herb is quite useful in controlling blood sugar. The plant as a whole is quite effective in controlling diabetes. We have given this to many diabetic patients and checked their blood for sugar in the modern way after satisfying ourselves with our own way. This herb is particularly useful for people with diabetes and skin problem. For people (especially diabetic type 2) with diabetes and eye problems (retinopathy) another herb Murraya Koenigii (―kariveppilai‖ in Tamil) is the useful. This herb has the effect of insulin in the body. These herbs can be added in the food daily without fear of side effect.

Results: The study on human beings by giving these herbs in forms depending upon the person showed very promising effects.

Conclusion: The modern scientific community must make note of these herbs and they can be added in the food items prepared in any modern restaurants.

PART FIVE

DEVELOPMENT OF FUNCTIONAL FOOD PRODUCTS AS THERAPEUTICS

PREBIOTIC SOLUBLE FIBER IN DAIRY DESSERTS FOR DIGESTIVE HEALTH

Lorraine Niba

Starch Food Innovation, Bridgewater, NJ, USA

Key Words: Soluble fiber, Digestive health, Prebiotics, Dairy deserts

Background: Soluble fiber plays a critical role in digestive health. Research has firmly established the importance of fiber in laxation and regularity (1, 2, 3, 4, 5). Fiber improves intestinal transit, laxation and regulatory through its bulking action in the small intestine (3, 6). Viscous soluble fibers with high water holding capacity contribute to bulking and laxation (7, 8).

In addition to the physical effects of bulking, certain fibers act as fermentation substrates for various beneficial bacterial species in the colon, demonstrating prebiotic activity. Prebiotics are defined as selectively fermented ingredients which allow specific changes, both in the composition and/or activity in the gastrointestinal microbiota, which confers benefits upon host well-being and health (9).

Colonic fermentation of prebiotic soluble fibers by beneficial colonic microflora impacts digestive health in various ways, including: increase in protective bacterial populations, and inhibition of proliferation and adhesion of pathogens (10, 11) and production of short chain fatty acids which are nutrients for colonic mucosal epithelial cells, protecting cell wall integrity (12,13), and decrease in colonic and fecal pH. Consumption of fiber among American

consumers falls far below the recommended daily amount of 25 - 38 g. This is reflected in the high incidence of digestive diseases: 22 % of American consumers reported being affected by gastrointestinal diseases in 2007, while 37 % of consumers reported being concerned about gastrointestinal disease (14). Dairy deserts could be a convenient delivery system for prebiotic soluble fibers.

Methods: The objective of this study was to develop ice cream containing at least 3 g of prebiotic soluble fiber per serving. This would provide 10 % of the RDI for fiber.

Prebiotic soluble fiber was used in a standard ice cream formula; controls were formulated with corn syrup solids. The ice cream was evaluated by a trained sensory panel on a 15-point difference scale for appearance, taste and texture. A melt down test (4 hours,19 ^{o}C) was used to assess melting characteristics.

Results: Ice cream containing prebiotic soluble fiber did not show any significant difference from the control with respect to sweetness, taste, firmness. Panelists observed greater smoothness, and less iciness in ice cream containing prebiotic soluble fiber, than in the control. Prebiotic soluble fiber slowed the melting of ice cream. The observed improvement in functional properties of ice cream is likely due to the replacement of corn syrup solids with prebiotic soluble fiber.

Conclusions: Prebiotic soluble fiber can be successfully incorporated into a popular dairy desert, ice cream with no adverse impact on

sensory or eating quality of the product. Ice cream containing 4 g of fiber per serving was formulated, with no significant difference from a control. As food manufacturers increasingly seek options for functional foods with physiological benefits, prebiotic soluble fibers offer viable potential for use in foods targeted at enhanced digestive health.

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ANTHOCYANIN-RICH PLUM AS A BEVERAGE BASE WITH POTENTIAL HEALTH FUNCTIONALITY

Fanning K., Netzel M., Netzel G., Russell D., and Stanley R.

Department of Employment Economic Development and Innovation, Queensland, Australia

Key Words: Anthocyanin, oxidative stress, bioavailability, plum

Background: There is an increasing accumulation of literature supporting an in vivo ability of anthocyanins to ameliorate symptoms of metabolic syndrome. In humans supplementation with anthocyanin has improved lipid profile, with increased HDL cholesterol, lowered LDL cholesterol and increased cellular efflux of cholesterol to serum, and lowered plasma inflammatory markers. Animal studies from the past decade have supported a role for anthocyanins in a variety of diet-induced models. Anthocyanin-rich purple corn extracts have shown beneficial effects in preventing obesity and normalising blood glucose, leptin and insulin levels in mice fed a high fat diet. An anthocyanin extract from blueberries and strawberries, included in a high fat diet, reduced weight gain in mice. An anthocyanin-rich purple carrot juice in a high-carbohydrate, high-fat, diet-fed rats attenuated or reversed all symptoms of the diet-induced state - hypertension, cardiac fibrosis, increased cardiac stiffness, endothelial dysfunction, impaired glucose tolerance, increased abdominal fat deposition, altered plasma lipid profile, liver fibrosis, increased plasma liver enzymes, increased plasma markers of oxidative stress

and inflammation, and increased inflammatory cell infiltration. A new variety of Japanese plum (Prunus Salicina), Queen Garnet plum (QGP), was developed in a Queensland government breeding program. It was identified as an anthocyanin rich fruit which could provide consumers with a significant anthocyanin intake through consumption of fresh fruit in season, or processed into a beverage base for use in nutraceutical products.

Objective: To investigate the relevant amount of anthocyanin in QGP fruit and juice, and the urinary excretion kinetics of the anthocyanin.

Methods: In the 2003/4 growing season 8 commercial varieties of Japanese plum were compared with the QGP for anthocyanin content. In 2008 the effect of maturity of anthocyanin content in QGP was examined over a five week period. A pilot study was undertaken to examine the bioavailability of anthocyanins, in human urine, from QGP juice (QGPJ). Juice was prepared from QGP by pre-heating, enzyme treatment, juicing and finally pasteurisation. Two male volunteers drank 400ml of QGPJ and urine was collected pre-dose and over 24-h post dose.

Results: QGP had significantly greater anthocyanin content compared to other commercial varieties. QGP anthocyanin content was seen to increase by 55% over a five week harvest period to a maximum of 272 mg cya-3-glucoside equivalents/100g fresh weight. QGPJ was a very good source of anthocyanins (1.117 g/400ml) and total phenolics (2.66 g gallic acid equivalents/400ml). Following

QGPJ, the urinary excretion of total phenolic content was increased, malondialdehyde was decreased and hippuric acid was increased (3-fold).

Both native QGP anthocyanins and four identified metabolites were also present in the urine following QGPJ ingestion. The sum of urinary excreted anthocyanin and metabolites totalled 0.5% of ingested dose.

Conclusion: The anthocyanin content of QGP (170-280) in 2008, over a five week period, was comparable to literature values for the richest berry sources of anthocyanin (bilberry 215-300, blackcurrant 128-411, blueberry 63-331). Additionally QGP yields at up to 40 tonne per hectare which is 7-8 times the yield of blueberries/blackcurrants, thus making it a very attractive source of anthocyanin. As urinary malondialdehyde is regarded as a non-invasive biomarker for oxidative stress, it's reduction following QGPJ ingestion suggests that anthocyanins and/or other polyphenolics from QGPJ, either in intact form or as metabolites, may have a role in ameliorating oxidative stress. Further testing is required to better understand the potential in vivo benefits of consuming anthocyanins from QGP fruit, juice or powder.

STUDY THE EFFECT OF PROBIOTICS BACTERIA ISOLATED FROM EGYPTIAN INFANTS IN THE CHOLESTEROL REMOVAL FROM MEDIA AND THEIR PRESENCE IN YOGHURT

Hoda Mahrous

Genetic Engineering and Biotechnology Research Institute, Menofiya University, Egypt

Key Words: Lactobacillus, probiotics, and cholesterol removal

Probiotics are defined as ―living organisms which upon ingestion in certain numbers exert health benefits beyond inherent basic nutrition.‖ One of the most significant groups of probiotic organisms are the lactic acid bacteria, commonly used in fermented dairy products. In this study cultures were isolated from two infants After screening for the classic properties of probiotic organisms, four promising isolates were identified as two strains of Lactobacillus acidophilus(P106, P110), strain of Lactobacillus plantarum (P164) and Lactobacillus. Pentosus (P191) were tested for capability to remove cholesterol and to deconjugate sodium taurocholate from the culture medium. Results showed that a considerable variation existed among cultures in their growth viability in the presence of bile salt, deconjugation of sodium taurocholate and assimilation of cholesterol from the medium. All tested strains removed less cholesterol from the broth (ranged from 4.02-24.32%) compared to those grown in broth supplemented

with 0.2% bile salts (from 29.02 to 45.3). Lactobacillus acidophilus P106 appeared to be more active in bile salt hydrolase compared to the other strains, and therefore, is regarded as a suitable candidate probiotic and adjunct culture.

These strains were employed to make yoghurt and, in order to achieve a short production time; a two-stage fermentation procedure was used with Streptococcus thermophilus and Lactobacillus delbrueckii subsp. bulgaricus providing the rapid acidification. Storage trials at 4o C showed that the viability of the probiotic cultures was retained over 15 days.

MEDICINAL PLANTS BASED FUNCTIONAL (HERBAL) FOOD PRODUCTS

Goyal S. K.[1] and Samsher S.V.[2]

[1]Institute of Agricultural Sciences, Barkachha, BHU, Mirzapur, India
[2]Department of Agricultural Engineering and Food Technology, Patel University of Agriculture and Technology, Meerut, India

Key Words: Stevia, Safed musli, Herbal beverage, stevioside

Background: Preparation of herbal foods is a new concept in food industry. Herbal food products are those products which are prepared with the herbs. Presently, herbal products either in the form of foods or cosmetics have become more popular in the international market. American dietetic association noted that consumption of herbal plants and other foods containing anti-oxidants can provide protection against certain diseases (Pazczola, 2001). Epidemiological data as well as in vitro studies strongly suggested that foods containing phytochemicals with intoxication potential have strong protective effects against major disease risks including cancer and cardiovascular diseases (Kaur and Kapoor, 2002). Herbs play a significant role especially in modern time. The movement is based on the belief that the medicinal plants have a vast potential for their curative medicinal uses as herbal food products. Safed musli (Chlorophytum borivilianum) is characterized by the presence of saponin. In the Indian traditional system of medicine (Ayurveda), Safed musli (Chlorophytum borivilianum) roots are used in many theraptic preparations. Roots are used in commercial preparation of

steroidal hormones. Generally it is used for increasing vitality. Stevia (Stevia rebaudiana) used as a sweetener for tea, coffee and food additives in Japan. It is also used as herbal medicine for hypoglycemia, hypotensive, diuretic, cardio tonic and other tonics, etc. It has zero calories sweetening agent and can be used by diabetic patient (Kumar et al, 2006). Stevioside is one of the sweet substances isolated from the leaves. Chemically, stevioside is the diterpene glycoside with the aglycone part called steviol. Stevia has similar sweetening taste as sugar (sucrose) but it has 300 times sweetness as compared to sucrose (Kohda, et al., 1976; Pederson, 1987). At present, herbal food products are gaining popularity among different section of the society. Attempts were therefore made to develop herbal beverage and herbal burfi from safed musli roots and stevia leaves.

Objective: To develop the herbal foods by the use of medicinal plants.

Methods: Various quantity of orange juice (80, 85 and 90%, v/v) and stevia extract (10, 13, 16 and 19%, v/v) were selected on the basis of trial runs with 2% safed musli extract in each sample for the development of herbal beverage. Similarly, for preparation of herbal burfi, different levels of khoa (95, 90 and 85%, w/w) and stevia powder (5, 10 and 15%, w/w) with 2% safed musli powder and other minor ingredients were used. The herbal burfi samples were compared with market burfi. The physico-chemical, microbial and sensory quality attributes of these products were evaluated during storage.

Results: The combination of 90% orange juice and 10% stevia extract with 2% of safed musli extract secured highest overall acceptability for herbal beverage. While, less numbers of microbes (1.843x105 cfu/ml) were found, those prepared with highest level of stevia extract (19%) and lowest orange juice (80%) when samples stored upto 180 days at ambient condition. Other physico-chemical qualities of this ratio were found satisfactory. Herbal burfi samples prepared with 90% khoa, 10% stevia powder and 2% safed musli powder ratio was found best. This ratio scored highest overall acceptability. Less microbial growth (2.557x105 cfu/g) was noticed those sample having 85% khoa and 15% stevia powder. Other physico-chemical qualities of this ratio were also found satisfactory. The values of TSS, optical density, acidity and TPC were increased significantly however, the values of pH, total sugars and sensory scores decreased significantly during storage of herbal beverage. The composition of 90% khoa, 10% stevia powder and 2% safed musli powder ratio was found better in comparison to market burfi when evaluated for various qualities parameters. In case of herbal burfi and market burfi the values of optical density, acidity and TPC were increased significantly while, the values of pH, protein content, fat content and sensory scores decreased significantly during 10 days of storage.

Conclusion: The combination of 90% orange juice and 10% stevia extract with 2% of safed musli extract secured highest overall acceptability for herbal beverage.Herbal burfi samples prepared with 90% khoa, 10% stevia powder and 2% safed musli powder ratio was found best.

A CRITIQUE OF THE METABOLIC SYNDROME

László Halmy

Hungarian Society for the Study of Obesity, Budapest, Hungary

Key Words: metabolic syndrome, oxidative stress, vitamin-D insufficiency, hypertension, and diabetes mellitus

The metabolic syndrome is a heterogeneous complex. The diagnosis of metabolic syndrome is made, if at least three out of five factors - dysglycemia, visceral obesity, increased triglycerides, decreased HDL cholesterol, arterial hypertension - are present. There is general consensus regarding the main components of the syndrome but different definitions require different cut points and have different mandatory inclusion criteria. Some of them are very frequent (hypertension, diabetes mellitus) others missing of the criteria (renal diseases, vitamin-D insufficiency, mental disturbances).

Although insulin resistance is considered a major pathological influence and only the International Diabetes Federation (IDF) definition has waist circumference as a mandatory component versus other systems. Therefore we cannot see the metabolic syndrome as a characteristic entity. Available studies on the pathogenesis of metabolic syndrome are discrepant. The oxidative stress is a primary pathogenic mechanism leading to the development of insulin resistance associated with energy

balance. Several organizations have recommended clinical criteria for the diagnosis of metabolic syndrome but they also reveal fundamental differences in their positioning of the predominant causes of the syndrome. Both the popularity and criticism of the metabolic syndrome have increased over recent years. The confusion associated with the meaning and usefulness of the metabolic syndrome.

The metabolic syndrome is a clustering of risk factors which predispose an individual to cardiovascular morbidity and mortality. The therapy of MS is diffuse and it has no main direction in therapy. Further problem is that the abdominal obesity is not obligatory in criteria so the body weight loss is not generally recommended. It seemed to be solved by cardio metabolic syndrome but this could not give answer for all questions. Since the metabolic syndrome is not a uniform genetic, pathogenetic or clinical entity, treatment should target the different components instead of a standardized common treatment.

EVALUATION OF ANTIOXIDANT POWER OF STONE FRUITS FOR DEVELOPMENT OF FUNCTIONAL FOOD

Imtiyaz Murtaza and Hafiza Ahsan

S.K. University of Agricultural Sciences and Technology, Division of Post Harvest Technology, Kashmir, India

Key Words: Stone fruits, total phenols, antioxidant activity

Background: Clearly, all foods are functional, as in addition to providing taste, aroma, or nutritive value they also provide additional physiological benefits. Overwhelming evidence from epidemiological, in vivo, in vitro, and clinical trial data indicates that antioxidant rich plant-based diet beyond that of meeting basic nutritional needs can reduce the risk of chronic diseases, particularly cancer. Among these, stone fruits are known to play an important role in human health due to the range of antioxidant rich phytochemicals in the form of phenolic compounds and carotenoids they contain. Besides sensory and nutritional quality, phenolic compounds are becoming of interest as researchers are discovering their functional activities as drugs, colorants, flavors, and antioxidants. One approach to increase the intake of these beneficial compounds is screening the best cultivars for such capacity and thus increases their concentration inside the fruits by breeding and selection.

Objective: To analyze functional properties of different stone fruit genotypes in terms of antioxidant concentration and antioxidant

activity and select those with highest potential for use in the fresh produce and processing industries.

Methods: Fruit samples of eight plum cultavers, five cherry, five peach and five apricot cultavers with different flesh and skin color were collected from fields of SKUAST (K), Shalimar Campus, Kashmir, India and analyzed for their antioxidant content and antioxidant activity. Total phenolics, anthocyanins, carotenoids and vitamin C content were analyzed spectrophotometrically. Antioxidant activity was evaluated using FRAP assay.

Results: In almost all of the selected stone fruits, a great variation was observed in total carotenoid content, vitamin C and anthocyanin content. However, the results clearly demonstrated that cultavers with higher phenolic content had higher antioxidant activity and thus suggesting a strong correlation between total phenol content and antioxidant activity. Plums showed the highest antioxidant activity followed by cherry, peachs and apricots respectively. Among the plum cultivars, ―Green Gage‖ though smallest in size demonstrates highest phenolic content as well as total antioxidant activity. Likewise, among cherry cultavers ―Siah Gole‖ shows the highest antioxidant activity, among peach ―July Elberta‖ and among apricot ―Hercot‖ cultivar the highest antioxidant activity.

Conclusion: The results clearly suggest that cultivars like Green Guage, showing high content of antioxidant compounds and activity may acquire new interests, mainly due to the fact that they can be considered as a ―functional food‖.

ROSA KANINA (MASUR) AS A PROSPECTIVE SOURCE OF HEALTHY AND FUNCTIONAL FOOD PRODUCT FOR THE PREVENTION OF HYPERTENSION

[1]Martirosyan S, [2]Sodhani S, and [1,2]Martirosyan D

[1]Functional Food Center Inc. Richardson TX, 75080, USA
[2]Department of Nutrition and Food Sciences, TWU, Denton, TX USA

Key Words: Rose hips, hypertension, and functional food

Botany: Rose hips grow from a perennial plant, which can grow one to 4 meters in height. Their thorny branches give way to pink and white flowers and scarlet fruits, called ―hips.‖[1,2] These rose hips are the ripe ovaries or seeded fruit of roses forming on branches after the flower.[3] They are approximately 1 to 3 cm long by 0.5 to 1.5 cm thick; oval in shape. Inside the hips are 3 or more small (3 to 5 mm), angular, yellow-brown seeds.[2] Rosa canina is native to Europe, North Africa, and temperate areas of Asia. Armeian landchaft, especially Artsakh is a very reach in Rosa canina.

History: Once used as a folk remedy for chest ailments, R. canina hips were popular in the Middle Ages.[1] They are a natural source of vitamin C, which has led to their widespread use in natural vitamin supplements, teas, and various other preparations including soups, preservatives, and marmalades.[4] Although these products have been used historically as nutritional supplements, they have also been used as diuretics.[5] Rose hip syrup was used as a nourishing drink for children.[1] It was also used to flavor teas and jams.[2]

Chemistry: Fresh rose hips contain 0.5% to 1.7% vitamin C, [4] usually determined as a combination of l-dehydroascorbic acid and l-ascorbic acid. 6 However, the vitamin C content of dried, commercially available rose hips products varies considerably. One report evaluates stability of vitamin C, using photometry and thin layer chromatography (TLC). Results showed that loss of vitamin C was dependent on ―degree of coarseness‖ of rose hips. Fruits cut in half lost less than 50% vitamin C in 18 months storage, while ground drug lost 100% in 6 months.[6]

While some accounts suggest that rose hips are the richest natural source of vitamin C, a number of more concentrated sources have been identified. Citrus fruits contain approximately 50 mg vitamin C per 100 g; uncooked broccoli, kale, and kiwi fruit, approximately 100 mg; black currants, guavas, and some tropical vegetables, 200 to 300 mg; rose hips (Rosa canina), 1250

mg; acerola or Barbados cherry (Malpighia punicifolia), 1000 to 2330 mg; and Terminalia ferdinandiana , up to 3150 mg.[7]

Rose hips also contain vitamins A, B 1 , B 2 , B 3 , and K. Other ingredients include pectin (11%), tannins (2% to 3%), malic and citric acids, flavonoids, red and yellow pigments, especially carotenoids, polyphenols, invert sugar, volatile oil, vanillin, and a variety of minor components.

Cardiovascular Benefits: According to Winston Craig, Ph.D., rose hips also contain high levels of anthocyanins, catechins, plant sterols and other phytochemicals that have been shown to help reduce the risk of developing cardiovascular disease. Specifically, this effect appears to be due to these agents decreasing levels of C-reactive protein, a protein manufactured in the liver in response to systemic inflammation. High levels of this protein indicate a higher risk for heart disease.

Anti-inflammatory Properties: Dr. Craig also says that rose hip preparations contain antioxidants and may benefit patients suffering from inflammatory disorders, such as osteoarthritis and rheumatoid arthritis. While Craig cites experimentation by Danish scientists with a powder made from rose hip seed, there is evidence that the extract also demonstrates these effects. For instance, in a paper published in Inflammopharmacology, researchers at Kolding Hospital in Denmark reported that rose hip extract produced anti-

inflammatory effects by reducing chemotaxis of peripheral blood neutrophils and monocytes in cultured cells.

Dosage: There is no recent clinical evidence upon which dosage recommandations can be based. Classical use of rose petals was 3 to 6 g daily.[8]

Toxicology: Rose hips ingestion is not generally associated with toxicity.

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CHANGES IN LYCOPENE AND VITAMIN C CONTENT DURING TOMATO RIPENING AFFECT DIETARY REFERENCE INTAKE MANAGEMENT FOR CARDIOMETABOLIC SYNDROME PATIENTS

Majeed R. Al-Ani1[1], Umezuruike Linus Opara[2], Nadia Al-Rahbi[3]

[1]Department of Food Science and Human Nutrition, Sultan Qaboos University, Muscat, Oman,
[2]South African Chair in Postharvest Technology, University of Stellenbosch, South Africa,
[3]Department of Nutrition, Ministry of Health, Oman

Key Words: Tomato; lycopene, vitamin C; biological variability; DRI.

Background: Tomato is one of the most widely grown and consumed product in the world. Several intervention studies suggested that tomato consumption increases protection against oxidative stress, thereby reducing the risk of chronic diseases such as cardiovascular disease and cancer. This protective action is attributed to its high content of bioactive components such as lycopene and vitamin C. Dramatic changes occur in the physico-chemical properties of tomato fruit during maturation and ripening.

Objective: To investigated the changes in lycopene and vitamin C content of cherry tomatoes during ripening in relation to dietary reference intake (DRI).

Methods: Fruit at six ripening stages were analyzed spectrophotometerically for nutrient content. Variations in lycopene and vitamin C were used to calculate the amount of fruit needed to meet DRI.

Results: Nutrient content changed significantly during fruit maturation and ripening ($P \leq 0.05$). In cherry tomato, lycopene content changed significantly from 6.2 to 56.1 mg/100gFW in mature-green and dark-red fruit, respectively, while ascorbic acid content changed from 32.0 to 42.7 mg/100gFW mg/100gFW in mature-green and red fruit, respectively.

Conclusion: The amount of bioactive components in tomato is affected by degree of fruit ripening, which must be considered in nutrition planning and diet management of metabolic syndrome patients.

ADMINISTRATION OF PROBIOTIC BIFIDOBACTERIUM LACTIS 420 REDUCES METABOLIC SYNDROME IN MICE UNDER HIGH-FAT DIET

[1]Olli K., [2]Waget A, [2]Klopp P, [1]Lahtinen S, [1]Ouwehand A, [3]Carcano D, [1]Rautonen N, and [2]Burcelin R

[1] Danisco Health and Nutrition, Kantvik, Finland
[2] Institut National de la Santé et de la Recherche Médicale (INSERM), Toulouse, France
[3]Danisco Health and Nutrition, Paris, France

Key Words: metabolic syndrome, inflammation, probiotic, Bifidobacterium, and LPS

Background: Recent findings indicate that high-fat diet (HFD) is associated with metabolic syndrome and type II diabetes and it induces also metabolic endotoxemia (elevated plasma lipopolysaccharides; LPS) originating from gram-negative microbes. LPS is a potent pro-inflammatory agent, which may be one of the triggering molecules in the chronic low-grade inflammation associated with obesity and related disorders.

Objective: We hypothesized that administration of a probiotic Bifidobacterium lactis 420 may reduce endotoxemia and subsequently inhibit inflammation, improve glucose tolerance and contribute to weight maintenance by reducing weight gain.

Methods: C56Bl/6 mice and CD14 knock-out mice were fed with

normal or high-fat diet to induce a diabetic state. Bifidobacterium lactis 420 was administered daily for four weeks. Diabetic status was measured by intraperitoneal glucose tolerance test (IPGTT) and hyperinsulinemic euglycemic clamp. RNA was extracted from liver and subcutaneous and visceral (mesenteric) adipose tissues for the determination of inflammatory cytokines. Body composition was measured by NMR and bacteriological analyses were done by qPCR. LPS was determined from blood. Epithelial integrity was measured in vitro with Caco-2 cell culture model.

Results: Bifidobacterium lactis 420 reduced fasting glycemia, improved glucose tolerance and reduced insulin resistance. Probiotic treatment reduced tissue inflammation. Weight gain, total fat mass and mesenteric adipose tissue weight of the treated mice were lower than those of the control mice. Probiotic treatment also reduced plasma LPS. Probiotic effect was blunted in knock-out mice lacking CD14 (LPS receptor), suggesting a central role of LPS. Additional mechanistic studies showed that B. lactis 420 improved epithelial barrier function.

Conclusion: Probiotic administration may offer a new strategy for treatment of metabolic syndrome, type II diabetes, and related disorders including obesity and chronic inflammation. The mechanism associated with the positive effect of Bifidobacterium lactis 420 involves reduction of translocation of gut-derived LPS into the host, thus reducing inflammation.

STRUCTURE ANALYSIS OF A NEUTRAL POLYSACCHARIDE ISOLATED FROM FRUIT OF PISTACIA ATLANICA

Omar Ishurd, Mohamed Elmabruk, Faraj Zgheel, Mohamed Elghazoun

Biotechnology Research Center, Twisha, Tripoli, Libya

Key Words: Polysaccharides; Pistacia atlantica, D-Glucan

Background: Pistacia atlantica. is a tree located in north Africa, which can reach over 15 m in height and grows in arid and semi-arid areas, its vernacular name is ―Butom‖. Pistacia atlantica is valued because it is the source of mastic gum, exudates which strengthens gums, deodorizes breath, fights coughs, chills and stomach diseases. Moreover; the galls of Pistacia atlantica which are edible and sold in markets are used as an embalming gradient by rural habitants. Previous studies on P. atlantica deals with flavonoids) , fatty acids and triglycerides, chemical composition of the oleoresin, and chemical composition of the essential oils. Recently, a new hispolone compound has been isolated from the methanolic extract. The structure of the polysaccharide has not been investigated, and we now report on a D-glucan isolated from fruit of Pistacia atlantica that has not been previously studied in detail.

Objective: report on a D-glucan isolated from fruit of Pistacia atlantica that has not been previously studied in detail.

Methods: Dried crushed fruiting (500 g) were cut in to small pieces, crushed by mechanical hammering, and dried. The material was then extracted with hot water (5 L) for 3 h. The process of extraction was repeated, and the extracts were combined, and than centrifuged for 20 min at 5000 rpm. The supernatant solution was diluted with EtOH, and the resulting precipitate was collected by centrifugation, washed four times with acetone, and dried. A solution of the polysaccharide in water (200 mL) was diluted with EtOH, and the precipitate was collected by centrifugation (5000 rpm, 15 min). This process was repeated several times. A portion (100 mg) of the resulting polysaccharide (5 g) was then eluted from a column (2.8×25 cm) of DEAE-cellulose (Pharmacia) anion-exchange resin with water (500 mL), followed by 0.05 M NaOH (500 mL). Fractions (5 mL) were assayed for carbohydrate by the phenol–sulfuric acid method. (Du Bois et al., 1956) Four fractions were obtained, of which NP-1, which had [α]D26 +33° (c 0.1, water) and constituted 43% of the polysaccharide, was used in the subsequent studies. Gel filtration in borate buffer (pH 9.5), through a column (1×30 cm) of Sephadex G-100 (Phamacia), and high-voltage paper electrophoresis at 2 °C in the same borate buffer showed NP-1 to be homogeneous.

Results: Polysaccharide named as NP-1 was isolated from the fruiting of Pistacia atlantica by hot water extraction and 0.05 M NaOH, DEAE-cellulose and Sephadex G-100 column chromatography. Its structural characteristics were investigated by FT-IR, NMR spectroscopy, GLC-MS, methylation analysis, periodate oxidation and Smith degradation. Based on the data obtained, NP-1

was found to be a β-d-glucan containing a (1→3) - and (1→4)-linkage. The anomeric NMR measurements confirm that the sugar residues are β-glycosidically linked.

Conclusion: Based on the data obtained, NP-1 was found to be a β-d-glucan containing a (1→3) - and (1→4)-linkage. The anomeric NMR measurements confirm that the sugar residues are β-glycosidically linked.

EFFECT OF VARIETY AND PROCESSING CONDITIONS ON CURCUMIN AS A BIO ACTIVE COMPONENT AND ITS POTENTIAL IN FORMULATING FUNCTIONAL FOODS

Sawate A. R.,' *Pawar V. N., Yadav G. B., and Kshirsagar R.B.

Department of Food Science and Technology, College of Food Technology, Marathwada Agricultural University, Parbhani, India

Key Words: Curcumin, ration, flavored skim milk, threshold concentration

Background: The turmeric (*Curcuma Longa L*) is a member of the ginger family (Van et al.2004). Turmeric rhizome powder, a recognized seasoning agent has gained its popularity as a spice in various food preparations. In addition to food value, its medicinal importance over the centuries is underlined by its efficacy against non-communicable diseases (Cardiovascular diseases, cancer, liver disorders, rheumatism, diabetic wounds, runny nose, cough and sinusitis). The efficacy governing constituent as a curcumin is notified as a bio active component responsible for reducing the risk of above referred non-communicable diseases. Curcumin as Chinese and Hindu medicine against abdominal pain and sprains and swelling is in use since ancient times (Araujo and Leon 2001). Historical utilization of turmeric as crude medicine provided an option to undertake need base research for therapeutic assessment on the basis of its bio active nature. Recent research findings emerged out from consolidated

medical research progressively helped in elucidating beneficial effects of curcumin over a wide range of diseases and conditions. The predominant efficacy against reducing risk of cancer is justified on the basis of therapeutic and prophylactic mode of action coupled with suppressed proliferation of tumor cells and inhibition of harmful molecules and enzymes by its antioxidant and anti-inflammatory abilities during successive treatment (Agrawal et al. 2003). The earlier studies documented the therapeutic value of curcumin to formulate wide range of drugs on the basis of its medicinal properties. In this investigation efforts have been made to utilize curcumin as one of the constituent ingredients to formulate specialty functional food to justify the innovative emerging slogan as let food be a medicine. The standard formulation of curcumin enriched flavored skim milk emerged out from this investigation provided a horizon to develop a specialty functional foods to harvest health benefits and also to justify its potential as a nutraceutical.

Objectives: To formulate standard recipe of curcumin enriched flavored skim milk to ensure therapeutic health benefits.

Methods: The agro base source of curcumin as rhizomes of Salem and Krishna turmeric cultivars procured from Associate Director (Seed) and Breeder Seed production unit, Marathwada Agricultural University, Parbhani (MS) were processed as per the standard method specified by CFTRI, Mysore.

The standard recipe of flavored skim milk was developed on trial and error basis and assessed on the basis of sensorial quality

parameters by using a semi-trained panel of 11 members. The baseline clinical study to justify its efficacy as a bioactive component by serving 200 ml/day a flavored skim milk ration for one month was assessed on the basis of lipid profile and blood pressure.

Results: The superiority of salem cultivar turmeric powder (curcumin 3.7 per cent) obtained from cabinet drying (temp. at 60^0C) processing techniques was justified for its techno-economic feasibility on the basis of yield and sustainable molecular structure. The characterization profile of curcumin extract, assessed by TLC technique against standard curcumin coupled with RF values at par, recorded its purity.

The per cent decrease in serum cholesterol (2.7 to 9.4 mg %), triglycerides (17.7 to 45.8 mg %), LDL cholesterol (8.9 to 30.3 mg %), VLDL cholesterol (17.7 to 45.8 %) and relative increase in serum HDL cholesterol (3.57 to 20.6 %) was recorded encouraging for further investigation. The hypothetical conclusions emerged out on the basis of data generated from this investigation, primarily justified the efficacy of curcumin as a agro based nutraceuticals to determine its threshold concentration on the basis of further critical clinical studies.

Conclusions: The standard recipe of flavored skim milk containing 0.075 % curcumin, 0.005 % pineapple flavor and 0.5 % fat emerged out from the data of this investigation gave way for development of innovative type of functional food.

PHYSIOLOGICAL FUNCTIONS OF BITTER MELON VARIETIES (MOMORDICA CHARANTIA L.) IN RELATION TO THEIR BIOACTIVE COMPOUNDS

Shahidul Islam*, Jalaluddin M.*, and Hettiarachchy, N. S.**

*Department of Agriculture, University of Arkansas at Pine Bluff, Pine Bluff, AR, USA
**Department of Food Science, University of Arkansas, Fayetteville, AR, USA

Key Words: Bittermelon, protein, polyphenol, antioxidant, antimutagenicity

Background: Many exotic vegetables are known for their special nutritional and medicinal properties. Bitter Melon (Momordica charantia L.), an annual vegetable of Cucurbitaceae family, is found to be one of the important vegetables of special nutritional and medicinal qualities in southern United States. Nutritional qualities of Bitter Melons were examined by chemical analyses namely minerals, polyphenolics, protein and amino acids. Analyses for antioxidants and other biochemical attributes as well as cooking qualities are also investigated. Several recipes have been tasted for consumer acceptance. Along with the potential chemo preventative activities, the popular belief of bitter melon improving glucose tolerance in Type II diabetes and lowering blood cholesterol are being investigated. However, it has not been determined if or which alkaloids, polypeptides, or combination of the chemicals, found in bitter melon, are responsible for the beneficial medicinal effect.

Objective: The functional compounds contained in these foods and their medicinal effects are needed to thoroughly studied and clinically proven.

Methods: Four varieties of bitter melon: Indian Green (IG), Indian White (IW), Chinese Green (CG) and Chinese White (CW), were used for phytochemical analyses for protein extraction and determination, protein hydrolysis, determination of amino acid, antioxidant and atimutagenic activity. All the analyses were done as per the standard methods. All values are reported as means of three determinations. Split plot complete randomized design was conducted using JMP 5 software package (SAS) and Tukey HSD procedure was performed for the significance of differences at the 5% level.

Results: Melon flesh contained approximately 93% moisture for all four varieties, whereas the moisture content of melon seeds ranged from 53.3% in Indian Green to 75% in Indian White. Bitter melon flesh contained 8.4% to 9.8% protein, whereas seed contained 27% to 31% protein.Seed protein was higher in glutamic acid and arginine but lower in lysine compared to flesh proteins. Glycine was also higher in bitter melon compared to soy proteins. Other amino acids contents were similar levels as flesh proteins. . Phenolic contents of seed, SCT, and flesh ranged from 4.67-8.02, 4.64-8.94, and 5.36-8.90 mg/CAE dry matter, respectively. Phenolic contents of the flesh were significantly higher than those of the SCT and seed and phenolic contents of the seed was the lowest among those of all the tissues. The total phenolic contents of four varieties were significantly different

with the highest was Indian white followed by China white, China Green, and Indian green. The antioxidant activities of Indian green, Indian white, China green and China white ranged from 79-88, 79-87, 80-86, and 79-87% inhibition, respectively. The antioxidant activities of the oven-dried samples and the freeze-dried samples were 79-88 and 79-86% inhibition, respectively. Bitter melon varieties IW and CG showed higher antimutagenic effects against benzo(a)pyrene with Salmonella TA98 (92-100% inhibition) and Salmonella TA100 (79-86% inhibition) but lower antimutagenic effects against sodium azide.

Conclusion: Bitter melon is a good source of phenolic compounds. The phenolic extracts showed high inhibition effect to Prevent lipid oxidation. These natural plant phenolics can be a good antioxidant which may be applied in many food systems to maintain the food quality. More Detailed investigations on bitter Melon phenolics, peptides, and proteins are needed to provide information for their nutraceutical values. Bitter melon has high demand in the ethnic market especially in Asian, African and certain South American countries. To the best of our knowledge in normal vegetable usage of bitter melon no toxicity has been reported. However high dose of bitter melon capsule which is now available in the US as well as international market may cause some kind of toxic effect, like over dose of any other polyphenolics it is reported in some non referred sources that higher dose may cause abortion.

ANTIOXIDANT ACTIVITY AND PHENOLIC CONTENT OF COMMONLY CONSUMED INDIAN FOODS

Sreeramulu D. and Raghunath M.

Endocrinology and Metabolism Division, National Institute of Nutrition (ICMR), Tarnaka, Hyderabad, (A.P), India

Key Words: Indian foods, polyphenolic content, and antioxidants

Background: Epidemiological studies indicate the protective effects of plant foods against degenerative dieses and ageing. They exert these effects due to their antioxidant activity (AOA) attributed to their high total polyphenolic content (TPC). Data on AOA and TPC of Indian plant foods is scanty.

Objective: To determined AOA and TPC in most commonly consumed Indian plant foods.

Methods: Based on the National Nutrition Monitoring Bureau surveys. About 110 most commonly consumed foods in India were chosen. Food samples were obtained from three local markets of Hyderabad, India.Edible portions of food samples were extracted with 80% methanol containing 0.1% HCl. TPC and AOA contents were analyzed
by Folin – Ciocalteu method and DPPH / FRAP by standard methods.

Common Name	n (110)	Antioxidant Activity (mg/ 100g)		TPC (mg/100g Gal. equ)
		DPPH (Trol equ)	FRAP ($FeSo_4$ equ)	
Cereals and Millets	9	24 - 173	450 - 13093	47 – 373
Dry Fruits	10	271 - 1541	1174 - 32416	99 – 959
Fresh Fruits	14	32 - 891	22 - 496	26 – 374
Green Leafy Vegetables	11	21 - 1020	1380 – 27 827	77 – 1077
Milk and Milk products edible oils and Sugars	14	3 - 208	11 - 11674	0.72 - 336
Nuts and Oil Seeds	12	20 - 28622	220 -4220341	10– 10841
Pulses and Legumes	11	26 - 107	1469 - 10362	62 – 418
Roots and Tubers	10	11 - 125	256 - 6308	22 – 169
Vegetables	19	12 - 466	243 – 105	27 - 339

The results are expressed as Gallic acid equivalents and Trolox / $FeSo_4$ equivalents respectively.

Results: Data on TPC and AOA content of commonly cosumed foods in India, the range of values are as fallows. Foods studied had good amount of polyphenols and antioxidant activity, despite the fact that they belonged to different groups. Interestingly, significant correlation was observed between AOA (DPPH and FRAP) and TPC in most of the foods corroborating literature from other parts of the world. Findings suggest that polyphenolics are potent antioxidants and that they may be important contributors to the AOA of plant foods studied.

Conclusion: To the best of our knowledge, results obtained in this study, the first of its kind from India. This data would be useful to

nutritionists and consumers to assess and/or formulate antioxidant – rich therapeutic diets. In addition, they will be valuable addition to the scanty knowledge on commonly consumed Indian foods.

FOOD BORNE OUTBREAK OF GASTROENTERITIS IN TIBETAN TRANSIT SCHOOL, DHARAMSHALA, HIMACHAL PRADESH, INDIA, 2006

Gupta SN[1], Gupta NN[2] , Mehta N[3]

[1] MAE-FETP Graduate from NIE, Chennai; presently Epidemiologist at Regional Health and Family Welfare Training Centre, Chheb, Kangra, Himachal Pradesh, India
[2] Freelance researcher in Epidemiology, Kangra
[3] District Leprosy Officer, Kangra

Key Words: Outbreak, gastroenteritis, Tibetan Transit School, Himachal Pradesh

Background: On 28th June, 2006 we investigated a food borne outbreak of gastroenteritis among the hostellers of Tibetan Transit School to identify the source, propose control and preventive measures.

Methods: We defined a case of gastroenteritis as occurrence of more than three smelly loose motions between 28th of June to the 2nd of July, 2006 among some sections of the hostellers. We determined age and sex specific attack rate. We hypothesized it as a food borne beef meat outbreak. We conducted case control study and collected the information about the food items consumed inside and outside the hostel at dinner using the standardized questionnaire. We calculated floor wise incidences of four hostels, odds ratios and attributable fractions. We interviewed food handlers. We lifted seven rectal and four water samples for culture and sensitivity.

Result: We identified 116 cases patients. Overall attack rate (AR) as 14% with maximum AR (16%) and floor wise incidences as highest (40%) in the youngest group (15-20yrs) and nil in staff, median age 20 yrs (Range 17-40 yrs). Sex specific attack rate was more (18%) in females. Of the six edible items examined, food specific AR was highly statistically significant in the beef meat eaters (82% with PAF 71%; OR 19.19; 95% C.I. as 9.3-140). The food handlers and cooking conditions were unhygienic. Escherichia coli were detected in the given samples. No fatality was reported.

Conclusion: The beef meat purchased from outside was implicated for the explosive common source outbreak. The school authorities were counseled for hygienic food handling.

BHUX: A PATENTED POLY-HERBAL FORMULATIONS TO PREVENT PROGRESSION OF ATHEROSCLEROSIS AND PLAQUE STABILIZATION

Yamini Bhusan Tripathi

Department of Medicinal Chemistry, Institute of Medical Sciences, Banaras Hindu University, Varanasi, India

Key Words: Atherosclerosis, inflammation, herbal preparation, lipid profile, HDL cholesterol, functional food

Atherosclerosis is associated with free radical mediated reactions including inflammation. BHUx (EU#1583499 dated-25.6.2009, US#20060147555, ZL200380109770.X dated-17.06.2009), consisting specific fraction of 5 medicinal plants namely *Termenalia arjuna, Strychnox nux vomica, Boswellia serrata, Commiphora mukul, and Semecarpus anacardium,* has significantly inhibited LPS induced changes and specifically inhibited lipoxygenase-15 and cyclooxygenase-2 enzymes, suggesting its anti-inflammatory property. It has significantly prevented the progression of vascular lipid deposition and reduced raised lipid profile with rise in HDL cholesterol in diet induced hyperlipidema and atherosclerosis in rabbits and in ApoE knockout mice. It maintained the collagen cap and reduced Ca content in existing plaque. These observations were substantiated with cell culture experiments with HUVEC cell lines to explore its role on expression of M-CSF and with VSMC culture to assess its role on

proliferation and calcification. BHUx was found to be safe in preclinical toxicity study in treatment span of 90 days. Thus it was suggested that BHUx is a promising herbal preparation, which can be used as functional food for persons, predisposed to atherosclerosis and associated complications.