

DIAMEL INTERVENTION TRIAL ON METABOLIC SYNDROME: BASELINE DATA

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INTRODUCTION

The metabolic syndrome (MS) has attracted attention of the scientific community since it constitutes one of the most polemic medical entities on the last years. MS is defined as an association of several risk factors that precede atherosclerotic cardiovascular disease and type 2 diabetes. This concept was unified by Reaven in 1988, after observing that dyslipemia, hypertension, and hyperglycemia used to commonly appear grouped, naming this association X syndrome. He postulated that insulin resistance and compensatory hyperinsulinemia played an important role in X syndrome physiopathology. Thus, some scientists called this condition insulin resistance syndrome. At present, prestigious health and scientific association and organizations have proposed clinic guides to facilitate the diagnosis and treatment of MS in the adulthood: WHO in 1999, the European Group for Study of Insulin Resistance (EGIR) in 1999, The National Cholesterol Education Program (NCEP), the Adult Treatment Panel III (ATP III) in 2002, the American Association of Clinical Endocrinologist (AACE) 2003, and the International Federation of Diabetes (IDF) in 2005. At present, several drugs have been used for the treatment of MS like metformin, statins, fenofibrate, gemfibrozil, thiazolidinediones among others. On the other hand, DIAMEL, a natural product derived from lettuce and blueberry extracts has been incorporated in the market. DIAMEL also contains vitamins and trace elements acting as biocatalysts and antioxidants. DIAMEL administration in type 2 diabetic patients has been proved to reduce blood triglycerides and glucose concentrations after 2 months of treatment indicating the probable positive action on insulin resistance. Hernández Yero and Vargas in 2006, found that patients treated with a combination of DIAMEL and glimeclamide improved metabolic control and beta cell function after 6 months, compared to patients treated with glimeclamide alone. Accordingly, we believe that DIAMEL could be effective in the treatment of persons with MS. There are no reports of the use of DIAMEL on MS. Thus, we propose to carry out a randomized double blind controlled intervention trial in persons with MS with confirmed presence of insulin resistance.

METABOLIC SYNDROME (MS)

Several cardiometabolic risk factors, that are likely to be linked to insulin resistance

Diamelel administration

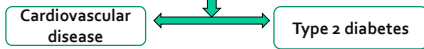


Fig 1. Diamelel intervention trial on Metabolic Syndrome

AIMS

To evaluate the effect of Diamelel administration on the clinical and metabolic parameters of metabolic syndrome. To set up a clinical trial to establish whether Diamelel influence on the clinical and metabolic parameters of metabolic syndrome. To access the effect of DIAMEL on clinic and metabolic characteristics of persons with MS

SUBJECTS AND METHODS

Subjects

We screened 180 overweight or obese subjects. **Inclusion Criteria:** Individuals who had criteria of the Metabolic Syndrome (MS) according to WHO but without previous or current drug treatment for elevated blood glucose concentrations, age 19-70 years. **Exclusion Criteria:** We excluded people who had a chronic disease that was likely to affect outcome, toxicity, or adherence to the protocol; women who were breastfeeding, pregnant. Participants who were shown to have type 1 diabetes, type 2 diabetes treated with oral hypoglycemia agents; as well as subjects with insulin resistance diseases (acromegaly, endogenous hypercortisolism, among others); and sepsis were also excluded. Data for gender, color of the skin, BMI, waist and hip circumference, blood pressure, serum glucose and lipids concentrations were obtained. Subjects of the study group was further studied for the presence of acanthosis nigricans and antibodies to GAD, IA-2, as well as for free cholesterol, creatinine and uric acid concentrations. All subjects gave their informed consent to be studied. The local Ethical and Research Committee approved the protocol. Diamelel are register as a food supplement in the National Institute of Nutrition and Food. Diamelel is a food supplement made of lettuce and blueberry extracts as well oligolements. **Ingredients:** Arginine, Glycine, Fumaric Acid, Ascorbic Acid, L-Carnitine, Ornithine, Acetylcysteine, Calcium Pantothenate, Pyridoxal, Folic Acid, Cyanocobalamin, Lettuce Extract, Blueberry Extract and Zinc Sulfate.

Definition

Metabolic syndrome was considered present by modified WHO definition if the subject had fasting glucose ≥ 6.1 mmol/L or was in the highest quartile of HOMA (insulin resistance when HOMA IR ≥ 2.64) or had previously diagnosed diabetes and at least two of the following: blood pressure $\geq 140/90$ mmHg; triglycerides ≥ 1.7 mmol/L or HDL-cholesterol < 1.0 mmol/L in women and < 0.9 mmol/L in men; BMI > 30.0 kg/m² or waist-to-hip ratio (WHR) > 0.85 in women and > 0.90 in men.

Study Design: The Diamelel intervention trial is a randomized, double-blind, placebo-controlled intervention trial undertaken in Cuba. Participant were randomly allocated either oral Diamelel or placebo at a dose of two capsules before the three main meals of each day for one year. After stratifying by age (< 45 and ≥ 45 years of age), codes were randomly assigned to each participant. Randomization was generated using computerized random number generator. All personnel involved in the study remained unaware of the correspondence between codes and the content of the capsules. Study food supplement (Diamelel, 660 mg and placebo) was supplied by Catalysis, s.l. Madrid, Spain labeled with the randomization codes only. Code-to-pill-content addition was kept in a sealed envelope under the custody of the Catalysis group. The envelope will be open at the end of the study.

Adverse Effects: Every 3 month clinical examination will be performed. Adverse events such as rashes and dyspepsia will be recorded.

Methods

Fasting blood glucose: Plasma glucose concentration was determined by glucose oxidase method.

Fasting insulin: Fasting plasma insulin concentration was measured by radioimmunoassay (RIA).

Insulin resistance index: Insulin resistance index was calculated according to the homeostasis model assessment (HOMA IR) of Matthews (insulin x glucose/22.5).

Cholesterol and Triglycerides: Serum cholesterol and triglycerides were determined using a routine enzymatic methods.

HDL- Cholesterol: HDL-cholesterol was determined after precipitation of apolipoprotein B-containing lipoproteins.

GAD65 and IA-2 autoantibodies: Autoantibodies against GAD65 and tyrosine phosphatase were detected by quantitative radioimmuno-precipitation assays, using a commercial kit (SHERING SA, CI bio international, France), which utilizes human recombinant 125I-labelled GAD65 or 125I-labelled IA-2 and protein A-sepharose to separate free from antibody-bound labelled GAD65 or IA-2, respectively. Values above 1 U/ml were considered as pathological for GAD and IA-2A. These values were determined in sera of apparently healthy controls without family history of diabetes (n=200) with anti-GAD and anti-IA-2 immunoradiometric assay. 99.5% of anti-GAD concentrations and 100% of anti-IA-2 concentrations were below 1 U/ml respectively.

Statistical analysis: Data are presented as means \pm SD. T-tests tested for differences in means of continuous variables with Gaussian distributions. Mann-Whitney test was used otherwise. Proportions were compared using the chi-squared test or Fisher exact test, whenever appropriated. p values less than 0.05 were considered significant.

RESULTS

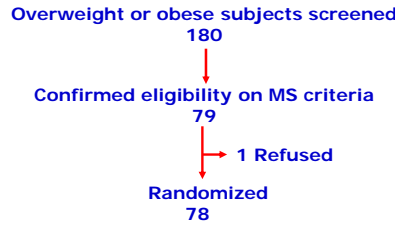


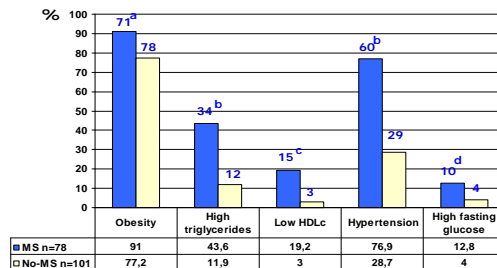
Fig. 2. Subject disposal

Table 1. Comparison among subjects free of metabolic syndrome and WHO-defined metabolic syndrome

Characteristics	WHO-MS* n = 78 N (%)	No-MS n = 101 N (%)	p value
Gender			
Female	57 (73.1)	94 (93.1)	
Male	21 (26.9)	7 (6.9)	< 0,0005
Color of the skin (White)	37 (47.4)	57 (56.4)	
Corporal weight			
Overweight	7 (8.9)	23 (22.7)	
Obese	71 (91.0)	78 (77.2)	0,026
Acanthosis Nigricans	61 (78.2)	60 (59.4)	0,010
Iset autoantibodies			
GADA- positive	2 (2.5)	1 (0.9)	
IA-2A- positive	1 (1.2)	0 (0.0)	
	Mean \pm SD	Mean \pm SD	
Age (years)	44.13 \pm 11.73	40.15 \pm 11.16	0,022
Weight (kg)	100.29 \pm 20.76	89.32 \pm 17.67	< 0,0005
Height (cm)	163.76 \pm 9.31	162.169 \pm 8.09	
BMI (kg/m ²)	37.28 \pm 6.51	33.91 \pm 5.93	< 0,0005
Waist circumference (cm)	108.83 \pm 14.11	99.39 \pm 11.33	< 0,0005
Waist-to-hip ratio	0.905 \pm 0.083	.859 \pm 0.088	0,001
Systolic b pressure (mmHg)	132.27 \pm 26.58	115.61 \pm 14.53	< 0,0005
Diastolic b pressure (mmHg)	88.96 \pm 16.47	76.93 \pm 11.69	< 0,0005
Fasting glucose (mmol/L)	4.97 \pm 1.03	4.43 \pm 0.82	< 0,0005
Fasting insulin (μ U/ml)	23.37 \pm 12.51	12.78 \pm 7.20	< 0,0005
HOMA-index	5.09 \pm 2.72	2.53 \pm 1.52	< 0,0005
Total cholesterol (mmol/L)	4.81 \pm 0.91	4.56 \pm 0.78	0,058
Triglycerides (mmol/L)	1.65 \pm 0.38	1.43 \pm 0.32	< 0,0005
HDL-Cholesterol (mmol/L)	1.18 \pm 0.30	1.29 \pm 0.31	0,022
Creatinine (mmol/L)	98.28 \pm 36.39	92.61 \pm 23.31	
Uric Acid (mmol/L)	348.24 \pm 73.64	286.43 \pm 73.91	< 0,0005

*WHO-MS: metabolic syndrome (randomized) by WHO definition

No-MS: subjects free of metabolic syndrome (WHO negative)

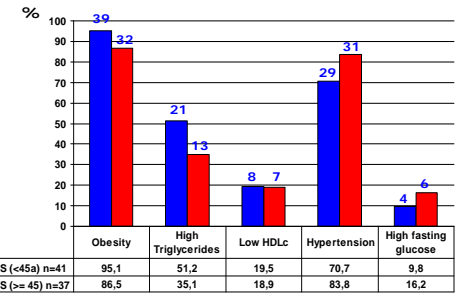


n Total of subjects
a p = 0,026 vs No-MS; b p < 0,0005 vs No-MS; c p = 0,001 vs No-MS; d p = 0,046 vs No-MS. The number of subjects for each component is reflect over the columns

Graphic 1. Frequency of the individual components of Metabolic Syndrome according to the presence or not of MS

Table 2. Baseline characteristics of trial entrants

Characteristics	Age below 45 n = 41 N (%)	Age \geq 45 n = 37 N (%)	p value
Gender			
Female	31 (75.6)	26 (70.3)	
Male	10 (24.4)	11 (29.7)	
Color of the skin (White)	17 (41.5)	20 (54.1)	
Corporal weight			
Overweight	2 (4.9)	5 (13.5)	
Obese	39 (95.1)	32 (86.5)	
Acanthosis Nigricans	35 (85.4)	26 (70.3)	
Iset autoantibodies			
GADA- positive	0 (0.0)	2 (5.4)	
IA-2A- positive	0 (0.0)	1 (2.7)	
	Mean \pm SD	Mean \pm SD	
Age (years)	35.46 \pm 6.74	53.73 \pm 7.97	NA
Weight (kg)	103.83 \pm 20.09	96.37 \pm 21.06	
Height (cm)	163.76 \pm 9.31	162.169 \pm 8.09	
BMI (kg/m ²)	38.04 \pm 5.95	36.43 \pm 7.06	
Waist circumference (cm)	109.37 \pm 15.11	108.23 \pm 13.09	
Waist-to-hip ratio	0.903 \pm 0.094	0.907 \pm 0.072	
Systolic b pressure (mmHg)	130.27 \pm 30.04	134.49 \pm 22.32	
Diastolic b pressure (mmHg)	89.41 \pm 19.30	88.46 \pm 12.90	
Fasting glucose (mmol/L)	4.98 \pm 1.12	4.97 \pm 0.93	
Fasting insulin (μ U/ml)	24.70 \pm 11.78	21.89 \pm 13.27	
HOMA-index	5.49 \pm 2.87	4.65 \pm 2.51	
Total cholesterol (mmol/L)	4.66 \pm 0.89	5.03 \pm 0.88	0,036
Triglycerides (mmol/L)	1.70 \pm 0.42	1.60 \pm 0.34	
HDL-Cholesterol (mmol/L)	1.16 \pm 0.26	1.21 \pm 0.34	
Creatinine (mmol/L)	101.17 \pm 45.03	95.08 \pm 23.66	
Uric Acid (mmol/L)	322.27 \pm 51.74	365.95 \pm 89.51	0,043



n Total of subjects
The number of subjects for each component is reflect over the columns

Graphic 1. Frequency of the individual components of Metabolic Syndrome according to age of the entrants

Table 3. Concentration of total cholesterol and Uric acid in No-MS subjects after stratification by age

Characteristics	Age below 45 n = 69	Age \geq 45 n = 32	p value
	Mean \pm SD	Mean \pm SD	
Total cholesterol (mmol/L)	4,43 \pm 0,66	4,86 \pm 0,93	0,023
Uric Acid (mmol/L)	280,57 \pm 78,13	299,06 \pm 63,15	

- Utilizing WHO criteria to define MS, the reported frequency of MS in the present study (43,8%; 79/180) is similar to that reported for type 2 diabetic European subjects (20-50%).
- The frequency of MS of our sample is similar to a previous work conducted in first degree relatives of Cuban type 2 diabetic patients (42,9%)
- Individual components of MS according to WHO criteria do not appear to be modified by age in the trial entrants

CONCLUSIONS

- Diamelel intervention trial has shown that the use of natural products together with indications for lifestyle improvement aiming at diminishing risk factors for future development of type 2 diabetes or cardiovascular disease is feasible and has high acceptance levels on obese or overweight subjects informed to be "labeled" as persons with MS.
- Uric acid but not cholesterol appears to be associated with age only on MS individuals indicating that this marker could be useful for the screening of MS