Insulin Resistance, Inflammation, and Serum Fatty Acid Composition

José-Manuel Fernández-Real, md, phd[1]
Montserrat Broch, phd[2]
Joan Vendrell, md, phd[2]
Wifredo Ricart, md[1]

OBJECTIVE — Fatty acids (FAs) have been involved in the development of chronic inflammatory conditions such as insulin resistance and obesity. However, the relation among insulin resistance, obesity, inflammatory activity (circulating interleukin [IL]-6) and dietary FAs has been scarcely studied in otherwise healthy subjects.

RESEARCH DESIGN AND METHODS — We aimed to study these interactions in 123 overweight (BMI 26.9 ± 2.4 kg/m^2 [means ± SD]) subjects and 109 lean (BMI 21.7 ± 1.7 kg/m^2, $P < 0.000001$) subjects. IL-6 was measured by immunoassay and FA by gas liquid cromatography.

RESULTS — The percentage of saturated FAs ($r = 0.30$, $P = 0.01$) and ω-6 FAs ($r = -0.32$, $P = 0.001$) were significantly associated with circulating IL-6, whereas the percentage of ω-3 FAs correlated negatively with C-reactive protein in overweight subjects ($P = 0.04$). Saturated–to–ω-3 and saturated–to–ω-6 FA ratios were significantly and positively associated with C-reactive protein ($P < 0.0001$) and IL-6 ($P < 0.001$), respectively. In contrast, none of these associations reached statistical significance in lean subjects. Those subjects in the most insulin-sensitive quintile (homeostasis model assessment value) showed a significantly higher percentage of linoleic acid (C18:2 ϖ6) ($P = 0.03$) and a significantly lower level of araquidic (C20:0) ($P = 0.04$), behenic (C22:0) ($P = 0.009$), lignoceric (C24:0) ($P = 0.02$), and nervonic (C24:1 ϖ9) ($P = 0.001$) FAs than the remaining subjects. In parallel, the most insulin-sensitive subjects showed significantly decreased C-reactive protein ($P = 0.03$). Serum C-reactive protein was significantly associated with percent linoleic acid and eicosapentaenoic acid in nonsmoking men ($P = 0.03$ and $P = 0.04$, respectively) and with docosahexaenoic acid in nonsmoking women ($r = -0.46$, $P < 0.0001$). We constructed a multivariant regression analysis to predict circulating IL-6. Age, BMI, waist-to-hip ratio (WHR), smoking status, and the relation of saturated to ω-6 or saturated to ω-3 FAs were considered as independent variables separately in men and women. In overweight men, the ratio of saturated to ω-3 FAs ($P = 0.01$), but not age, sex, BMI, WHR, or smoking status, independently contributed to 17% of IL-6 variance. In lean men, smoking status ($P = 0.02$), but not the remaining variables, contributed to 8% of IL-6 variance.

CONCLUSIONS — Dietary FAs (as inferred from plasma FA concentration) seem to be linked to inflammatory activity in overweight subjects and in subjects with insulin resistance. Being overweight modulates the relation of FAs to inflammatory markers.

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Insulin resistance is increasingly viewed as a chronic inflammatory disease (1). Different cytokines and chemical messengers constitute the main regulators of this inflammatory process, achieving biological effects individually as well as in association with each other. Interleukin (IL)-6 is a proinflammatory cytokine produced by many different cell types, including immune cells and adipose tissue (2,3). The major effects of IL-6 take place at sites distinct from its origin and are consequent upon its circulating concentrations. IL-6 seems to play a role in general lipid metabolism. For instance, IL-6 inhibits adipocyte lipoprotein lipase activity (4) and induces increases in hepatic triglyceride secretion in rats (5). In humans, the action of IL-6 is associated with increased plasma free fatty acids (FFAs) (6), fasting triglycerides, and VLDL triglycerides, and post–glucose load FFAs are also linked to IL-6 (7). These associations seem important because elevated IL-6 levels and C-reactive protein, a surrogate marker for IL-6 activity, predict the development of type 2 diabetes and mortality (8–10).

The amount and quality of fat in the diet could be of importance for the development of insulin resistance and related inflammatory activity. A high proportion of long-chain unsaturated fatty acids (FAs) and a low proportion of saturated FAs in the diet have been associated with improved insulin action (11). Highly unsaturated FAs, and n-3 FAs in particular, are receiving increasing attention as potential anti-inflammatory agents. It is well known that dietary FAs appear to modulate the release of different cytokines (12). The production of IL-6 and other cytokines by peripheral mononuclear cells was significantly decreased after dietary polyunsaturated FA supplementation (13,14). Serum FA concentration reflects to some extent the composition of dietary fat (15). No information is available regarding the possible links between serum FA profile and circulating IL-6 in a large sample, so we aimed to study this association in 232 healthy subjects.

RESEARCH DESIGN AND METHODS

Subjects

A total of 232 subjects were evaluated as part of an ongoing epidemiological study dealing with nonclassical cardiovascular risk factors. The study was approved by the hospital ethics committee, and in-

From the [1]Unitat de Diabetologia, Endocrinologia i Nutrició, University Hospital of Girona "Dr. Josep Trueta," Girona, Spain; and the [2]University Hospital of Tarragona "Joan XXIII," Tarragona, Spain.

Address correspondence and reprint requests to J.M. Fernandez-Real, MD, PhD, Unitat d'Endocrinologia, Diabetes i Nutrició, Hospital de Girona, Carretera de Francia s/n, 17007 Girona, Spain. E-mail: uden.jmfernandezreal@htrueta.scs.es.

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Abbreviations: DHA, docosahexaenoic; EPA, eicosapentaenoic; FA, fatty acid; HOMA, homeostasis model assessment; IL, interleukin; WHR, waist-to-hip ratio.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

formed consent was obtained from each subject.

Anthropometric and clinical measurements. BMI and waist-to-hip ratio (WHR) were measured in all subjects. BMI was calculated as weight (in kilograms) divided by height (in meters) squared, and each subject's waist was measured with a soft tape midway between the lowest rib and the iliac crest. The hip circumference was measured at the widest part of the gluteal region.

Smokers were defined as any person consuming at least one cigarette a day in the previous 6 months. We had included a small number of smoking women (n = 16), so we decided to exclude them from this analysis. All subjects reported that their body weight had been stable for at least 3 months before the study. Resting blood pressure was measured after subjects had been in a sitting position for a minimum of 15 min. Using a mercury sphygmomanometer, blood pressure was read three times on the right arm by the same investigator. The mean of three measurements was used for this study. None of the subjects was taking any medication (including glucocorticoids or estrogens) or had any evidence of metabolic disease other than obesity. Subjects with impaired fasting glucose were specifically excluded. All women were premenopausal and studied in their follicular phase. Liver disease and thyroid dysfunction were specifically excluded by biochemical workup. All women had regular menstrual cycles.

Table 1—*Anthropometric and biochemical variables in men*

	Lean	Overweight	*P*
n	64	70	—
Age (years)	37.9 ± 12	43.7 ± 10.1	0.004
Weight (kg)	67.5 ± 7.9	82.7 ± 8.6	<0.0001
BMI (kg/m^2)	22.2 ± 1.8	27.9 ± 2.4	<0.0001
WHR	0.94 ± 0.05	0.99 ± 0.02	<0.0001
Smokers (yes/no)	24/40	26/44	NS
Systolic blood pressure (mmHg)	119.3 ± 11.1	130 ± 12	<0.0001
Diastolic blood pressure (mmHg)	67.4 ± 7	76.8 ± 9.9	<0.0001
Fasting glucose (mmol/l)	4.8 ± 0.5	5.1 ± 0.3	0.02
Fasting insulin (mU/l)	6.3 ± 2.5	8.8 ± 4.1	<0.0001
HOMA value	1.22 ± 0.5	1.83 ± 0.9	<0.0001
Cholesterol (mg/dl)	215 ± 45	226 ± 46	NS
Triglycerides (mg/dl)	97 ± 45	141 ± 87	0.001
IL-6 (pg/ml)*	4.4–11.9	5.0–17.1	0.06
C-reactive protein (mg/dl)	0.35 ± 0.17	0.44 ± 0.2	0.03

Data are n, means ± SD, and interquartile range.

Serum samples

Analysis of serum FAs. Following the method by Lepage and Roy (16), 100 μl serum obtained after a 12-h fast was precisely weighted in glass tubes and diluted with methanol-benzene 4:1 (vol/vol). Acetyl chloride (200 μl) was slowly added over a period of 1 min. After transesterification, the pooled solvent extracts were dried under a gentle stream of nitrogen at room temperature. Residues were dissolved in 500 μl hexane, and an aliquot was injected into the chromatograph. FAs were chromatographed as methyl esters on a 30-m fused silica column with an internal diameter of 0.25 mm. Analysis was performed on a Hewlett-Packard 5890 gas chromatograph equipped with a flame ionization detector. The column temperature was held at 80°C for 3 min and in a stepwise fashion reached a plateau of 220°C. The injection port and detector temperature were 250°C and 270°C, respectively. Helium was used as carrier gas. An internal standard consisting of 50 μg pentadecanoic acid (C15:0) was precisely weighed and added to the serum.

Serum IL-6 concentration was measured using a commercial immunoassay (MEDGENIX IL-6 EASIA; BioSource Europe SA, Zoning Industriel B-6220, Fleunes, Belgium), with coefficients of variation <6%. The minimum detectable concentration of IL-6 was 0.094 pg/ml. Serum C-reactive protein (Beckman, Fullerton, CA) was determined by routine laboratory tests with intra- and interassay CVs <4%.

The serum glucose concentration was measured in duplicate by the glucose oxidase method. The serum insulin level was measured in duplicate by monoclonal immunoradiometric assay (IRMA; Medgenix Diagnostics, Fleunes, Belgium). The lowest limit of detection was 4.0 mU/l. The intraassay CV was 5.2% at a concentration of 10 mU/l. The interassay CV was 6.9% at 14 mU/l. The fasting insulin resistance index (homeostasis model assessment [HOMA]) was calculated with the formula: HOMA = fasting glucose (mmol/l) × fasting insulin (mU/l)/22.5. In our experience, HOMA fairly correlates with the insulin sensitivity index calculated using the minimal model approach (r = 0.79, P < 0.0001 [17]).

Total serum cholesterol was measured by the reaction of cholesterol esterase/cholesterol oxidase/peroxidase. Total serum triglycerides were measured through the reaction of glycerol-phosphate-oxidase and peroxidase.

Statistical analysis

Descriptive results of continuous variables are expressed as the mean ± SD. Before statistical analysis, normal distribution and homogeneity of the variances were tested. Parameters that did not fulfill these tests (individual FAs and ratios and IL-6) were log transformed. The relations between variables were analyzed by simple correlation analysis. Subjects were divided into quintiles of HOMA value. Comparison of variables in subjects of the highest versus the lowest insulin sensitivity quintiles was performed using Student's t test and by stepwise multivariate linear regression analysis. Smoking status was considered as a categorical variable (0 = nonsmoker, 1 = smoker). Levels of statistical significance were set at P < 0.05.

RESULTS — The main characteristics and serum FA composition of the study subjects are shown in Tables 1–3. The absolute FA concentration did not differ significantly between men and women or between overweight and lean subjects. Percent palmitic acid (19.8 ± 2 vs. 18.8 ± 3, P = 0.004) and percent docosahexaenoic (DHA) acid (2.15 ± 0.6 vs. 1.92 ± 0.6, P = 0.03) were significantly higher in men than in women.

We divided this population into over-

Table 2—*Serum fatty acid profile in men and women*

Total concentration (μmol/l)	Overweight	Lean	*P*	Relative concentration (%)	Overweight	Lean	*P*
Men				Men			
14:0	70.72 ± 61	58.08 ± 37.79	NS	14:0	0.34 ± 0.22	0.45 ± 0.26	0.01
16:0	2,853 ± 825	2,837 ± 954	NS	16:0	19.63 ± 1.88	20.13 ± 2.20	NS
16:1 (ω-9)	54.46 ± 23.62	52.33 ± 23.80	NS	16:1 (ω-9)	0.34 ± 0.09	0.37 ± 0.11	NS
18:0	1,000 ± 315	1,046 ± 4,018	NS	18:0	7.82 ± 0.79	7.73 ± 0.76	NS
18:1 (ω-9)	3,424 ± 5,047	2,791 ± 1,165	NS	18:1 (ω-9)	20.98 ± 3.58	20.69 ± 3.37	NS
18:2 (ω-6)	4,261 ± 1,230	4,249 ± 1,614	NS	18:2 (ω-6)	32.12 ± 4.58	31.37 ± 4.52	NS
18:3 (ω-3)	48.43 ± 49.50	39.08 ± 22.38	NS	18:3 (ω-6)	0.43 ± 0.15	0.46 ± 0.14	NS
18:3 (ω-6)	58.48 ± 32.81	62.02 ± 41.35	NS	18:3 (ω-3)	0.27 ± 0.09	0.32 ± 0.18	0.09
20:0	35.26 ± 11.04	36.52 ± 13.34	NS	20:0	0.30 ± 0.06	0.29 ± 0.06	NS
20:4 (ω-6)	859.92 ± 290	849 ± 374	NS	20:4 (ω-6)	7.03 ± 1.41	7.13 ± 1.32	NS
20:5 (ω-3)	71.48 ± 54.56	73.51 ± 56.98	NS	20:5 (ω-3)	0.44 ± 0.21	0.67 ± 0.43	<0.0001
22:0	91.82 ± 30.81	97.08 ± 37.09	NS	22:0	0.86 ± 0.16	0.84 ± 0.18	NS
22:6 (ω-3)	248 ± 101.46	247 ± 128.79	NS	22:6 (ω-3)	2.00 ± 0.50	2.11 ± 0.68	NS
24:0	108.54 ± 32.31	112.25 ± 46.76	NS	24:0	1.06 ± 0.16	1.09 ± 0.16	NS
24:1 (ω-9)	125.28 ± 43.53	122.62 ± 49.27	NS	24:1 (ω-9)	1.23 ± 0.31	1.20 ± 0.29	NS
Essential FA	4,310 ± 1,244	4,288 ± 1,633	NS	Essential FA	43.4 ± 4.7	43.1 ± 4.6	NS
Saturated FA	4,196 ± 1,158	4,225 ± 1,445	NS	Saturated FA	30.3 ± 1.7	30.8 ± 1.9	NS
Monounsaturated	3,978 ± 3,117	3,334 ± 1,336	NS	Monounsaturated	22.35 ± 4.66	22.79 ± 4.23	NS
Polyunsaturated ω-3	319 ± 144	321 ± 178	NS	Polyunsaturated ω-3	2.72 ± 0.6	3.1 ± 1	0.01
Polyunsaturated ω-6	1,140 ± 376	1,152 ± 494	NS	Polyunsaturated ω-6	40.7 ± 4.6	40 ± 4.7	NS
Women				Women			
14:0	85.96 ± 89.30	67.27 ± 57.33	NS	14:0	0.40 ± 0.24	0.42 ± 0.34	NS
16:0	2,064 ± 1,171	2,875 ± 996	NS	16:0	18.9 ± 2.95	18.94 ± 3.27	NS
16:1 (ω-9)	57.60 ± 23.96	53.18 ± 28.35	NS	16:1 (ω-9)	0.34 ± 0.12	0.36 ± 0.09	NS
18:0	1,005 ± 317	1,026 ± 337	NS	18:0	7.52 ± 1.11	7.72 ± 0.95	NS
18:1 (ω-9)	2,853 ± 1,209	2,825 ± 1,290	NS	18:1 (ω-9)	22.36 ± 8.72	20.95 ± 3.53	NS
18:2 (ω-6)	4,337 ± 1,316	4,129 ± 1,218	NS	18:2 (ω-6)	32.1 ± 5.80	31.89 ± 5.13	NS
18:3 (ω-3)	41.58 ± 24.87	36.02 ± 15.81	NS	18:3 (ω-6)	0.42 ± 0.15	0.44 ± 0.19	NS
18:3 (ω-6)	61.42 ± 32.75	59.38 ± 33.59	NS	18:3 (ω-3)	0.29 ± 0.15	0.34 ± 0.35	NS
20:0	35.93 ± 11.04	34.64 ± 13.33	NS	20:0	0.30 ± 0.08	0.30 ± 0.07	NS
20:4 (ω-6)	859 ± 295	853 ± 296	NS	20:4 (ω-6)	6.92 ± 1.43	7.1 ± 1.35	NS
20:5 (ω-3)	81.29 ± 71.93	61.27 ± 50.11	NS	20:5 (ω-3)	0.47 ± 0.33	0.59 ± 0.38	0.1
22:0	94.11 ± 29.92	92.89 ± 30.94	NS	22:0	0.88 ± 0.20	0.88 ± 0.19	NS
22:6 (ω-3)	239 ± 110	204 ± 104	NS	22:6 (ω-3)	1.82 ± 0.63	2.06 ± 0.55	0.05
24:0	111.38 ± 34.12	106.80 ± 33.09	NS	24:0	1.11 ± 0.21	1.08 ± 0.20	NS
24:1 (ω-9)	120.51 ± 40.46	125.64 ± 53.12	NS	24:1 (ω-9)	1.25 ± 0.32	1.21 ± 0.33	NS
Essential FA	4,379 ± 1,326	4,165 ± 1,225	NS	Essential FA	43.1 ± 6.6	43.5 ± 5.4	NS
Saturated FA	4,333 ± 1,350	4,241 ± 1,362	NS	Saturated FA	29.4 ± 3.6	29.6 ± 3.2	NS
Monounsaturated	3,438 ± 1,431	3,407 ± 1,533	NS	Monounsaturated	24.6 ± 9.4	22.3 ± 4.73	NS
Polyunsaturated ω-3	320 ± 174	265 ± 142	NS	Polyunsaturated ω-3	2.59 ± 0.93	3.01 ± 0.96	0.04
Polyunsaturated ω-6	1,148 ± 386	1,155 ± 410	NS	Polyunsaturated ω-6	40.5 ± 6.4	40.5 ± 5.4	NS

Data are means ± SD.

weight (BMI >24.4 kg/m^2 in men and >22.9 in women) and lean subjects. This cut point corresponds approximately to a BMI of 25 kg/m^2 in the Caucasian population of Spain (18). By definition, overweight subjects showed increased BMI (26.9 ± 2.4 vs. 21.7 ± 1.7 kg/m^2, $P <$ 0.000001) and WHR (0.94 ± 0.07 vs. 0.88 ± 0.6, $P < 0.03$) and were significantly older (42.1 ± 8.9 vs. 37.4 ± 10 years, $P = 0.006$). Circulating IL-6 level was below the minimum detectable concentration of the assay (0.094 pg/ml) in 58 subjects, and they were thus excluded from further analysis. These subjects did not significantly differ regarding age, sex, and BMI from the remaining subjects.

The percentage of saturated FAs (29.9 ± 1.8 vs. 30.4 ± 2.1) and ω-6 FAs (40.6 ± 5.0 vs. 40.6 ± 5.2) were not significantly different in overweight and lean subjects, whereas ω-3 FAs were significantly lower in the former (2.7 ± 0.8 vs. 3.0 ± 1.0, $P = 0.04$). The same findings were observed separately in men and women (Table 3), except in that overweight men also had a decreased percentage at 14:0 and 20:5 FA, and women with overweight had decreased (22:6) ω-3. The proportions of saturated/ω-3 or satu-

Table 3—*Anthropometric and biochemical variables in women*

	Lean	Overweight	*P*
n	45	53	—
Age (years)	35.2 ± 7.4	40.8 ± 9.8	0.003
Weight (kg)	54.5 ± 5.2	68.3 ± 7.5	<0.0001
BMI (kg/m^2)	20.4 ± 1.5	26.4 ± 3.2	<0.0001
WHR	0.84 ± 0.84	0.86 ± 0.04	0.1
Smokers (yes/no)	9/36	7/46	NS
Systolic blood pressure (mmHg)	111.3 ± 10	121.4 ± 12	<0.0001
Diastolic blood pressure (mmHg)	63.6 ± 5.5	69.1 ± 10.1	0.002
Fasting glucose (mmol/l)	4.5 ± 0.5	4.8 ± 0.5	0.008
Fasting insulin (mU/l)	6.5 ± 2.3	8.5 ± 4.1	0.006
HOMA value	1.18 ± 0.4	1.67 ± 0.8	0.002
Cholesterol (mg/dl)	202 ± 38	205 ± 43	NS
Triglycerides (mg/dl)	73.3 ± 27	76.2 ± 29	NS
IL-6 (pg/ml)*	3.8–13.8	5–20.3	0.055
C-reactive protein (mg/dl)	0.37 ± 0.1	0.57 ± 0.6	0.02

Data are *n*, means ± SD, and interquartile range.

rated/ω-6 FAs were not significantly different between groups.

In overweight subjects, the percentage of saturated FAs ($r = 0.30$, $P = 0.01$) and ω-6 FAs ($r = -0.32$, $P = 0.001$) were significantly associated with circulating IL-6 (Fig. 1), whereas the percentage of ω-3 FAs correlated negatively with C-reactive protein ($r = -0.25$, $P = 0.04$). Saturated–to–ω-3 and saturated–to–ω-6 FA ratios were significantly and positively associated with C-reactive protein ($P < 0.0001$) and IL-6 ($P < 0.001$), respectively. The relations were stronger in smokers. In contrast, none of these associations showed statistical significance in lean subjects. All of these results persisted after controlling for age. We constructed a multivariant regression analysis to predict circulating IL-6. Age, BMI, WHR, smoking status, and the relation of saturated/ω-6 or saturated/ω-3 FAs were considered as independent variables separately in men and women. In overweight men, saturated/ω-3 FAs ($P = 0.01$), but not age, sex, BMI, WHR, or smoking status, independently contributed to 17% of IL-6 variance (Table 4). In lean men, smoking status ($P = 0.02$), but not the remaining variables, contributed to 8% of IL-6 variance. In women, any of these variables contributed independently to IL-6 variance.

Those subjects in the most insulin-sensitive quintile (HOMA value) (quintiles mean ± SD: 0.73 ± 0.19, 1.13 ± 0.08, 1.45 ± 0.09, 1.89 ± 0.16, and 3.41 ± 1.6 in men and 0.75 ± 0.11, 1.02 ± 0.06, 1.35 ± 0.12, 1.76 ± 0.13, and 3.12 ± 0.8 in women) had a significantly higher percentage of linolenic acid (C18:3 ϖ3) (0.48 ± 0.18 vs. 0.42 ± 0.15, $P = 0.032$) and significantly lower araquidic (C20:0) ($P = 0.04$), behenic (C22:0) ($P = 0.009$), lignoceric (C24:0) ($P = 0.02$), and nervonic (C24:1 ϖ9) ($P = 0.001$) FAs than the remaining subjects. In parallel, the most insulin-sensitive subjects had significantly decreased C-reactive protein (0.35 ± 0.12 vs. 0.44 ± 0.36, $P = 0.03$). Specifically, serum C-reactive protein was significantly associated with percent linoleic acid (C18:2 ϖ6) and eicosapentaenoic (EPA) acid (C20:5 ϖ3) in nonsmoking men ($P = 0.03$ and $P = 0.04$, respectively) and with DHA acid (C22:6 ϖ3) in nonsmoking women ($r = -0.46$, $P < 0.0001$) (Table 5).

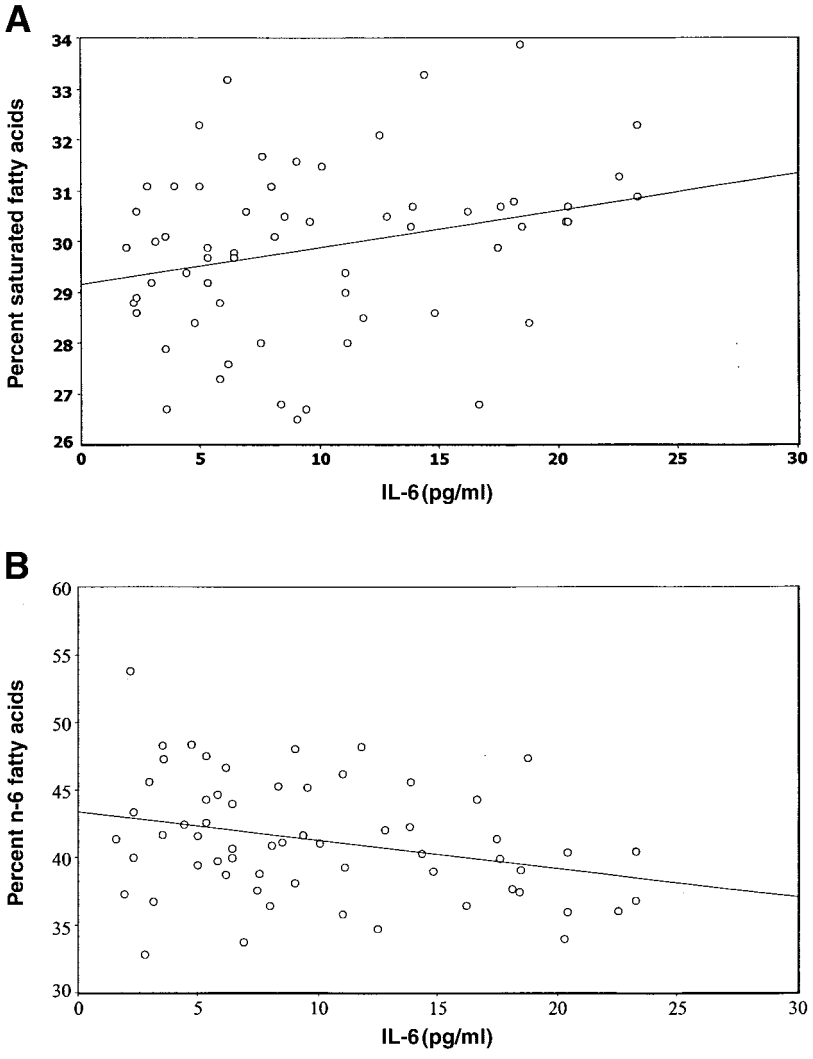

Figure 1—*Association between percent saturated FAs and IL-6 (A) and between percent ω-6 polyunsaturated FAs and IL-6 (B) in overweight subjects with detectable IL-6 levels.*

Table 4—*Multiple linear regression analysis equations in overweight men*

Variable	B	SE B	β	T	Sig T
Age	0.165728	0.125602	0.187013	1.319	0.19
BMI	−0.642939	0.500064	−0.179762	−1.286	0.20
TAB	0.253204	0.152662	0.220966	1.659	0.10
WHR	−30.817504	47.512974	−0.096397	−0.649	0.51
HOMA	1.892206	6.046741	0.044868	0.313	0.75
SAT/ω3	1.072576	0.439140	0.323850	2.442	0.0181
(Constant)	35.565789	43.175453		0.824	0.41

Multiple $r = 0.41$; $r^2 = 0.17$. SAT/ω3, ratio of percent saturated to percent ω-3 fatty acids in serum. TAB, smoking.

When evaluating individual FAs in all subjects, serum IL-6 concentration correlated positively with the proportion of the saturated myristic (C14:0) and palmitic acid (C16:0) in smoking men and in nonsmoking women (Table 5). The absolute concentrations of the saturated FA C18:0 (stearic) and C24:0 (lignoceric) were also positively associated with IL-6 among smoking men ($r = 0.31$, $P = 0.031$ and $r = 0.30$, $P = 0.038$, respectively). A tendency toward a negative association with linoleic acid (C18:2 ϖ6) was shown in smoking men and in nonsmoking women (Table 5).

CONCLUSIONS — Dietary FAs (as inferred from plasma FA concentration) seem to be strongly linked to inflammatory activity. Interestingly, this association is especially remarkable in subjects with increased fat mass, defined as BMI >24.4 kg/m^2 in men and >22.9 kg/m^2 in women. Our population showed increased fat mass in comparison with other populations, so these cut points correspond to a BMI of 25 kg/m^2 in the American Caucasian population (18). We found that the percentage of saturated FAs and ω-6 FAs were significantly associated with circulating IL-6, whereas the percentage of ω-3 FAs correlated negatively with C-reactive protein in overweight subjects. Saturated–to–ω-3 and saturated–to–ω-6 FA ratios were significantly and positively associated with C-reactive protein and IL-6, respectively. All of these associations hint at obesity as an inflammatory disease of dietary origin, although cause and consequence cannot be derived from the present study. In sharp contrast, any of these associations showed statistical significance in lean subjects. However, it should be considered that the greater IL-6 and C-reactive protein variances in overweight versus lean individuals could explain the lack of relation in lean subjects.

Table 5—*Study of the associations of circulating IL-6 and C-reactive protein according to sex and smoking status*

	n	Myristic (C14:0)	Palmitic (C16:0)	Linoleic (C18:2 ω-6)	EPA (C20:5 ω-3)	DHA (C22:6 ω-3)
IL-6						
Nonsmoking men	84	0.04	−0.09	0.02	−0.03	−0.03
P		NS	NS	NS	NS	NS
Smoking men	48	**0.35**	**0.42**	**−0.27**	−0.10	−0.09
P		**0.01**	**0.002**	**0.06**	NS	NS
Nonsmoking women	80	**0.26**	**0.28**	**−0.19**	−0.06	0.11
P		**0.01**	**0.01**	**0.09**	NS	NS
C-reactive protein						
Nonsmoking men	84	0.05	0.11	**−0.23**	**−0.22**	−0.17
P		NS	NS	**0.03**	**0.04**	0.10
Smoking men	48	−0.12	−0.11	−0.11	0.01	0.11
P		NS	NS	NS	NS	NS
Nonsmoking women	80	0.08	0.04	0.11	−0.003	**−0.46**
P		NS	NS	NS	NS	**<0.0001**

Bold data indicate significance difference.

Serum FA composition is largely determined by dietary intake and is a good indicator of habitual dietary fat intake in middle-aged adults (15,19). It is well known that dietary composition of long-chain FAs influence inflammation and atherogenesis in in vitro, animal, and human studies involving relatively few subjects (12–15). In a recent in vitro study, saturated FAs induced nuclear factor κB activation, an important mediator in the production of IL-6 and several other cytokines (20). DHA (C22:6 ϖ3) and EPA acids (C20:5 ϖ3) inhibited in vitro production of IL-6 by human endothelial cell (21). DHA also reduced endothelial expression of IL-6 in response to different stimulus (22). In parallel to these observations, supplementation with EPA and DHA appears to reduce cytokine production in humans. Meydani et al. (13) studied the effect of EPA and DHA as dietary supplements in 12 women; this supplementation significantly decreased cytokine production by peripheral mononuclear cells. Endres et al. (14) gave supplements to nine volunteers consisting of 18 g/day of fish oil that contained 2.7 g EPA and 1.85 g DHA and found a significant decrease (43%) in the production of IL-1β by peripheral blood mononuclear cells.

The association of percent ω-3 and C-reactive protein herein described seems especially noticeable when considering recent findings in American women. An increased intake of the ω-3 polyunsaturated FA, EPA, and DHA was associated with reduced risk of thrombotic brain infarction (23). These relations were most likely to have a dietary explanation because food is the major source of these FAs. EPA, in part converted from DHA, is transformed into a nonaggregatory agent that increases the synthesis of a vasodilator, prostaglandin I_3, leading to further reduction in platelet aggregation and to increased vasodilation (24).

The physiological relevance of a significant difference in percent araquidic (C20:0), behenic (C22:0), and lignoceric (C24:0) in insulin-sensitive versus insulin-resistant subjects cannot be inferred from the present study. However, substituting dietary saturated fat with polyunsaturated fat improved insulin sensitivity in recent studies (25).

The low proportion of arachidonic

acid in men with overweight is also remarkable. In fact, a high ratio between arachidonic (20:4 n-6) and dihomo-γ linolenic (20:3 n-6) acid, as a measure of Δ5 desaturase activity, in the skeletal muscle phospholipids has been related to good insulin sensitivity (26).

The associations of percent FAs and IL-6 need also to be interpreted in the context of the atherosclerotic process. It is well known that inflammation in the vessel wall plays an essential part in the initiation and progression of atherosclerosis (27–29). Damage to the vessel wall leads to endothelial cell disruption, resulting in exposure of the underlying vascular smooth muscle cells. Endothelial and smooth muscle cells produce IL-6, and IL-6 gene transcripts are expressed in human atherosclerotic lesions (30). Prospective studies in apparently healthy and high-risk individuals indicate that increased IL-6 and C-reactive protein concentration on one hand (9,31–33) and FA composition on the other hand are associated with insulin resistance, type 2 diabetes, and cardiovascular events (34). The findings of the present study suggest that these associations are interrelated events.

One limitation of this study is its transversal design and, thus, the findings shown here are hypothesis generating. The associations that have been found could be due to a variety of other factors and do not indicate cause and effect. It will be necessary to demonstrate that IL-6 levels can be modulated by feeding certain FAs (35) or by changing serum FA composition.

In summary, dietary FAs (as inferred from plasma FA concentration) seem to be linked to inflammatory activity in overweight subjects and in subjects with insulin resistance. Being overweight modulates the relations of FAs to inflammatory markers.

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