Nutrición Hospitalaria

Nutr Hosp. 2015;32(2):710-721
ISSN 0212-1611 • CODEN NUHOEQ
S.V.R. 318

Original/*Alimentos funcionales*

Impact of improved fat-meat products consumption on anthropometric markers and nutrient intakes of male volunteers at increased cardiovascular risk

Paloma Celada[1], Gonzalo Delgado-Pando[2], Begoña Olmedilla-Alonso[2], Francisco Jiménez-Colmenero[2], Mar Ruperto[1] and Francisco J. Sánchez-Muniz[1]

[1]Departamento de Nutrición y Bromatología I (Nutrición). Facultad de Farmacia. Universidad Complutense, Madrid. [2]Instituto de Ciencia y Tecnología de los Alimentos y Nutrición (ICTAN), CSIC. 28040-Madrid, (Spain).

Abstract

Introduction: **meat products have been recognized to be adequate matrix for incorporating functional ingredients. The impact of meat products formulated by replacing animal fat with a combination of olive, linseed and fish oils on energy and nutrient intakes and anthropometric measurements were tested in a non-randomized-controlled-sequential study.**

Methods: **eighteen male volunteers at high-CVD risk consumed weekly 200 g frankfurters and 250 g pâtés during three 4-wk periods (reduced fat (RF); n3-enriched-RF (n-3RF), and normal fat (NF)), separated by 4-wk washout. Energy and nutrient intakes, healthy eating index (HEI), and anthropometric changes were evaluated.**

Results: **body fat mass rate-of-change and the waist/hip ratio significantly differs (p = 0.018 and p = 0.031, respectively) between periods, decreasing body fat mass, waist circumference and waist/hip ratio in RF period and increasing body fat mass in NF one (all p = 0.05). Significant inverse correlations were observed between rate-of-change of BMI and ideal body weight with dietary carbohydrate/SFA ratio in n-3RF period (p = 0.003 and p = 0.006, respectively). Initial diets presented low HEIs (means < 60). Carbohydrate, fat and protein energy contribution was 40%, 41%, and 16%, respectively. More than 33% of volunteers did not initially cover 70% of several minerals and vitamins RDAs. Product consumption improved dietary Zn, Ca, retinol equivalent, folate and vitamin B_{12} contents in all periods, and ameliorated n-3 PUFA contents and n-6/n-3 PUFA ratio over the n-3RF period.**

Correspondence: Francisco José Sánchez-Muniz.
Departamento de Nutrición y Bromatología I (Nutrición).
Facultad de Farmacia. Universidad Complutense.
28040-Madrid, Spain.
E-mail: frasan@ucm.es

Recibido: 11-V-2015.
Aceptado: 27-V-2015.

IMPACTO DEL CONSUMO DE PRODUCTOS CÁRNICOS CON CALIDAD GRASA MEJORADA SOBRE MARCADORES ANTROPOMÉTRICOS E INGESTA DE NUTRIENTES EN VOLUNTARIOS CON RIESGO CARDIOVASCULAR ELEVADO

Resumen

Introducción: **la carne es una matriz adecuada para la inclusión de ingredientes funcionales. En un estudio no secuencial controlado y aleatorio se evaluó el impacto del consumo de productos cárnicos, en los que se sustituyó la grasa animal por una combinación de aceite de oliva, de linaza y de pescado, sobre la ingesta de energía y nutrientes y sobre los marcadores antropométricos.**

Métodos: **dieciocho voluntarios con elevado riesgo cardiovascular consumieron semanalmente 200 g de salchichas tipo frankfurt y 250 g de paté durante tres períodos sucesivos de 4 semanas (bajo en grasa (RF); enriquecidos en n-3 (n-3RF), y grasa normal (NF)), separados por un lavado de 4 semanas. Se evaluó la ingesta de nutrientes y energía, el índice de alimentación saludable (HEI) y los cambios antropométricos.**

Resultados: **hubo diferencias significativas entre períodos para las tasas de cambio de la grasa corporal y de la relación cintura/cadera (p = 0,018 y p = 0,031, respectivamente), disminuyendo la masa grasa, el perímetro de la cintura y la relación cintura/cadera en el periodo RF, e incrementándose la grasa corporal en el periodo NF (todos p = 0,05). En el período n-3RF las tasas de cambio de IMC y del peso ideal correlacionaron inversa y significativamente (p = 0.003 y p = 0.006, respectivamente) con el cociente hidratos de carbono/AGS. El HEI inicial de las dietas fue muy bajo (valor medio < 60). La contribución energética de carbohidratos, grasa y proteínas fue 40%, 41% y 16%, respectivamente. Más del 33% de los voluntarios no cubrían al inicio el 70% de las RDA para minerales y vitaminas. La intervención mejoró en todos los períodos la ingesta de Zn, Ca, equivalentes de retinol, folatos y vitamina B_{12}. En el período n-3RF incrementó los AGPn-3 y redujo el cociente n-6/n-3.**

Conclusion: **improved-fat meat products appear as functional foods for overweight/obeses since their consumption improved selected body-fat markers, without affecting HEI, macronutrient and energy but their n-3 PUFA and n6/n3 ratio intakes.**

(*Nutr Hosp*. 2015;32:710-721)

DOI:10.3305/nh.2015.32.2.9231

Key words: *Dietary assessment. Pâtés. Frankfurters. n-3PUFA. Reduced-fat. Functional foods. Omega-3-enriched meat.*

Conclusión: **los productos cárnicos con menos grasa o enriquecidos en AGP n-3 son alimentos funcionales para personas con sobrepeso/obesidad, ya que su consumo mejora los marcadores de grasa corporal, los niveles de AGP n-3 y el cociente n-6/n-3 sin afectar al HEI ni a la ingesta de energía y macronutrientes.**

(*Nutr Hosp*. 2015;32:710-721)

DOI:10.3305/nh.2015.32.2.9231

Palabras clave: *Evaluación nutricional. Patés. Salchichas frankfurt. AGPn-3. Productos cárnicos enriquecidos en omega-3. Productos cárnicos reducidos en grasa. Alimentos funcionales.*

Abbreviations

CVD: Cardiovascular diseases.
MUFA: Monounsaturated fatty acids.
NF: Normal-fat.
n-3RF: n-3PUFA-reduced fat.
PUFA: Polyunsaturated fatty acids.
RDA: Recommended dietary allowances.
RF: Reduced-fat.
SFA: Saturated fatty acids.
%En: Contribution to the total energy.

Introduction

Meat is recognized as a central food for human beings as it is a major source of high biological value proteins, minerals and vitamins, contributing in some countries to 20 to 50% of energy and several nutrient intakes[1]. Meat and meat derivatives are widely consumed in Spain[2]. However, the image of this food-group has deteriorated considerably due to meat and processed foods consumption has been associated with increased risk of cardiovascular disease (CVD) and other degenerative diseases[3,4]. Moreover, excessive meat intake is considered an indicator of unhealthy diets[2]. Nonetheless, moderate amounts of lean red meat provide a wide range of important nutrients without substantially increasing intakes of energy and saturated fatty acids (SFA)[5].

Functional foods, including meat-products[6], are foods with demonstrated beneficial effects on one or more target functions in the body beyond adequate nutritional effects, in a way that is conducive to either an improved state of health and well-being and/or reduction of risk of disease[7]. Functional foods must be demonstrably effective in amounts that can normally be expected to be consumed as part of a normal food pattern[7].

SFA replacement by polyunsaturated or monounsaturated fatty acids (PUFA or MUFA, respectively) seems to reduce the CVD risk, with stronger evidence for PUFA[8-10]. Several strategies (e.g. using olive oil or n-3-rich oil/fat to partially replace SFA) can be used for their healthy properties[11,12]. In fact, the consumption of n-3PUFA- or fat impoverished-meat products induced beneficial effects on the lipoprotein profiles of volunteers at increased CVD risk[11].

Dietary PUFA *vs*. SFA have been found to modify the lipogenesis and lipolysis in the adipose tissue[13] with modest effect of fish oil vs. sunflower oil on body fat but not on body weight[14]. In addition, the carbohydrate/SFA ratio has been related with insulin resistance[15,16]. However, a study of diet characteristics during dietary interventions with functional foods is essential since positive or negative interactions would affect results.

This paper hypothesized that the inclusion in the habitual diet of modified fat-frankfurters or pâtés improves some anthropometric measurements but did not significantly modify food consumption and dietary patterns. This paper, therefore, aimed to study in volunteers at increased CVD risk who participated in a placebo-controlled study with improved-fat meat products a) the basal diet characteristics; b) the impact of dietary modifications induced by consumption of three meat-derivatives in which animal fat was replaced by a combination of olive, linseed and fish oils, on the macro- and micronutrient intakes; c) to analyse effects on body weight, BMI, body fat, and conicity index; and d) to find out relationships between changes in anthropometric measurements with those in dietary intakes.

Methods and materials

Subjects

Twenty-two volunteers, 18 men and 4 women, were enrolled and completed the study. Participants were selected from 48 subjects who were interested and contacted through advertisements in different universities, research centres and notice boards. Study design and enrolment inclusion and exclusion criteria have been published in detail elsewhere.[11] In short, the selected subjects fulfilled the inclusion criteria: total cholesterol levels > 200 mg/dl, LDL cholesterol levels > 110 mg/dl, overweight (BMI > 25 and < 34.9 kg/m^2) and willingness to consume 200 g of frankfurters and 250 g of pâté per week. The exclusion criteria were: use of drugs or plant sterol-enriched beverages/foods to control cholesterol levels, hypertension or obesity; regular consumption of n-3-enriched food-products; and intolerance or food allergy to any of the components of the meat-products. They were also instructed to maintain a mixed diet (no avoidance of any food-groups) and to replace the helpings of meat and meat-products of their habitual diet with helpings of the meat-products provided. Moreover, as all selected participants were sedentary, they were also instructed to maintain the physical activity patterns to avoid possible results biases. The study was approved by the Ethical Committee for Clinical Investigation of the Hospital Universitario Puerta de Hierro-Madajahonda (Spain) (Acta nº 261, dated 20/12/2010) and the Bioethical Committee of the Spanish National Research Council. All subjects gave their written informed consent after receiving verbal and written information about the study.

However, anthropometric markers (e.g. hip and waist circumferences and conicity index) differ between men and women[17] and several clinical and intervention studies have confirmed differences in response to diet on the body weight and/or fat percentage between males and females[18]. Thus, present paper was performed only in the 18 men. To confirm such previous facts in our study, rate of change of two selected anthropometric parameters % body fat and BMI- in men and women were compared. Both gender showed differences in response to diet (Supplementary figures 1 and 2).

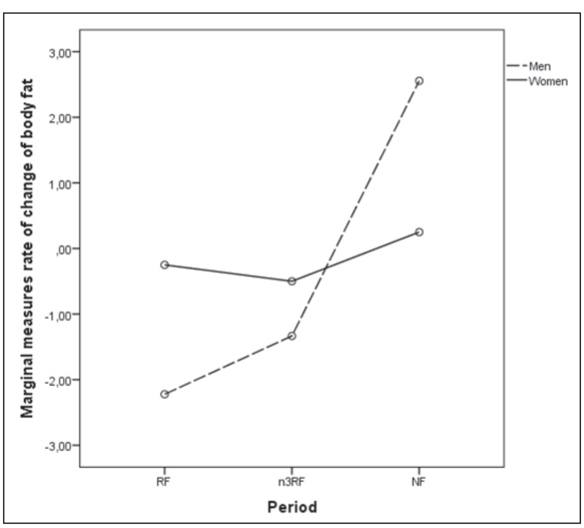
Meat products

Frankfurters and pâtés were developed at the Institute of Food Science, Technology and Nutrition (ICTAN, CSIC, Madrid, Spain). Three different batches were produced: a) reduced-fat-products (RF) (15% fat); b) reduced-fat (15% fat) n-3-enriched-meat-products (n-3RF) where the animal fat source (pork backfat) was replaced by a combination of olive, linseed and fish oils; and c) normal-fat control-products (NF) (18% fat for frankfurters, 30% fat for pâtés). This oil combination was used as an ingredient in an oil-in-water emulsion, with olive, linseed and fish oil proportions of 44.39%, 37.87% and 17.74%, respectively. RF frankfurters and pâtés were reformulated by reducing their fat content, and n-3RF products by reducing the fat content and replacing the pork backfat with the oil combination mentioned above, yielding meat products with an improved lipid profile[19,20]. The NF products were similar (in fat content to those usually found in the market. Additionally, the technological viability of these products has been established[19,20]. In the present study, some slight composition modifications have been introduced in the frankfurter development in order to achieve the established fat levels.

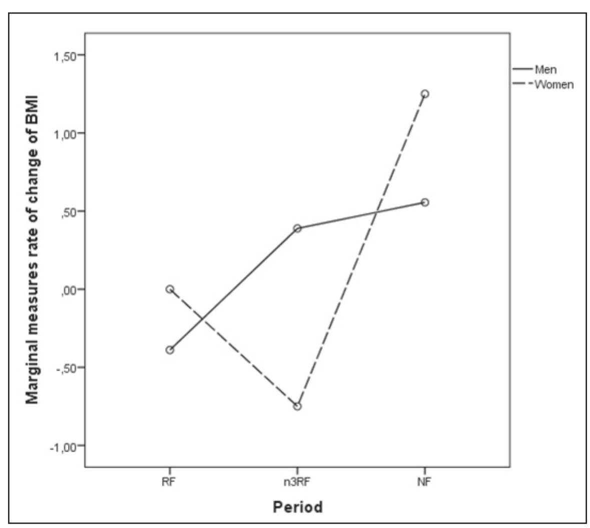
Containers of 250 g of pâté and 200 g of vacuum packed frankfurters were prepared. At the beginning of every intervention period, the meat products were delivered under refrigeration and stored at 4ºC until consumption. Frankfurters and pâtés were ready for consumption. Once open, the containers should be consumed during a week period.

Intervention study design

Volunteers were enrolled in a non-randomized sequentially controlled study of 5 months duration. Each one consumed 200 g frankfurters and 250 g pâtés per week, in each of the three periods, followed



Supplementary figure 1.—Marginal measures rate of change (%) of body fat in men and women for the three experimental periods. RF, Reduced-fat-meat-product; n3RF, n3-modified-reduced-fat-product; NF, Normal-fat-product.



Supplementary figure 2.—Marginal measures rate of change (%) of body mass index (BMI) in men and women for the three experimental periods. RF, Reduced-fat-meat-product; n3RF, n3-modified-reduced-fat-product; NF, Normal-fat-product.

by a 4-week washout in each study phase[11]. During the washout periods the subjects followed their habitual diets, making it possible to compare the effect of consumption of the "healthier" meat-products against their habitual diets and thus proving the effectiveness of regular consumption of these products[21]. During the first dietary intervention volunteers consumed RF-products in which the fat source was 100% of animal origin. In the second dietary intervention they consumed 450 g of n-3RF-enriched frankfurters and pâtés, providing 2 g of n-3PUFA per day, of which 1.5 g was linoleic acid and around 0.4 g eicosapentaenoic acid *plus* docosahexaenoic acid. Finally, during the third dietary intervention normal fat frankfurters and pâtés (NF) were consumed. These NF-products were similar in fat content to those usually found in the market.

Anthropometric measurements

Anthropometric measurements were performed in each period. Body weight and height were measured in fasting conditions to the nearest 0.1 kg and 0.1 cm respectively using an electronic digital scale (Omron BF400 Body Fat Monitor with scale, Hoofddorp, the Netherlands). Body Mass Index (BMI) was calculated as body weight (kg) divided by the square of body height (m^2). Waist circumference was measured to the nearest 1mm midway between the lowest rib margin and the iliac crest while hip perimeter was the higher circumference obtained at the level of greater trochanteres. Circumferences were measured with the subject standing with weight distributed equally between the two legs and feet about 12-15 cm apart. The ratio between waist and hip perimeters was obtained. Body fat mass (in percentage) was assessed by bioimpedance (Omron BF306, Hoofddorp, the Netherlands). In addition to BMI and other commonly used indices, the conicity index[22] was evaluated. Conicity index (CI) was calculated using the following formula CI = Waist circumference / [0.109 x the square root of (weight / height)] where waist circumference and height were expressed in meters and weight in kg[22]. The Ideal-body-weight (in percentage) was calculated dividing weight between ideal weight and multiplying this ratio by 100. Ideal weight data was taken from Alastrué tables[23].

Dietary assessment

In order to evaluate food consumption and hence nutrition intake and nutritional status, volunteers were interviewed by a dietician for 1 to 1.5-h. Food intake and food habits were registered. Photographs of sample portions were used to estimate the serving size and volumes consumed when this information was not clearly available. The dietary information was divided into 72-h food and drink consumption, food frequency and food consumption pattern registries. The energy and nutrient intakes per person and the health eating index (HEI) were calculated using a computer program (DIAL™ Madrid, Spain)[24] to evaluate global diet quality consumed. The foods consumed were grouped into cereals, milk, eggs, sugar, oils and fats, vegetables, legumes, fruits, meat and meat products, fish, shellfish, drinks and miscellaneous. The following nutrients were quantified: proteins, lipids, carbohydrates, fibre, vitamin B_1, vitamin B_2, niacin equivalents, vitamin B_6, folates, vitamin B_{12}, vitamin C, retinol equivalents (retinol eq.), vitamin D, vitamin E, biotin, vitamin K, vitamin B_5, calcium, iron, iodine, zinc, magnesium, selenium, and fluorine. The HEI index used, based on a 10-component, 100-point scale, was a slight modification of Norte Navarro and Ortiz Moncada[25], taking into account recommended energy intakes of 1600, 2200 and 2800 kcal[24,26] and the required servings[24,27]. Diets with HEI scores of < 70 were labelled "inadequate", while those with HEI-scores of ≥ 70 were considered "adequate"[28].

Statistical analysis

The Kolmogorov-Smirnov test was used to assess normal value data distribution. The paired Student *t*-test was employed to determine the effect of the different meat-products in each intervention period. The general linear model of repeated measures followed

Table I
Baseline characteristics of the volunteers (n=18) at the start of the intervention study

	Baseline levels
Age (years)	44.9±10.3
BMI (kg/m²)	28.6±2.5
Weight (kg)	84.8±10.3
Waist circumference (cm)	100.3±7.1
Fat Body Mass (%)	29.2±4.0
Total cholesterol (mg/dL)	229.5±20.5
Triglycerides (mg/dL)	131.5±76.9
Glucose (mg/dL)	89.4±16.7
Blood pressure (mm Hg)	
Systolic	120.3±10.5
Diastolic	76.6±9.6
Alcohol consumption	7 never
	9 sometimes (1-2 drink/week)
	2 daily
Smoking	13 non-smokers
	5 smokers (10-20 cigarettes/day)

Values are mean ± SD of 18 volunteers. To transform mg/dL of cholesterol, triglycerides and glucose in mmol/L divide them by 38.68, 89, and 18, respectively.

by Least significant different (LSD) *post hoc* test were used to evaluate differences among the three periods, based on rate of change (final value - baseline value / baseline value). Significance was set at $p < 0.05$. Contingency tests such as the Chi-square test were employed to assess change prevalence in the contribution of nutrients to the RDA. Pearson correlations between the rate of change of selected anthropometrical parameters and those of dietary compounds were tested. All statistics were performed with the SPSS v.15.0 statistical package.

Results

As commented, 22 volunteers completed the study. However, the low number of participating women and the differences in response in weight and fat changes between male and female (Supplementary figures 1 and 2) encouraged us to study male volunteers' data. Baseline characteristics of volunteers are shown in table I. According to the BMI, 22% were obese while 33% exceeded the waist circumference cut-off points of 102 cm for men. A large proportion presented high basal levels of cholesterol (72% ≥ 220 mg/dL), triglycerides (22% ≥ 150 mg/dL) and blood pressure (11% ≥ 130 mm Hg of systolic and 33% ≥ 85 mm Hg diastolic blood pressure). Twenty eight percent of volunteers smoke between 10 and 20 cigarettes a day.

The composition of meat products included in the diet of the volunteers is shown in table II. Pâtés were richer than frankfurters in energy, fat, cholesterol, most minerals and vitamins. RF and n-3RF presented lower energy, fat and SFA, MUFA and PUFA than NF-products, with n-3RF pâtés/frankfurters registering a n-6/n-3 ratio 10-times lower than the other assayed meat products.

Table III shows the dietary intervention effect on anthropometric measures. Differences between periods for the rate of change were significant for body fat mass ($p = 0.018$) and waist/hip ratio ($p = 0.031$). Body fat mass significantly increased ($p < 0.05$) during the NF period and decreased in the RF period. The waist circumference ($p < 0.05$) and the waist/hip ratio ($p < 0.01$) were significantly decreased in the RF-period.

The energy and macronutrient contributions of the diets consumed during the three intervention phases are shown in table IV. At baseline for the RF period, proteins, carbohydrates, lipids, and alcohol contribution to the total energy (%En) was approximately 16.1%En, 40.2%En, 41%En, and 2.7%En, respecti-

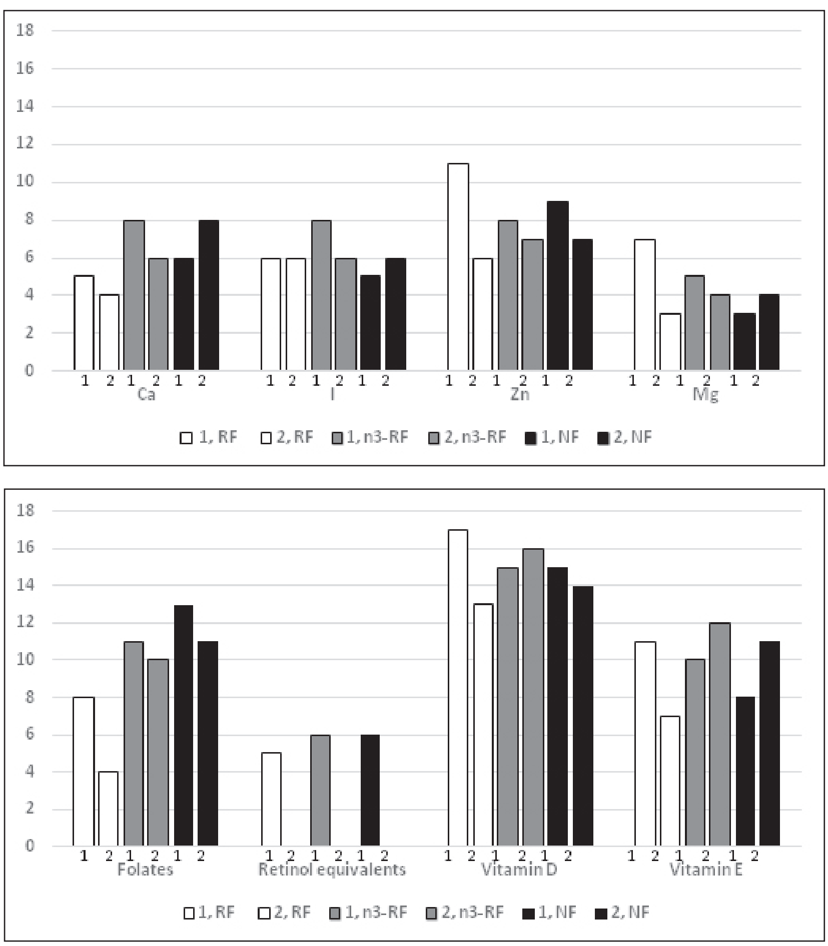


Fig. 1.—Effect of the three intervention phases on non-fulfilment of the recommended dietary allowances for some minerals (figure 1a) and vitamins (figure 1b). Bars show number of volunteers failing to provide 70% of RDA in each intervention and their respective baseline.
□ (1) Reduced-fat-meat-product (RF) baseline,
□ (2) Reduced-fat-meat-product (RF) intervention;
■ (1) n3-modified-reduced-fat-product (n-3RF) baseline,
■ (2) n3-modified-reduced-fat-product (n-3RF) intervention;
■ (1) Normal-fat-product (NF) baseline,
■ (2) Normal-fat-product (NF) intervention.

Table II
Composition of frankfurters and pâtés per 100 g wet matter

	Frankfurters			*Pâtés*		
	RF	*n-3 RF*	*NF*	*RF*	*n-3 RF*	*NF*
Macronutrients[1]						
Energy (kcal)	213	217	239	203	212	346
Protein (g)	17.9	19.4	18.3	13.3	14.2	13.2
Carbohydrates (g)	0	0.09	0	1.3	1.4	1.3
Dietary fiber (g)	0	0.31	0	0	0.29	0
Total fat (g)	15.3	15.1	18	15.2	15.5	30.8
SFA (g)	5.7	2.8	6.7	4.7	3.1	9.5
MUFA (g)	7.2	6.7	8.5	8.4	6.3	17.1
PUFA (g)	1.5	4.8	1.8	1.3	5.2	2.6
PUFA/SFA	0.3	1.71	0.27	0.28	1.68	0.27
[PUFA+MUFA]/SFA	1.5	4.11	1.54	2.06	3.71	2.07
n6/n3 PUFA	6.5	0.47	7.5	5.5	0.46	11
Cholesterol (mg)	49.8	41	51.4	138	129	147
Water (g)	66.8	65.2	63.7	69.2	64.8	50.7
Minerals[2]						
Ca (mg)	4.7	7.1	4.8	30.2	32.4	30.7
Fe (mg)	1.3	1.2	1.3	6.4	6.4	6.5
I (mg)	4.1	2.5	4.5	10.6	9	12.4
Mg (mg)	21.2	23.7	21.2	21	23.3	21.7
Zn(mg)	1.6	1.5	1.6	2.8	3.7	2.9
Se (mg)	9.1	9.2	9	21.5	21.6	21.7
Na (mg)	909	823	927	909	827	1004
K (mg)	198	211	199	218	231	232
P (mg)	116	117	116	192	194	199
Vitamins[2]						
Vitamin D (μg)	0	0	0	0.47	0.47	0.47
Vitamin E (mg)	0.017	0.75	0.02	0.22	0.89	0.24
Vitamin B_1 (mg)	0.54	0.54	0.54	0.28	0.27	0.29
Vitamin B_2 (mg)	0.14	0.14	0.14	1.1	1.1	1.1
Niacin Eq. (mg)	8.8	8.3	8.9	9.5	9	10.2
Vitamin B_6 (mg)	0.36	0.31	0.37	0.34	0.3	0.4
Folates (μg)	3.9	6.8	4	46.7	49.3	47.2
Vitamin B_{12} (μg)	2	2	2	13.5	13.5	13.5
Vitamin C (mg)	0	0.04	0	7.7	7.7	7.7
Retinol Eq. (μg)	0	0	0	11882	11882	11882

[1]Adapted from Delgado-Pando et al.[19,20]. [2]Calculated from DIAL data base[24].

vely. Rate of change in energy and in macronutrient did not significantly differ between periods except for n-3 (g/day and %En) (both $p < 0.001$), the n-6 PUFA/n-3 PUFA ratio ($p < 0.001$) and the carbohydrates/SFA ratio ($p = 0.023$). Rate of change displayed significant differences for PUFA (g/day) and the n-6 PUFA/n-3 PUFA ratio, lipids (%En), SFA (%En) and the carbohydrates/SFA ratio during the n-3RF period. Rate of change were significant for MUFA (g/day and %En) at the NF period.

The vitamin and mineral contents of the basal diets and the changes induced through the three dietary interventions are shown in table V. With the exception of vitamin B_5 and biotin and sodium, P, Cu and Ni

Table III
Basal anthropometrical measurements and rate of changes due to the intervention

	RF-baseline	RF	RF Rate of change	n-3RF-baseline	n-3RF	n-3RF Rate of change	NF-baseline	NF	NF Rate of change	P_r
Weight (kg)	84.8±10.3	84.6±10.3	0.15 (-0.76, 1.1)	84.3±9.9	84.4±10.2	0.11 (-0.82, 1.04)	84.5±10.5	84.9±10.5	0.44 (-0.24, 1.13)	0.490
Ideal Body Weight (%)	111.2±10.2	109.9±10.6	-0.39 (-1.03, 0.25)	109.5±10.4	109.7±10.9	0.11 (-0.82, 1.04)	109.9±10.9	110.2±10.6	0.44 (-0.24, 1.13)	0.326
BMI (kg/m²)	28.6±2.5	28.5±2.6	-0.39 (-1.03, 0.25)	28.4±2.7	28.5±2.7	0.39 (-0.53, 1.31)	28.5±2.7	28.6±2.6	0.56 (-0.23, 1.34)	0.211
Hip circumference (cm)	107.1±5.4	107.7±6.5	0.61 (-0.34, 1.56)[a]	108.0±4.9	107.2±5.2	-0.72 (-1.55, 0.11)[b]	106.6±5.4	107.2±5.6	0.61(-0.95, 2.18)[ab]	0.151
Waist circumference (cm)	100.3±7.1	99.4±7.2*	-0.94 (-1.73, -0.15)	98.9±6.2	99.0±6.1	0.11 (-1.09, 1.32)	99.1±6.5	99.0±6.4	-0.11 (-1.19, 0.97)	0.279
Waist/Hip ratio	0.94±0.04	0.92±0.04**	-1.56 (-2.46, -0.65)[a]	0.92±0.04	0.92±0.04	0.94 (-0.66, 2.55)[b]	0.93±0.03	0.92±0.04	-0.72 (-2.37, 0.93)[ab]	0.031
Conicity index	1.3±0.1	1.3±0.1	-0.78 (-1.57, 0.01)	1.3±0.04	1.3±0.04	0.17 (-1.21, 1.54)	1.3±0.05	1.3±0.04	-0.22 (-1.21, 0.77)	0.447
Body Fat Mass (%)	29.2±4.0	28.6±3.1*	-2.03 (-4.13, -0.09)[a]	28.5±4.1	28.1±4.0	-1.3 (-3.98, 1.31)[a]	28.2±4.0	28.8±3.7*	2.56 (0.37, 4.74)[b]	0.018

Mean ± SD; RF, reduced fat product; n-3RF, n-3 enriched reduced fat product; NF, normal fat product; +*p*<0.1, marginally significant; *$p < 0.050$, **$p < 0.010$, ***$p < 0.001$ with respect to its respective baseline; Rate of change (%), 100*(mean (CI 95%) of RF or n-3 RF or NF - baseline/baseline). Values in the same row bearing different letter were significantly different (repeated measures lineal general mode, $p < 0.05$) $_r$ significance for rate of change.

the mineral and vitamin changes observed were similar for the three periods. Some minerals and vitamin intakes as Cl ($p < 0.001$), Na and Cu (both at least $p < 0.05$), retinol equivalents ($p < 0.001$), vitamin B_{12} (at least $p < 0.01$) increased due to the inclusion of the meat-products in the three phases of the study.

The basal diets for the three periods failed to provide 70% of RDA for some minerals (Ca, I, Zn, Mg) and vitamins (folates, retinol equivalents, vitamin D, vitamin E) in many volunteers. The number of volunteers in this situation tended to decrease for some vitamins and minerals (marginally significantly $p < 0.1$) (Fig. 1a and 1b).

Basal data for HEI and the changes that occurred during the three intervention periods are shown in figure 2. There was no significant change in any of the three periods.

Rate of change in BMI and in ideal body weight percentage correlated inversely and significantly with rate of changes in the carbohydrate/SFA ratio for the n3RF period ($r = -0.658$; $p = 0.003$, and $r = -0.618$; $p = 0.006$, respectively)

Discussion

Present study, reports for the first time that the consumption of modified-RF exerts, in addition to the previous CVD risk improvement observed[11], modest benefits on some selected obesity markers, without affecting HEI, macronutrient and energy but their n-3PUFA and n-6/n-3 ratio intakes. The initial volunteers' characteristics clearly suggest that participants were at increased CVD risk. The anthropometrical changes observed, although modest, seem relevant as

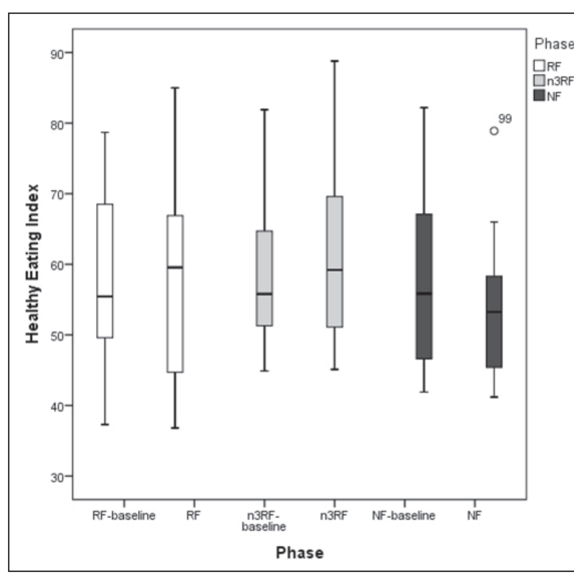


Fig. 2.—Change in the healthy eating index (HEI) of the 18 volunteers during the three intervention periods. □ Reduced-fat-meat-product (RF) diet; ▨ n3-modified-reduced-fat-product (n-3RF) diet; ■ Normal-fat-product (NF).

Table IV
Energy and macronutrients contents of each basal diet and the changes due to the interventions

Macronutrients	*RF-baseline*	*RF*	*RF Rate of change*	*n-3RF-baseline*	*n-3RF*	*n-3RF Rate of change*	*NF-baseline*	*NF*	*NF Rate of change*	p_r
Energy (kcal)	2393±754	2528±692	12.2 (-7.9, 32.3)	2379±686	2436±552	8.5 (-7.9, 25.0)	2684±748	2692±711	1.6 (-6.6, 9.7)	0.464
Carbohydrates (g)	234.7±107.4	255.2±89.9⁺	18.5 (-1.9, 38.9)	235.3±92.2	245.2±85.8	9.8 (-4.6, 24.2)	265.9±117.4	253.1±86.4	0.5 (-10.2, 11.1)	0.228
Proteins (g)	93.6±23.5	105.2±35.0	18.9 (-6.0, 43.8)	95.5±25.7	103.0±29.4	13.7 (-8.9, 36.2)	102.1±28.1	105.3±26.2	6.3 (-6.4, 19.0)	0.596
Cholesterol (mg)	339.5±104.1	402.4±207.8	23.2 (-6.7, 53.0)	365.6±146.9	397.8±123.1	27.4 (-8.2, 63.1)	388.1±124.9	408.3±117.1	9.3 (-5.4, 23.9)	0.531
Lipids (g)	107.7±39.8	108.9±39.7	11.4 (-19.8, 42.6)	105.9±31.4	103.3±24.7	4.8 (-15.4, 25.1)	115.6±34.5	125.0±39.1	10.8 (-1.4, 23.0)	0.854
SFA (g)	37.6±16.8	36.1±16.6	8.7 (-28.6, 46.0)	35.4±13.5	32.2±11.6	-3.7 (-21.1, 13.7)	38.2±13.4	40.6±16.5	8.7 (-4.6, 23.0)	0.602
MUFA (g)	47.4±18.6	49.1±17.9	20.6 (-17.6, 58.8)	47.2±11.2	46.9±11.2	11.7 (-16.7, 40.1)	52.8±16.8	58.5±18.7*	15.4 (-0.4, 31.1)	0.856
PUFA (g)	13.2±5.3*	14.3±5.2	18.3 (-7.7, 44.3)	13.8±4.9	15.5±3.7*	24.7 (1.0, 48.3)	15.1±6.0	15.2±5.4	7.3 (-12.8, 27.5)	0.403
n6 PUFA/n3 PUFA	7.9±1.9	7.3±2.5	-4.1 (-19.5, 11.3)[a]	7.2±2.6	3.2±0.7***	-49.4(-61.4,-37.5)[b]	8.0±3.3	8.3±3.3	14.4 (-14.8, 43.7)[a]	0.001
Alcohol (g)	10.0±13.9	8.1±10.0	-11.4 (-57.2, 34.4)	7.1±9.0	9.2±13.1	-18.6 (-72.0, 34.7)	18.4±22.4	12.8±13.6	2.4 (-61.1, 65.9)	0.802
Carbohydrates (%En)	40.2±8.4	42.9±8.5	8.9 (-1.9, 19.5)	41.1±7.8	42.7±10.0	3.7 (-3.4, 10.8)	40.6±9.8	39.4±8.1	-1.1 (-8.1, 5.9)	0.244
Proteins (%En)	16.1±3.3	16.5±2.6	4.6 (-3.7, 12.9)	16.4±2.8	16.7±2.6	4.4 (-5.5, 14.2)	15.3±1.9	15.9±3.2	4.7 (-6.2, 15.6)	0.999
Lipids (%En)	41.0±9.2	38.5±6.9	-3.2 (-13.7, 7.3)[ab]	40.6±6.5	38.1±8.1*	-6.3 (-12.5, -0.1)[a]	39.4±9.2	41.6±7.2	8.3 (-1.5, 18.2)[b]	0.078
SFA (%En)	14.1±3.1	12.6±3.3	-8.1 (-21.4, 5.2)[ab]	13.5±2.9	11.8±2.9**	-11.3 (-20.4, -2.2)[a]	12.8±2.9	13.3±2.3	6.5 (-4.3, 17.4)[b]	0.054
MUFA (%En)	18.3±5.8	17.6±4.0	4.8 (-14.4, 24.1)	18.2±4.3	17.9±4.9	0.1 (-12.2, 12.4)	18.2±5.5	19.8±4.9*	12.7 (-0.5, 26.0)	0.516
PUFA (%En)	5.1±1.6	5.1±1.2	7.5 (-9.3, 24.4)	5.4±1.6	5.9±1.5	16.3 (-1.1, 33.6)	5.2±1.8	5.1±1.4	5.1 (-13.6, 23.8)	0.516
Alcohol (%En)	2.7±3.7	2.1±2.6*	-34.6 (-64.1, -5.1)	2.0±2.6	2.5±3.3	-15.6 (-68.3, 37.2)	4.7±5.8	3.1±3.1	1.0 (-58.7, 60.6)	0.505
Carbohydrates/SFA[1]	6.5±2.6	7.9±3.4*	26.1 (3.5, 48.8)[a]	6.9±2.7	8.5±3.7**	23.6 (6.5, 40.7)[a]	7.5±3.4	6.5±2.4	-4.9 (-17.6, 7.8)[b]	0.023

Mean ± SD; RF, reduced fat product; n-3RF, n-3 enriched reduced fat product; NF, normal fat product; +p<0.1, marginally significant; *p<0.050, *p<0.010, ***p<0.001 with respect to its respective baseline; Rate of change (%), 100*(mean (CI 95%) of RF or n-3 RF or NF - baseline/baseline). Values in the same row bearing different letter were significantly different (repeated measures lineal general mode, p<0.05); $_r$ significance for rate of change; [1]based on g/g

Table V
Vitamin and mineral contents of each basal diet and the changes due to the interventions

	RF-baseline	*RF*	*RF Rate of change*	*n-3RF-baseline*	*n-3RF*	*n-3RF Rate of change*	*NF-baseline*	*NF*	*NF Rate of change*	p_r
Retinol Eq. (μg)	1046±640	5888±2902***	694 (359, 1030)	1045±652	5054±384***	609 (346, 872)	911±346	5105±577***	541 (418, 665)	0.589
Vitamin C (mg)	99.7±55.3	146.1±90.9*	57.2 (13.8,100.5)	141.1±105.4	136.3±90.7	47.3 (-29.5, 124)	120.4±61.9	109.6±57.6	5.7 (-23.0, 34.4)	0.260
Vitamin B_{12}(μg)	5.8±2.6	14.1±11.9**	176.4 (72.6, 280.2)	5.8±2.4	11.4±2.6**	152.3 (72.6, 280.2)	6.6±3.1	11.1±3.0***	94.8 (56.1, 133.5)	0.328
Niacin Eq. (mg)	37.9±8.1	48.7±15.9*	36.5 (1.4, 71.7)	37.4±10.3	46.0±13.5*	31.5 (2.0, 60.9)	41.8±11.2	46.3±13.4	16.0 (-4.7, 36.7)	0.525
Folates (μg)	302.7±163.9	358.1±142.8*	37.2 (4.5, 70.0)^{a}	322.9±197.9	302.1±128.9	15.2 (-14.4, 44.7)^{ab}	259.3±69.0	261.6±79.3	2.3 (-9.8, 14.4)^{b}	0.186
Vitamin B_1 (mg)	1.5±0.4	1.9±0.7	35.6 (-6.5, 77.8)	1.5±0.6	1.6±0.6	19.5 (-7.5, 46.4)	1.4±0.4	1.8±0.7*	33.1 (3.8, 62.4)	0.717
Riboflavin (mg)	1.9±0.6	2.7±1.0**	41.8 (8.8, 74.8)	2.0±0.8	2.1±0.6	13.5 (-9.2, 36.1)	2.0±0.9	2.3±0.7*	20.0 (6.6, 33.3)	0.180
Vitamin B_6 (mg)	2.1±0.5	2.6±0.9	32.3 (-8.3, 72.9)	2.2±0.7	2.2±0.7	18.8 (-23.2, 60.8)	2.3±0.5	2.3±0.7	5.1 (-17.8, 28.0)	0.489
Vitamin D (mg)	1.8±1.1	3.9±3.7**	168.1 (52.4, 283.8)	3.5±6.2	2.3±1.4	58.6 (-27.5, 144.7)	2.2±1.8	4.1±5.5	-132.3(-286, 21.1)	0.388
Vitamin E_*	7.9±2.9	9.3±4.5	22.0 (-5.8, 49.8)	9.2±5.3	8.2±3.4	10.0 (-22.0, 42.1)	8.9±3.7	8.8±5.5	1.8 (-22.4, 26.1)	0.313
Vitamin K (μg)	185.7±183.4	177.7±85.4	42.7 (-15.1, 100.6)	144.9±84.1	125.3±52.1	14.8 (-28.1, 57.6)	113.1±50.3	141.8±52.2**	40.3 (9.1, 71.4)	0.547
Vitamin B_5 (mg)	5.5±1.6	7.8±2.6**	47.9 (22.4, 73.5)^{a}	6.0±2.2	6.7±1.8	20.7 (-3.0, 44.5)^{b}	6.2±1.9	6.8±1.7*	13.8 (0.7, 26.8)^{b}	0.015
Biotin (μg)	26.7±11.5	40.7±20.6**	75.9 (20.5, 131.2)^{a}	33.2±16.4	32.9±11.5	16.3 (-12.6, 45.2)^{b}	36.6±19.8	32.8±12.4	-1.1 (-17.4, 15.2)^{b}	0.006
Ca (mg)	978.7±432.2	1090.5±438.0	32.3 (-22.0, 86.5)	961.6±463.0	849.7±363.4	-2.8 (-20.8, 15.3)	1027±588.2	943.2±482.2	-4.9 (-18.6, 8.6)	0.156
Fe (mg)	15.6±5.8	21.1±8.7*	43.5 (10.3, 76.6)	16,4±6.8	17.4±5.2	17.2 (-3.7, 38.1)	15.1±4.4	17.4±7.1	16.8 (4.3, 29.4)*	0.163
I (μg)	115.3±60.3	132.7±52.8*	31.5 (0.6, 62.4)	105.8±49.2	109.3±40.2	18.5 (-9.1, 45.9)	123.0±49.8	115.1±38.8	4.6 (-18.8, 28.0)	0.269
Zn (mg)	10.3±3.4	12.7±5.2*	35.2 (-1.2, 71.5)	11.2±3.3	11.2±3.4	10.0 (-16.4, 36.4)	10.9±3.7	11.7±3.5	12.5 (-3.5, 28.5)	0.287
Na (mg)	2343±749	4024±1672***	79.7 (42.4, 117.1)^{a}	2699±982	3068±715*	25.8 (3.6, 47.9)^{b}	2970±1237	3440±930**	26.1 (7.4, 44.7)^{b}	0.006
K (mg)	3155±1074	3590±1256	21,7 (-4.8, 48.2)	3290±1344	3173±887	8.2 (-15.5, 31.8)	3298±1089	3134±941	-3.0 (-13.5, 7-5)	0.149
Se (μg)	121.4±50.5	159.6±50.2*	46.6 (11.2, 81.9)	119.4±42.8	140.1±37.5*	26.1 (4.8, 47.4)	133.6±58.5	142.6±43.8	21.8 (-8.4, 51.9)	0.412
P (mg)	1506.0±472.4	1789.3±565.0	28.7 (-1.5, 58.8)^{a}	1571.3±533.4	1546.1±470.3	5.7 (-17.4, 28.7)^{b}	1734.7±603.3	1627.8±485.9	-2.7 (-14.3, 8.8)^{b}	0.028
Mg (mg)	319.9±124.6	370.6±127.3	26.4 (-3.9, 56.7)^{a}	332.9±154.7	319.4±97.0	8.7 (-12.2, 29.5)^{ab}	334.2±122.6	316.9±101.6	-2.6 (-12.4, 7.3)^{b}	0.064
Cu (mg)	1.5±0.6	2.3±1.2**	67.9 (22.7, 113.1)^{a}	1.7±0.8	1.9±0.7*	30.9 (-0.7, 62.6)^{ab}	1.6±0.6	1.7±0.6*	15.5 (0.04, 30.9)^{b}	0.041
Cr (μg)	39.9±17.4	53.7±34.1**	41.1 (11.9, 70.4)	53.2±36.9	50.2±25.0	36.4 (-29.9, 102.6)	56.8±51.8	48.0±22.0	4.9 (-18.2, 27.9)	0.401
Ni (μg)	106.1±72.3	151.3±122.4*	63.8 (13.4, 114.2)^{a}	164.0±194.9	112.2±111.5	14.9 (-23.4, 53.1)^{b}	110.1±111.1	99.0±49.6	12.5 (-7.1, 32.1)^{ab}	0.044
Cl (mg)	2445.6±721.4	3787±891***	64.3 (37.7, 90.9)	2598±927	3412±806***	40.2 (21.8, 58.5)	2537±628	3783±897***	55.2 (34.2, 76.2)	0.277
F (μg)	477.1±354.5	633.4±333.5**	71.2 (19.3, 123.1)^{a}	549.1±335.3	594.0±325.8	36.7 (-9.3, 82.8)^{ab}	607.4±408.8	607.2±340.9	13.8 (-8.0, 35.7)^{b}	0.130
Mn (mg)	2.7±1.1	3.6±1.7	61.2 (-8.2, 130.6)	2.8±1.3	3.2±1.9	26.7 (-4.0, 57.5)	5.3±9.2	3.4±2.8	13.4 (-16.4, 43.1)	0.277

Mean ± SD; RF, reduced fat product; n-3RF, n-3 enriched reduced fat product; NF, normal fat product; +$p<0.1$, marginally significant; *$p<0.050$, *$p<0.010$, ***$p<0.001$ with respect to its respective baseline; Rate of change (%), 100*(mean (CI 95%) of RF or n-3 RF or NF - baseline/baseline). Values in the same row bearing different letter were significantly different (repeated measures lineal general mode, $p<0.05$) $_r$ significance for rate of change reduction in body fat and waist circumference or the waist/hip ratio have been found to decrease both CVD and Metabolic Syndrome risks[29].

The energy intake at baseline for the RF-period was balanced with the theoretical energy expenditure in only 47.6% of the volunteers, which consumed less than 80% of the theoretical energy expenditure. Diet at baseline for the RF-period were similar to those observed in other Spanish studies, with a low energy contribution from carbohydrates, a high to very high contribution from fat, and a moderately high contribution from proteins[30,31]. Diets were also rich in SFA, exceeding the recommended level of 10%En[30,31], or the 7%En suggested as a secondary outcome for prevention of some chronic diseases[13]. These diet results explain, at least partially, the high cholesterol, triglyceride and LDL cholesterol levels observed in these volunteers[11], as diets rich in SFA are known to increase serum total and LDL cholesterol[13,32] and to contribute to lipogenesis, insulin resistance, blood pressure and other components of the Metabolic syndrome[33,34].

The initial dietary n-6/n-3 ratio of volunteers was higher than the ideal 4 suggested[13], but according to the WHO[35] it is not necessary to consider this ratio when n-6 and n-3PUFA contributions are within nutritional targets. Fifty-six per cent of volunteers consumed more than the recommended range of 10-15%En for proteins, 77.8% exceeded the recommended intake for lipids (20-35%En); 88.9% consumed less than the recommended carbohydrate intake (50-60%En); and 5.6% consumed more than 10%En from alcohol.

Diets followed during the three intervention periods were similar. Thus, volunteers seem to have maintained their habitual diets as regards macronutrients, one of the aspects addressed in the study (Table IV). As expected, during the n-3RF dietary period the n-3PUFA content increased while the n-6/n-3 ratio decreased with respect to RF and NF periods. Again, these dietary modifications support the lipoprotein profile improvement previously observed in these volunteers during the n3RF-period[11] while during the RF-intervention the diet changed slightly, explaining the absence of effects with respect to baseline.

The contribution of minerals and vitamins in the RF basal diet had the same limitations as other studies[36,37], because it failed to provide 70% of the RDA for some minerals (Fig. 1a) and vitamins (Fig. 1b) in many volunteers, suggesting the need for nutritional advice and/or intervention. Some mineral and vitamin intakes increased due to the inclusion of the meat-products in the three phases of the study. As indicated, the pâtés contained liver, which is known to contain high concentrations of some vitamins (e.g. vitamin A and vitamin B_{12}) and minerals (e.g. Fe)[38], thus explaining the observed increases. Due to the similarity of composition of the products tested, changes observed in vitamins and minerals were similar for the three periods. The reasons for sodium and folate changes are not clear given the similarity of food intake between phases and the standard content of the assayed meat-products.

The number of volunteers who did not cover 70% of the RDA for some vitamins (e.g. retinol equivalents and vitamin D) and minerals (e.g. Zn) tended to decrease (Figs. 1a and 1b). This situation must be related to the high concentrations of these micronutrients in the meat-derivatives tested. However, in most cases the dietary changes observed in those micronutrients were not relevant but similar in all three periods.

The dietary similarity in energy, fiber, macro and most micronutrients between periods explain the absence of significant changes in diet HEI during the study. As noted, HEI gives information on global diet quality based on food group consumption and variety and some quality aspects of diet such as SFA, fat and sodium[25]. As in other studies[28,39,40], a high percentage of individuals followed unacceptable diets (HEI < 70). This percentage was not significantly affected in the course of the present study (Fig. 2).

In comparison to SFA, PUFA have been found to decrease lipogenesis and increase lipolysis[13]. Furthermore, diets enriched in n-3PUFA *vs.* n-6PUFA induced modest decrease in fat mass[14]. These facts explain the correlations found between differences in rate of change for BMI and ideal-body-weight with that of body fat mass. As the carbohydrate/SFA ratio[17,39] and the body fat mass[41] have been related to insulin sensitivity, it can be speculated that the changes observed in the carbohydrate/SFA ratio and body fat mass would also affect insulin resistance. This hypothesis needs further confirmation and must be tested in future studies.

Conclusions

Modified meat products decrease body fat mass. With the exception of the amount and energy contribution of n-3PUFA, the n-6/n-3 ratio and a few minerals and some vitamins, the inclusion of modified meat-products with improved-fat content and composition in the volunteer diets, did not significantly modify their eating habits and the variety and quality of the diet. Thus, present pâtés and frankfurters act as functional food, adding benefits to the ones already observed in the lipoprotein profiles of same volunteers during the intervention study[11]. Furthermore, taking into account present results, it will be able for us to associate, in future studies, possible changes in the biochemical profiles of volunteers only with the nutritional input from the meat-products consumed.

Acknowledgements

This research was supported by Consolider-Ingenio 2010: CARNISENUSA (CSD2007-00016), and AGL 2011-29644-C02-01 y AGL 2011-29644-C02-02 projects. We acknowledge volunteers participation and

the statistical study assessment of Dr Laura Barrios from the Subdirección General de Apoyo a la Investigación (SGAI) of CSIC.

The authors have declared no conflict of interest.

References

1. Olmedilla-Alonso B, Jiménez-Colmenero F, Sánchez-Muniz FJ. Development and assessment of healthy properties of meat and meat products designed as functional foods. *Meat Sci*. 2013; 95: 919-30.
2. Varela-Moreiras G, Ruiz E, Valero T, Ávila JM, Del Pozo S. The Spanish diet: an update. *Nutr Hosp*. 2013; 28 suppl. 5: 13-20.
3. Sinha R, Cross AJ, Graubard BI, Leitzmann MF, Schatzkin A. Meat intake and mortality. A prospective study of over half a million people. *Arch Intern Med*. 2009; 169: 562-71.
4. Astrup A, Dyerberg J, Elwood P et al. The role of reducing intakes of saturated fat in the prevention of cardiovascular disease: where does the evidence stand in 2010? *Am J Clin Nutr.* 2011; 93: 684-8.
5. Ruxton CHS, Derbyshire E, Pickard RS. Micronutrient challenges across the age spectrum: Is there a role for red meat? *Nutr Bull*. 2012; 38: 178-90.
6. Jiménez-Colmenero F. Potential applications of multiple emulsions in the development of healthy and functional foods. *Food Res Int*. 2013; 52: 64-74.
7. Diplock AT, Agget PJ, Ashwell M et al. Scientific concepts of functional foods in Europe: Consensus document. *Br J Nutr.* 1999; 81 (Suppl. 1): S1-S27.
8. Bhupathiraju SN, Tucker KL. Coronary heart disease prevention: Nutrients, foods, and dietary patterns. *Clin Chim Acta*. 2011; 412 (17-18): 1493-514.
9. Jakobsen MU, O'Reilly EJ, Heitmann BL et al. Major types of dietary fat and risk of coronary heart disease: a pooled analysis of 11 cohort studies. *Am J Clin Nutr.* 2009; 89: 1425-32.
10. Piscopo S. The Mediterranean diet as a nutrition education, health promotion and disease prevention tool. *Public Health Nutr.* 2009; 12: 1648-55.
11. Delgado-Pando G, Celada P, Sánchez-Muniz FJ, Jiménez-Colmenero F, Olmedilla-Alonso B. Effects of improved fat content of frankfurters and pâtés on lipid and lipoprotein profile of volunteers at increased cardiovascular risk: a placebo-controlled study. *Eur J Nutr.* 2014; 53: 83-93.
12. Serra-Majem L, Calder PC, Uauy R. Systematic reviews of the role of omega-3 fatty acids in the prevention and treatment of disease. *Br J Nutr.* 2012; 107 Suppl 2: S1-S284.
13. Sánchez-Muniz FJ, Bastida. Lípidos. In: Varela Moreiras G, coordinator. Libro blanco de la Nutrición en España. Fundación Española de la Nutrición; pp. 113-24, Madrid, Spain, 2013.
14. Hill AM, Buckey JD, Murphy KJ, Howe PRC. Combining fish-oil supplements with regular aerobic exercise improves body composition and cardiovascular disease risk factors. *Am J Clin Nutr.* 2007; 85: 1267-74.
15. Smith CE, Arnett DK, Corella D et al. Perilipin polymorphism interacts with saturated fat and carbohydrates to modulate insulin resistance. *Nutr Metab Cardiovasc Dis*. 2012; 22: 449-55.
16. Martín de Santa Olalla L, Sánchez-Muniz FJ, Vaquero MP. n3 fatty acids in glucose metabolism and insulin sensitivity. *Nutr Hosp*. 2009; 24: 113-27.
17. Ruperto M, Barril G, Sánchez-Muniz FJ. Conicity index as a contributor marker of inflammation in haemodialysis patients. *Nutr Hosp*. 2013; 28: 1688-95.
18. Leblanc V, Bégin C, Hudon AM, Royer MM, Corneau L, Dodin S, Lemieux S. Gender differences in the long-term effects of a nutritional intervention program promoting the Mediterranean diet: changes in dietary intakes, eating behaviors, anthropometric and metabolic variables. *Nutr J*. 2014;13: 107.
19. Delgado-Pando G, Cofrades S, Ruiz-Capillas C, Jiménez-Colmenero F. Healthier lipid combination as functional ingredient influencing sensory and technological properties of low-fat frankfurters. *Eur J Lipid Sci Technol*. 2010; 112: 859-70.
20. Delgado-Pando G, Cofrades S, Rodríguez-Salas L, Jiménez-Colmenero F. A healthier oil combination and konjac gel as functional ingredients in low-fat pork liver pâté. *Meat Sci*. 2011; 88: 241-8.
21. Becker CC, Lund P, Holmer G, Jensen H, Sandstrom B. Effects of butter oil blends with increased concentrations of stearic, oleic and linolenic acid on blood lipids in young adults. *Eur J Clin Nutr.* 1999; 53: 535-41.
22. Valdez R. A simple model-based index of abdominal adiposity. *J Clin Epidemiol*. 1991; 44: 955-6.
23. Alastrué-Vidal A, Sitges-Serra A, Jaurrieta-Mas E, Sitges-Creus A. Valoración de los parámetros antropométricos en nuestra población. *Med Clin. (Barc)* 1982; 78: 407-15.
24. Ortega RM, López-Sobaler AM, Andrés P, Requejo AM, Aparicio A, Molinero LM. DIAL software for assessing diets and food calculations. Departamento de Nutrición (UCM) and Alce Ingeniería SA Madrid. 2004; (http://www.alceingenieria.net/nutrición.htm, accessed February 2014).
25. Kennedy ET, Ohls J, Carlson S, Fleming K. The Healthy Eating Index: design and applications. *J Am Diet Assoc*. 1995; 95: 1103-8.
26. Ortega RM, Requejo AM, Andrés P et al. El rombo de la alimentación. Guía útil en la planificación de dietas ajustadas a las pautas recomendadas. *Nutr Clin Diet Hosp*. 1998; 16: 35-43.
27. SENC, Sociedad Española de Nutrición Comunitaria in Guía de la alimentación saludable. Editorial Evergráficas, SL.; León, Spain, 2005,
28. Norte-Navarro AI, Ortiz-Moncada R. Spanish diet quality according to the healthy eating index. *Nutr Hosp*. 2011; 26: 330-6.
29. Martínez-Victoria E, Yago MD. Omega 3 polyunsaturated fatty acids and body weight. *Br J Nutr.* 2012; 107 suppl 2: S107-S116.
30. AESAN ENIDE: Encuesta Nacional de Ingesta Dietética (2009-2010). Evaluación nutricional de la dieta española. I. Energía y macronutrientes, 2011, http://aesan.msssi.gob.es/AESAN/web/notas_prensa/presentacion_enide.shtml
31. Gómez-Gerique JA, Gutiérrez-Fuentes JA, Montoya MT et al. Lipid profile of the Spanish population: the DRECE (diet and risk of cardiovascular disease in Spain) study. DRECE study group. *Med Clin*. 1999; 113: 730-5.
32. Sánchez-Muniz FJ, Bocanegra A, Bastida S, Benedí J. Algae and cardiovascular health. In: Domínguez H. editor. Functional ingredients from algae for foods and nutraceuticals, Woodhead Publishing; pp. 369-415, Oxford, UK, 2013.
33. Gesteiro E, Bastida S, Sánchez-Muniz FJ. Cord-blood lipoproteins, homocysteine, insulin sensitivity/resistance marker profile, and concurrence of dysglycaemia and dyslipaemia in full-term neonates of the Mérida Study. *Eur J Pediatr.* 2013; 172: 883-94.
34. Sánchez-Muniz FJ, Gesteiro E, Espárrago-Rodilla M et al. Maternal nutrition during pregnancy conditions the fetal pancreas development, hormonal status and diabetes mellitus and metabolic syndrome biomarkers at birth. *Nutr Hosp*. 2013; 28: 250-74.
35. FAO/WHO In The joint FAO/WHO expert consultation on fats and fatty acids in human nutrition. FAO food and nutrition paper 91. Food and Agriculture Organisation of the United Nations; ISSN 0254-4725, Rome, Italy, 2010.
36. Vaquero-Rodrigo MP, Navarro-Martos MP. Minerales. In Varela Moreiras G, coordinator. Libro blanco de la Nutrición en España. Fundación Española de la Nutrición; pp. 157-63, Madrid, Spain, 2013.
37. Olmedilla-Alonso B, Granado-Lorencio F. Vitaminas. In: Varela Moreiras G, coordinator, Libro blanco de la Nutrición

en España. Fundación Española de la Nutrición; pp. 145-55, Madrid, Spain, 2013.
38. Sociedad Española de la Nutrición. Departamento de Nutrición, Ingestas recomendadas de energía y nutrientes para la población española (revisadas in 2008) In: Moreiras O, Carbajal A, Cabrera L and Cuadrado C, editors. Tablas de composición de alimentos. Pirámide. Madrid, Spain, 2008.
39. Gesteiro E, Rodriguez-Bernal B, Bastida S, Sánchez-Muniz FJ. Maternal diets with low healthy eating index or mediterranean diet adherence scores are associated with high cord-blood insulin levels and insulin resistance markers at birth. *Eur J Clin Nutr.* 2012; 66: 1008-15.
40. Martínez-Sesmero JM, Riesgo cardiovascular en el estudio Área de Toledo. Ph.D. Thesis, Universidad Complutense de Madrid, Madrid, Spain, 2013.
41. Riccardi G, Aggett P, Brighenti F et al. Body weight regulation, insulin sensitivity and diabetes risk. *Eur J Nutr.* 2004; 43 Suppl 2: II7-II46.