Journal of the American College of Cardiology
© 2006 by the American College of Cardiology Foundation
Published by Elsevier Inc.

Vol. 48, No. 8, 2006
ISSN 0735-1097/06/$32.00
doi:10.1016/j.jacc.2006.06.057

Acute Effects of High-Fat Meals Enriched With Walnuts or Olive Oil on Postprandial Endothelial Function

Berenice Cortés, BS,*† Isabel Núñez, MD,‡ Montserrat Cofán, PHD,† Rosa Gilabert, MD, PHD,‡ Ana Pérez-Heras, RD,† Elena Casals, MD, PHD,§ Ramón Deulofeu, PHD,§ Emilio Ros, MD, PHD†

Barcelona, Spain

OBJECTIVES We sought to investigate whether the addition of walnuts or olive oil to a fatty meal have differential effects on postprandial vasoactivity, lipoproteins, markers of oxidation and endothelial activation, and plasma asymmetric dimethylarginine (ADMA).

BACKGROUND Compared with a Mediterranean diet, a walnut diet has been shown to improve endothelial function in hypercholesterolemic patients. We hypothesized that walnuts would reverse postprandial endothelial dysfunction associated with consumption of a fatty meal.

METHODS We randomized in a crossover design 12 healthy subjects and 12 patients with hypercholesterolemia to 2 high-fat meal sequences to which 25 g olive oil or 40 g walnuts had been added. Both test meals contained 80 g fat and 35% saturated fatty acids, and consumption of each meal was separated by 1 week. Venipunctures and ultrasound measurements of brachial artery endothelial function were performed after fasting and 4 h after test meals.

RESULTS In both study groups, flow-mediated dilation (FMD) was worse after the olive oil meal than after the walnut meal ($p = 0.006$, time-period interaction). Fasting, but not postprandial, triglyceride concentrations correlated inversely with FMD ($r = -0.324$; $p = 0.024$). Flow-independent dilation and plasma ADMA concentrations were unchanged, and the concentration of oxidized low-density lipoproteins decreased ($p = 0.051$) after either meal. The plasma concentrations of soluble inflammatory cytokines and adhesion molecules decreased ($p < 0.01$) independently of meal type, except for E-selectin, which decreased more ($p = 0.033$) after the walnut meal.

CONCLUSIONS Adding walnuts to a high-fat meal acutely improves FMD independently of changes in oxidation, inflammation, or ADMA. Both walnuts and olive oil preserve the protective phenotype of endothelial cells. (J Am Coll Cardiol 2006;48:1666–71) © 2006 by the American College of Cardiology Foundation

Endothelial dysfunction is a critical event in atherogenesis that is implicated both in early disease and in advanced atherosclerosis (1). It is characterized by a decreased bioavailability of nitric oxide (NO) and increased expression of proinflammatory cytokines and cellular adhesion molecules (2). The predominant mechanism of NO inactivation is a perturbation of the L-arginine–NO pathway by oxidative stress leading to elevations of plasma asymmetric dimethylarginine (ADMA), which in turn exacerbates oxidative stress (3). Both a pro-oxidant status and increased ADMA are common features of disease states associated with atherosclerosis, including hypercholesterolemia (3,4).

Food intake is an important factor that affects vascular reactivity. Short-term feeding trials have shown the potential of food for improving endothelial function, either as isolated nutrients, such as n-3 polyunsaturated fatty acids (PUFA), L-arginine, and antioxidant vitamins, or as healthy food patterns (5). A high-fat meal is usually followed by transient endothelial dysfunction in association with raised triglyceride-rich lipoproteins (6). Abnormal vasoactivity after a fatty meal is attenuated by pretreatment with antioxidant phytochemicals (7) or addition of antioxidants to the meal (8,9), suggesting that postprandial oxidative stress plays an important role. Elevated concentrations of the endogenous NO inhibitor ADMA may also contribute to fatty meal-induced endothelial dysfunction (3).

Walnuts are a rich source of antioxidants, L-arginine, and α-linolenic acid (ALA), a plant n-3 PUFA. Recently we showed that, compared with a Mediterranean diet, a walnut diet improves endothelial function in hypercholesterolemic patients (10). To test the hypothesis that walnuts also would have acute favorable effects on vasoactivity, we examined the effects of adding walnuts or olive oil to a single high-fat meal on postprandial endothelial function of the brachial

From the *Departament de Medicina, Universitat Autònoma de Barcelona, Barcelona, Spain; and the †Secció d'Ecografia, Centre de Diagnòstic per l'Imatge; ‡Unitat de Lípids, Institut Clínic de Malalties Digestives i Metabòliques; and §Centre de Diagnòstic Biològic, Institut d'Investigacions Biomèdiques August Pi i Sunyer, Hospital Clínic, Barcelona, Spain. Supported by an unrestricted grant and provision of walnuts by the California Walnut Commission (CWC), Sacramento, California, and by grants from the Spanish Ministry of Health (FIS 00/0992, RT/C03-01, RT/G03-140). Berenice Cortés was supported by a grant from Fundación Carolina, Madrid, Spain. Dr. Ros serves on the Scientific Advisory Board of the CWC. The first two authors contributed equally to this work.

Manuscript received March 22, 2006; revised manuscript received May 31, 2006, accepted June 6, 2006.

Abbreviations and Acronyms

ADMA = asymmetric dimethylarginine
ALA = α-linolenic acid
FID = flow-independent dilation
FMD = flow-mediated dilation
MUFA = monounsaturated fatty acids
NO = nitric oxide
PUFA = polyunsaturated fatty acids
sICAM-1 = soluble intercellular adhesion molecule 1
SFA = saturated fatty acids
sTNF-R = soluble tumor necrosis factor receptors
sVCAM-1 = soluble vascular cell adhesion molecule 1

artery and markers of oxidation and endothelial activation in controls and hypercholesterolemic subjects.

METHODS

Subjects. Twenty-four asymptomatic subjects were recruited into a protocol approved by the institutional review board and gave informed consent. Twelve subjects were healthy, normolipidemic controls, and 12 subjects had moderate hypercholesterolemia (low-density lipoprotein [LDL] cholesterol 150 to 220 mg/dl, triglycerides <200 mg/dl) (Table 1). All participants were nonsmokers, had normal body weight and blood pressure, consumed <30 g/day alcohol, and took no medications or antioxidant supplements. Lack of a history of allergy to nuts, normal fasting blood glucose and thyroid, renal, and hepatic function tests, and absence of carotid atherosclerosis at ultrasound examination were prerequisites for entry.

Table 1. Characteristics of Study Groups

Gender	Control Subjects (n = 12)	Hypercholesterolemic Subjects (n = 12)
Gender, men/women	9/3	11/1
Age, yrs	32 ± 8	45 ± 13
Body mass index, kg/m^2	24.7 ± 3.0	26.3 ± 3.5
Waist circumference, cm	93 ± 10	96 ± 7
Glycemic control		
Glucose, mg/dl	82 ± 4.5	85 ± 7.5
Insulin, mU/l	8.2 ± 2.4	9.1 ± 4.3
Free fatty acids, umol/l	673 ± 302	466 ± 202
Lipids		
Total cholesterol, mg/dl	185 ± 27	250 ± 25
HDL cholesterol, mg/dl	59 ± 13	58 ± 12
VLDL cholesterol, mg/dl	12 ± 9	19 ± 11
LDL cholesterol, mg/dl	115 ± 26	173 ± 22
Triglycerides, mg/dl	87 ± 47	128 ± 42
Apolipoprotein B, g/l	0.82 ± 0.17	1.18 ± 0.14
LDL cholesterol/LDL apoB	1.49 ± 0.10	1.57 ± 0.09
VLDL triglyceride/VLDL apoB	7.8 ± 3.1	8.2 ± 3.2
Inflammatory markers		
E-selectin, ng/ml	33 ± 8	37 ± 10
sICAM-1, ng/ml	265 ± 35	274 ± 44
sVCAM-1, ng/ml	917 ± 216	967 ± 319
sTNF receptors, ng/ml	2.12 ± 0.36	2.21 ± 0.44
ADMA, μmol/l	0.73 ± 0.1	0.74 ± 0.1

ADMA = asymmetric dimethylarginine; apoB = apolipoprotein B; HDL = high-density lipoprotein; LDL = low-density lipoprotein; sICAM-1 = soluble intercellular adhesion molecule 1; sTNF = soluble tumor necrosis factor; sVCAM = soluble vascular adhesion molecule 1; VLDL = very low-density lipoprotein.

Study protocol. Between 2 and 6 weeks before testing, candidates came to the clinic for clinical history, interview with the dietitian, anthropometric and blood pressure measurements, blood extraction, and carotid sonography to confirm eligibility. For 2 weeks before the first study and the ensuing period until the second study, participants were instructed to follow a cholesterol-lowering Mediterranean diet (10,11) and to abstain from physical exertion. Compliance with the background diet was assessed before testing using a 7-day food record. Participants were individually randomized in a crossover design between 2 meal sequences (high-fat meals with walnuts or olive oil) and were studied on 2 separate days 1 week apart. The experiments were performed in the afternoon to avoid confounding by the early morning blunting in endothelial function (12,13). On each study day, participants were asked to eat a low-fat breakfast with coffee ad libitum at 7:00 AM and to refrain from further food intake until 1:30 PM, when they reported to the clinic and had a blood extraction. At 2:00 PM a baseline ultrasound assessment of endothelial function in the brachial artery was performed. Thereafter, participants ate 1 of the 2 meals under the supervision of a clinical investigator. The protocol was repeated 4 h postprandially, with blood extraction at 5:30 PM and a second endothelial function test at 6:00 PM. In previous studies in subjects without overt hypertriglyceridemia, the largest changes in triglycerides and endothelial function have been observed at 3 to 4 h after the meal (4,12). During the 4-h interval, participants rested in a quiet room and were allowed to drink water. The primary end points were the between-meal differences in changes from baseline of flow-mediated dilation (FMD) assessed as the percentage change in brachial artery diameter during reactive hyperemia. Postprandial changes in flow-independent dilation (FID), glycemic control, lipoproteins, oxidation markers, and plasma concentrations of vitamins, ADMA, and inflammatory molecules were secondary end points.

Test meals. The meals were prepared at the hospital's kitchen and consisted of ~1,200 kcal with 63% fat (35% saturated fatty acids [SFA]), 15% protein, 22% carbohydrate, and 120 mg cholesterol, for a total fat content of 80 g. They included a sandwich with 100 g white bread, 75 g salami, and 50 g fatty cheese, 125 g fat-rich (10%) yogurt, and water ad libitum. Additionally, participants consumed 25 ml olive oil soaked into the bread (olive oil meal) or 40 g shelled walnuts (walnut meal). The unsaturated fatty acid content of the olive oil and walnut meals differed: 38% and 23% monounsaturated fatty acids (MUFA), and 7% and 23% PUFA, respectively. Only the walnut meal contained ALA (5.4 g). The nutrient composition of the walnuts used in the study has been published previously (10). The olive oil used contained 78% oleic acid and 30 mg/100 g α-tocopherol.