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A Monounsaturated Fatty Acid–Rich Pecan-Enriched Diet Favorably Alters the Serum Lipid Profile of Healthy Men and Women¹

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ABSTRACT Frequent consumption of nuts is associated with decreased risk of cardiovascular disease. We investigated the effect of pecans rich in monounsaturated fat as an alternative to the Step 1 diet in modifying serum lipids and lipoproteins in men and women with normal to moderately high serum cholesterol. In a single-blind, randomized, controlled, crossover feeding study, we assigned 23 subjects (mean age: 38 y; 9 women, 14 men) to follow two diets, each for 4 wk: a Step I diet and a pecan-enriched diet (accomplished by proportionately reducing all food items in a Step I diet by one fifth for a 20% isoenergetic replacement with pecans). The percentage of energy from fat in the two diets was 28.3 and 39.6%, respectively. Both diets improved the lipid profile; however, the pecan-enriched diet decreased both serum total and LDL cholesterol by 0.32 mmol/L (6.7 and 10.4%, respectively) and triglyceride by 0.14 mmol/L (11.1%) beyond the Step I diet, while increasing HDL cholesterol by 0.06 mmol/L (2.5 mg/dL). Serum apolipoprotein B and lipoprotein(a) decreased by 11.6 and 11.1%, respectively, and apolipoprotein A1 increased by 2.2% when subjects consumed the pecan compared with the Step I diet. These differences were all significant (P < 0.05). A 20% isoenergetic replacement of a Step I diet with pecans favorably altered the serum lipid profile beyond the Step I diet, without increasing body weight. Nuts such as pecans that are rich in monounsaturated fat may therefore be recommended as part of prescribed cholesterol-lowering diet of patients or habitual diet of healthy individuals. J. Nutr. 131: 2275–2279, 2001.

KEY WORDS: • lipids • lipoproteins • fatty acids • cardiovascular diseases

Dietary modification is the foundation of population strategies for the prevention and treatment of coronary heart disease (CHD).³ Because serum total and LDL cholesterol are risk factors for CHD (1), clinicians and practitioners recommend diets such as the National Cholesterol Education Program Step I diet to lower cholesterol. Although the cholesterol-lowering effect of the Step I diet is favorable (2,3), by virtue of its relatively high carbohydrate and low fat content, it tends to lower HDL cholesterol and raise triacylglycerol concentrations, both of which affect coronary risk factors adversely (4). It is imperative therefore to identify alternative diets that can more effectively modify the blood lipid profile, and thus reduce CHD risk. A simple and yet viable approach would be to introduce a whole food to the currently existing diet or to cholesterol-lowering diets of individuals.

There is epidemiologic evidence that frequent consumption of nuts protects both men and women from CHD (5–7). Previously we showed that polyunsaturated fatty acid (PUFA)rich walnuts reduced serum total and LDL cholesterol in healthy men compared with the Step I diet (8) or a Mediterranean diet (9). Recently, the Scientific Advisory of the American Heart Association reported (10) that high monounsaturated fatty acid (MUFA) diets tend to raise HDL and lower triacylglycerol concentration compared with low fat, carbohydrate-rich, cholesterol-lowering diets (11,12). In keeping with this evidence, we chose to study the effect of MUFArich pecans on blood lipids. Pecans are considered to be the traditional tree nut in the United States. In addition to being a rich source of MUFA, the unique nonfat component of pecans may also have a role in favorably modifying the blood lipid profile and potentially other cardiovascular risk factors. Thus, the objective of this controlled feeding study was to investigate the effect of pecans on blood lipids and lipoproteins in healthy men and women compared with the Step I diet.

SUBJECTS AND METHODS

Subjects. Healthy men and women volunteers from Loma Linda University and surrounding communities were selected at the completion of a multiphase screening process that included a questionnaire, group meeting, face-to-face interview with a senior investigator and an assessment of serum cholesterol and triacylglycerol concentrations. Subjects with serum cholesterol below the 15th or above the 80th percentile for their age, race and gender (13) and triacylglycerol >2.26 mmol/L (200 mg/dL) were excluded from the study. Also excluded were subjects who ate nuts (>2 times/wk), had food allergies, smoked cigarettes, drank caffeinated beverages (\geq 3 times/d) or alcohol (more often than on a rare social occasion), had a history of CHD or other metabolic diseases, took medication that interfered

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³ Abbreviations used: apo, apolipoprotein; CHD, coronary heart disease; Lp(a), lipoprotein(a); MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids; SFA, saturated fatty acids.

with lipid metabolism or had a body mass index >30 kg/m². Women who had started hormone replacement within 5 y of entry into the study and those with irregular menses were also excluded.

Statistical power calculations indicated that to detect mean differences of 0.26 mmol/L (10 mg/dL) for total and LDL cholesterol, 22 participants would have to complete the two treatment periods (statistic, 0.05; power >0.9) of this crossover design. Of the 24 subjects who were randomly assigned to groups, 23 (11 Caucasians, 6 Asians, 4 Hispanics and 2 African-Americans) successfully completed the study. There were 9 women and 14 men in the age range of 25–55 y. The body weight and baseline serum cholesterol of the subjects were 74.4 \pm 16.7 kg and 4.64 \pm 0.83 mmol/L [179.4 \pm 32.2 mg/dL], respectively. The study protocol was approved by the Institutional Review Board of the University; all interested subjects signed the informed consent form and were offered a cash incentive of \$200 upon successful completion of the study.

Study and diet design. The first 2 wk of this single-blind, crossover study comprised a run-in phase in which subjects were fed a typical American diet (34% energy from total and 15% from saturated fat). The subjects were randomly assigned after stratification based on two categories of age, gender and screening values of serum cholesterol, into either the Step I diet or the pecan-enriched diet for the first 4 wk. The groups then reversed their diet intervention and continued for another 4 wk. Given that lipoprotein values stabilize in <4 wk of a diet intervention (14), and based on our previous studies that showed no carry-over effects with walnuts (8,9), we did not include a washout period between the two diet periods in this study. The diets were isoenergetic, but the percentage of energy from fat was higher in the pecan (39.6%) than in the Step I diet (28.3%). Pecans did not replace a given food or fat in the Step I diet, but a portion of the entire diet. This was accomplished by reducing the portion size of all items on the menu of the Step I diet by one fifth (i.e., total energy was reduced by 20%) to accommodate the pecans, which were served plain, in salads, gravies, shakes and as toppings. The amount of pecans consumed daily by the subjects per 10,032 kJ (2400 kcal) was 72 g (2.5 oz).

Meal service and quality control. All meals were prepared in the University Metabolic Kitchen and each food accurately weighed to the nearest gram by trained weighers. Subjects ate breakfast and dinner, Sunday through Friday, in the Metabolic Kitchen dining facility, and all lunches and Saturday meals were packed for carry out. A total of nine different daily menus of commonly consumed foods were prepared using standardized recipes; menus were used in rotation to increase variety.

For subjects to maintain constant body weight during the study, energy intake had to be adjusted periodically. Frequent monitoring of body weight throughout the study and subjective feelings of hunger expressed by participants were used in making the necessary adjustments in energy intake. The energy intake used in this study ranged from 8368 (2000 kcal) to 15,062 kJ (3600 kcal) with 1674 kJ (400 kcal) increments. Homogenized samples of both diets were collected on 18 randomly selected days covering both diet periods. Samples were mixed and analyzed for macronutrients and fatty acid composition (Covance Laboratories, Madison, WI).

Compliance. Subjects were required to consume all meals served in the Metabolic Kitchen and were not allowed to consume nonstudy foods or beverages except water. They were required to maintain the same lifestyle activities as reported at the time of the screening interview. Subjects were asked to record any deviations from this "norm" in a diary, which was reviewed frequently by an investigator, one of whom was always present at meal times to interact with the subjects. Dietary compliance was also assessed by measuring plasma fatty acids (UC Davis Nutrition laboratory) at the end of each diet intervention.

Laboratory measurements. Blood was drawn from fasting subjects on two alternate days at the end of the run-in phase and the end of the two diet periods. All samples were processed immediately (Smith-Kline Beecham, Loma Linda, CA) and serum aliquots shipped to the Nutritional Assessment Core of the University of California, Davis (NIH Clinical Nutrition Research Unit, NIDDK 35747) for analyses. Serum total cholesterol, LDL cholesterol, HDL cholesterol and triacylglycerol were determined by enzymatic assays

using the 550 Express Chemistry Analyzer (Bayer, Tarrytown, NY). HDL cholesterol was determined after anionic precipitation of apolipoprotein (apo) B-containing lipoproteins. Apo A1 and B were measured by rate immunonephelometry (Beckman Coulter, Brea, CA). Lipoprotein(a) [Lp(a)] was determined turbidimetrically (Dia-Sorin, Stillwater, MN). The diet groupings were not disclosed to the laboratory personnel.

Statistical analysis. Mean and standard deviations are presented for each measurement. Differences between the pecan-enriched and Step 1 diets were tested by repeated-measures analysis of variance and covariance; baseline values or gender were used as covariates. Period and carry-over effects were evaluated using appropriate interaction terms. Analyses were performed using SAS software (15). Differences were considered significant at P < 0.05.

RESULTS

Dietary compliance. We observed a high degree of congruence between the planned and analyzed diet composition (Table 1). The percentages of energy from MUFA and PUFA were almost 100% greater in the pecan-enriched diet than in the Step I diet, resulting in markedly different PUFA:MUFA: saturated fatty acid (SFA) ratios in the Step I (6:11:8) and pecan-enriched (11:19:8) diets. Pecan consumption in the study was 100% because they were consumed under supervision. We estimated the overall dietary compliance to be >95% on the basis of supervision by investigators during on-site meal times, 6 d a week, and by personal diaries kept by subjects. In addition, the fatty acid composition of the triacylglycerol fraction of plasma lipids and those of the two diets were consistent. As expected, plasma triacylglycerol SFA and ratios of SFA to MUFA and SFA to PUFA were significantly lower when subjects consumed the pecan-enriched diet compared with the Step I diet (Table 2). This was expected given the higher percentage of energy from oleic acid in the pecanenriched diet (18.5%) compared with the Step I diet (10.5%).

TABLE 1

Planned and analyzed compositions of the Step I and pecan-enriched diets

	Step	o I diet	Pecan-enriched diet	
Nutrient	Planned	Analyzed ¹	Planned	Analyzed ¹
Energy,				
kJ/d	10042	9983	10042	10422
kcal/d	2400	2386	2400	2491
Fat, % energy	29.9	28.3	42.1	39.6
Saturated	9.3	8.2	8.8	8.1
8:0–16:0	—	5.9	—	5.9
18:0	—	2.2	—	2.1
Monounsaturated	11.1	11.0	20.2	18.9
18:1(n-9)	—	10.5	—	18.6
Polyunsaturated	5.3	6.3	8.8	10.7
18:2(n-6)	—	5.6	—	9.9
18:3(n-3)	—	0.7	—	0.7
Protein, % energy	13.2	14.5	11.6	13.1
Carbohydrate, % energy	59.4	56.8 ²	49.7	47.2 ²
Cholesterol, ⁴ mg/4184 kJ	—	88.1 ³	—	70.3 ³
Fiber, ⁴ g/4184 kJ	—	10.53	—	12.01

¹ Values obtained during chemical analysis of samples from the study diets.

² The values for carbohydrate intake were calculated by subtracting the values for fat and protein intake from those for total energy intake. ³ Values calculated from nutrient analysis of the menus.

4 4184 kJ = 1000 kcal.

TABLE 2

Fatty acid composition of plasma triacylglycerols in men and women who consumed Step I and pecan-enriched diets

Fatty acid	Step I diet	Pecan-enriched diet		
	g/100 g total fatty acids ¹			
Saturated fatty acids				
8:0–16:0	26.6 ± 2.6a	22.0 ± 2.3 ^b		
18:0	3.8 ± 1.0	2.9 ± 0.6		
Monounsaturated fatty acids				
18:1(n-9)	33.1 ± 3.4a	37.3 ± 2.7b		
Polyunsaturated fatty acids				
18:2(n-6)	$22.9~\pm~5.8$	24.9 ± 3.2		
18:3(n-3)	1.4 ± 0.5	1.3 ± 0.4		
Ratios				
SFA:MUFA ²	0.92a	0.67 ^b		
SFA:PUFA ²	1.25 ^a	0.95 ^b		
18:2(n-6):18:3(n-3)	16.4	19.1		
,				

¹ Values are means \pm sD, n = 23. Values in a row with different superscript letters differ, $P \leq 0.001$.

² Abbreviations: SFA, saturated fatty acids; MUFA, monounsaturated fatty acids; PUFA, polyunsaturated fatty acids.

Serum lipids and lipoproteins. Serum lipid, lipoprotein and apolipoprotein concentrations at the end of each diet period as well as the differences of effect between dietary interventions are given in Table 3. No carry-over effect was observed between the dietary periods. Serum total and LDL cholesterol were lower by 0.32 mmol/L (12.3 mg/dL), whereas HDL cholesterol was greater by 0.06 mmol/L (2.5 mg/dL) when subjects consumed the pecan-enriched diet compared with the Step I diet. This corresponded to 6.7 and 10.4% decreases in total and LDL cholesterol, respectively, and a 5.6% increase in HDL cholesterol when subject consumed the pecan-enriched diet beyond the effects of the Step I diet. The LDL cholesterol to HDL cholesterol ratio was lower by 0.44 when subjects consumed the pecan-enriched diet compared with the Step I diet. Plasma triacylglycerol, apo B and Lp(a) concentrations were significantly lower by 11.1% [0.14 mmol/L (12.7 mg/dL)], 11.6% (0.10 g/L), 15.1% (0.04 g/L) respectively, and apo A1 significantly greater by 2.2% (0.03 g/L) when subjects consumed the pecan-enriched compared with the Step I diet. All differences were significant.

Throughout the study, energy intake was adjusted to prevent changes in body weight. During the pecan-enriched diet period, subjects lost 0.43 ± 0.18 kg (P < 0.05) compared with the Step I diet period, a decrease of <1% from the mean baseline body weight. After adjusting for body weight, the differences in blood lipids between the two diet periods were maintained.

Figure 1 depicts the effectiveness of the pecan-enriched diet compared with the Step I diet by comparing the changes in serum lipid concentrations due to the two test diets to concentrations at the end of the run-in phase (baseline) when subjects consumed a typical American diet. As expected, the Step I diet lowered total and LDL cholesterol compared with the American diet. However, the pecan-enriched diet significantly lowered serum total and LDL cholesterol beyond that of the Step I diet. Furthermore, the pecan-enriched diet prevented the decrease in HDL cholesterol and the increase in triacylglycerol observed with the Step I diet and significantly decreased apo B.

DISCUSSION

In this randomized crossover study we found that 20% isoenergetic replacement of a Step I diet with pecans favorably altered multiple blood lipid variables in men and women with normal to moderately high serum cholesterol. Specifically, the pecan-enriched diet lowered serum total and LDL cholesterol compared with the Step I diet. Incorporation of pecans into self-selected diets has shown similar results with LDL cholesterol (16). Although the Step I diet is a cholesterol-lowering diet (2,3), it has previously been shown to increase serum triacylglycerol and lower HDL cholesterol concentrations (4,17). We found that the pecan-enriched diet, in addition to lowering serum cholesterol, also increased HDL cholesterol and lowered triacylglycerol concentrations. Overall, the pe-

TABLE 3

Serum lipid, lipoprotein and apolipoprotein (apo) concentrations in men and women who consumed Step I and pecan-enriched diets

Variable B		Step I diet ¹	Pecan-enriched diet ¹	Estimated differences in diet effects ²		
	Baseline values			% Change	Absolute	95% confidence interval
Total cholesterol,						
mmol/L	5.04 ± 0.84	$4.78 ~\pm~ 0.75$	4.47 ± 0.70	-6.7	-0.32	-0.45 to -0.19**
LDL cholesterol,						
mmol/L	3.27 ± 0.65	3.05 ± 0.56	2.73 ± 0.51	-10.4	-0.32	-0.41 to -0.22**
HDL cholesterol,						
mmol/L	1.20 ± 0.26	1.14 ± 0.23	1.21 ± 0.25	5.6	0.06	0.03 to 0.09**
LDL cholesterol:						
HDL cholesterol	2.86 ± 0.9	2.81 ± 0.9	2.37 ± 0.7	-15.7	-0.44	-0.56 to -0.32**
Triacylglycerol,						
mmol/L	1.23 ± 0.67	1.29 ± 0.77	1.16 ± 0.69	-11.1	-0.14	-0.29 to 0.001*
Apo A1, g/L	1.36 ± 0.20	1.30 ± 0.20	1.33 ± 0.21	2.2	0.03	0.026 to 0.032**
Apo B, g/L	0.87 ± 0.20	0.85 ± 0.21	0.75 ± 0.19	-11.6	-0.10	-0.105 to -0.094**
Lipoprotein (a),						
g/L	$0.21 ~\pm~ 0.19$	$0.25 ~\pm~ 0.22$	$0.21 ~\pm~ 0.18$	-15.1	-0.04	-0.041 to -0.033**

¹ Values are means \pm sp, n = 23.

² Pecan-enriched diet compared with Step I diet. * $P \le 0.05$. ** $P \le 0.001$.



FIGURE 1 Percentage of change from baseline concentrations (run-in phase) of serum total cholesterol (TC), LDL cholesterol (LDL), HDL cholesterol (HDL), triacylglycerol (TG), apolipoprotein A1 (apo A1) and apolipoprotein B (apo B) in men and women who consumed Step I and pecan-enriched diets. Values are means and 95% confidence interval, n = 23. Letters 'a' ($P \le 0.001$) and 'b' ($P \le 0.05$) indicate significant differences between the Step I and pecan-enriched diets.

can-enriched diet altered the lipid profile more favorably than the Step I diet.

Serum apo B concentrations decreased and apo A1 concentrations increased when subjects consumed the pecanenriched diet. These changes paralleled those observed with LDL and HDL cholesterol, respectively. A decrease in the synthesis of VLDL, the primary carrier of hepatic triacylglycerol, and an increase in its catabolism may be the cause of the decrease in plasma triacylglycerol (18). The lowering of triacylglycerol by other MUFA-rich foods has previously been demonstrated (11,19). The increase in HDL cholesterol due to the pecan-enriched diet was expected (12) given the higher percentage of fat contributed by the pecans. Also, the concomitant increase in apo A1, which stimulates cholesterol uptake by HDL, may explain in part the increase in HDL cholesterol observed when subjects consumed the pecan-enriched diet.

Changes in blood lipids were calculated by predictive equations that included changes in dietary fatty acid, cholesterol and total fat intake (20-23). These models estimated changes in serum total cholesterol ranging from 0.11 to -0.20 mmol/L (4.3 to -7.7 mg/dL) and in LDL cholesterol ranging from 0.11 to -0.25 mmol/L (-4.3 to -9.7 mg/dL). These values are much less than the mean decrease of 0.32 mmol/L (12.4 mg/dL) observed for both total and LDL cholesterol, and mainly outside their corresponding 95% confidence intervals. The lipid component of pecans contributed to the observed effects on blood lipids, but cholesterol was lowered more than the decrease estimated from differences in the fat content of the two diets. One third of pecans by weight is nonfat, and includes dietary fiber and proteins with a low ratio of lysine to arginine. Both fiber and a low ratio of lysine to arginine lower cholesterol (24–26). In addition to the traditional nutrients, the phytochemicals present in pecans may be quantified and explored in the future for their potential role in reducing some of the risk factors of CHD. Thus, the nonfat matrix of pecans may also contribute to the lipid-lowering effects observed in this study.

The beneficial changes in serum Lp(a), an independent risk factor for CHD (27) and the lipoprotein that is considered to be least affected by diet, are interesting. We and others (3,9)

have shown that Step I and Step II diets raise serum Lp(a) concentrations. In this study, serum Lp(a) was significantly lower in both men and women when they consumed the pecan-enriched diet. Walnuts (9) and fish oil (28), two excellent sources of PUFA of the (n-3) family, have both been shown to lower Lp(a) concentrations in healthy and hyper-cholesterolemic men. Given that walnuts and pecans, one rich in PUFA and the other in MUFA, both lowered Lp(a) suggests that nuts as a family may have some common component in addition to lipids that influences Lp(a) concentrations.

It has been shown that for every 1% reduction in LDL cholesterol there is a 1.5% reduction in the incidence of CHD (29). In this study, the 16% decrease in LDL cholesterol in the pecan-enriched diet compared with the American diet (Fig. 1) corresponds to a 25% decrease in CHD risk. Epidemiologic studies (5–7) have estimated that the percentage of decrease in CHD risk with frequent consumption of nuts is 40–50%. This suggests that the favorable changes in HDL cholesterol, Lp(a) and triacylglycerol (30,31) observed in the pecan group, and other mechanisms not determined in this study, may contribute to the cardioprotective effects of nuts.

Aside from the issue of suboptimal changes in the blood lipid profile, there is currently a heightened interest in searching for alternatives to the Step I diet because of difficulties with dietary compliance (32,33). Part of the challenge in complying with diets that are lower in total and saturated fat is that they require the elimination and restriction of foods. A viable alternative is an approach that relies on incorporating whole foods such as high MUFA nuts into self-selected diets or prescribed lipid-lowering diets. Recently, olive oil, a constituent of the Mediterranean diet, has been recommended as a source of MUFA (4). Because it is a fluid fat, olive oil and other high MUFA oils may be of somewhat limited use in everyday American cooking. In contrast, pecans and other high MUFA nuts are perhaps more versatile and easier to incorporate into different types of diets. This has the potential to improve dietary compliance in many patients consuming lipid-lowering diets.

In summary, the pecan-enriched diet favorably altered the blood lipid profile beyond that observed with the Step I diet. Because of the beneficial effects of nuts on several biomarkers of CHD risk and on the basis of previously reported epidemiologic evidence that showed a lowering of CHD risk with frequent consumption of nuts (5–7), it seems prudent to recommend inclusion of high MUFA nuts in cholesterol-lowering diets, or as a start, in self-selected diets.

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LITERATURE CITED

1. Davis, C. E., Rifkind, B. M., Brenner, H. & Gordon, D. J. (1990) A single cholesterol measurement underestimates the risk of coronary heart disease: an empirical example from the Lipid Research Clinics mortality follow-up study. J. Am. Med. Assoc. 264: 3044–3046.

 Stone, N. J., Nicolosi, R. J., Kris-Etherton, P., Ernst, N. D., Krauss, R. M. & Winston, M. (1996) Summary of the Scientific Conference on the Efficacy of Hypocholesterolemic Dietary Interventions. Circulation 94: 3388–3391.

3. Ginsberg, H. N., Kris-Etherton, P., Dennis, B., Elmer, P. J., Ershow, A., Lefevre, M., Pearson, T., Roheim, P., Ramakrishnan, R., Reed, R., Stewart, K., Stewart, P., Phillips, K.& Anderson, N. (1998) Effects of reducing dietary saturated fatty acids on plasma lipids and lipoproteins in healthy subjects: the DELTA Study, protocol 1. Arterioscler. Thromb. Vasc. Biol. 18: 441–449.

 Katan, M. B., Grundy, S. M. & Willett, W. C. (1997) Should a low-fat, high-carbohydrate diet be recommended for everyone? Beyond low-fat diets. N. Engl. J. Med. 337: 563–566. 5. Fraser, G. E., Sabate, J., Beeson, W. L. & Strahan, T. M. (1992) A possible protective effect of nut consumption on risk of coronary heart disease: the Adventist Health Study. Arch. Intern. Med. 152: 1416–1424.

6. Kushi, L. H., Folsom, A. R., Prineas, R. J., Mink, P. J., Wu, Y. & Bostick, R. M. (1996) Dietary antioxidant vitamins and death from coronary heart disease in postmenopausal women. N. Engl. J. Med. 334: 1156–1162.

7. Hu, F. B., Stampfer, M. J., Manson, J. E., Rimm, E. B., Colditz, G. A., Rosner, B. A., Speizer, F. E., Hennekens, C. H. & Willett, W. C. (1998) Frequent nut consumption and risk of coronary heart disease in women: prospective cohort study. Br. Med. J. 317: 1341–1345.

8. Sabate, J., Fraser, G. E., Burke, K., Knutsen, S. F., Bennett, H. & Lindsted, K. D. (1993) Effects of walnuts on serum lipid concentrations and blood pressure in normal men. N. Engl. J. Med. 328: 603–607.

9. Zambon, D., Sabate, J., Munoz, S., Campero, B., Casals, E., Merlos, M., Laguna, J. C. & Ros, E. (2000) Substituting walnuts for monounsaturated fat improves the serum lipid profile of hypercholesterolic men and women. Ann. Intern. Med. 7: 538–546.

10 Kris-Etherton, P. (1999) Monounsaturated fatty acids and risk of cardiovascular disease. Circulation 100: 1253–1258.

11. Grundy, S. M. (1986) Comparison of monounsaturated fatty acids and carbohydrates for lowering plasma cholesterol. N. Engl. J. Med. 314: 745–748.

12. Mensink, R. P. & Katan, M. B. (1987) Effect of monounsaturated fatty acids versus complex carbohydrates on high-density lipoproteins in healthy men and women. Lancet 1: 122–125.

13. National Cholesterol Education Program (1993) Second report of the Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel II), NIH Publication no. 93–3095. National Cholesterol Education Program, National Institutes of Health, National Heart, Lung and Blood Institute, Bethesda, MD

14. Kris-Etherton, P. M. & Dietschy, J. (1997) Design criteria for studies examining individual fatty acid effects on cardiovascular disease risk factors: human and animal studies. Am. J. Clin. Nutr. 65 (suppl.): 1590S–1596S.

15. The GLM Procedure (1990) SAS/STAT User's Guide, Version 6, 4th ed., vol. 2, pp. 891–996. SAS Institute, Cary, NC.

16. Morgan, W. A. & Clayshulte, B. J. (2000) Pecans lower low-density lipoprotein cholesterol in people with normal lipid concentrations. J. Am. Diet. Assoc. 100: 312–318.

17. Connor, W. E. & Connor, S. L. (1997) Should a low-fat, high-carbohydrate diet be recommended for everyone? The case for a low-fat, high-carbohydrate diet. N. Engl. J. Med. 337: 562–563.

18. Roche, H. M., Zampelas, A., Knapper, J. M., Webb, D., Brooks, C., Jackson, K. G., Wright, J. W., Gould, B. J., Kafatos, A., Gibney, M. J. & Williams, C. M. (1998) Effect of long-term olive oil dietary intervention on postprandial triacylglycerol and factor VII metabolism. Am. J. Clin. Nutr. 68: 552–560.

19. Kris-Etherton, P. M., Pearson, T. A., Wan, Y, Hargrove, R. L., Moriarty, K., Fishell, V. & Etherton, T. D. (1999) High-monounsaturated fatty acid diets lower both plasma cholesterol and triacylglycerol concentrations. Am. J. Clin. Nutr. 70: 1009–1015.

20. Keys, A., Anderson, J. T. & Grande, F. (1965) Serum cholesterol response to changes in the diet. IV. Particular saturated fatty acids in the diet. Metabolism 14: 776–787.

21. Mensink, R. P. & Katan, M. B. (1992) Effect of dietary fatty acids on serum lipids and lipoproteins. A meta-analysis of 27 trials. Arterioscler. Thromb. 12: 911–913.

22. Yu, S., Derr, J., Etherton, T. D. & Kris-Etherton, P. M. (1995) Plasma cholesterol-predictive equations demonstrate that stearic acid is neutral and monounsaturated fatty acids are hypocholesterolemic. Am. J. Clin. Nutr. 61: 1129–1139.

23. Clarke, R., Frost, C., Collins, R., Appleby, P. & Peto, R. (1997) Dietary lipids and blood cholesterol: quantitative meta-analysis of metabolic ward studies. Br. Med. J. 314: 112–117.

24. Sabaté, J., Bell, H.E.T. & Fraser, G. E. (1996) Nut consumption and coronary heart disease risk. In: Handbook of Lipids in Human Nutrition (Spiller, G. A., ed.), pp. 145–151. CRC Press, Boca Raton, FL.

25. Anderson, J. W., Smith, B. M. & Gustafson, N. J. (1994) Health benefits and practical aspects of high-fiber diets. Am. J. Clin. Nutr. 59 (suppl.): 1242S–1247S.

26. Anderson, J. W., Johnstone, B. M. & Cook-Newell, M. E. (1995) Metaanalysis of the effects of soy protein intake on serum lipids. N. Engl. J. Med. 333: 276–282.

27. Orth-Gomer, K., Mittleman, M. A., Schenck-Gustafsson, K., Wamala, S. P., Eriksson, M., Belkic, K., Kirkeeide, R., Svane, B. & Ryden, L. (1997) Lipoprotein(a) as a determinant of coronary heart disease in young women. Circulation 95: 329–334.

28. Eritsland, J., Arnesen, H., Berg, K., Seljeflot, I. & Abdelnoor, M. (1995) Serum Lp(a) lipoprotein concentrations in patients with coronary artery disease and the influence of long-term n-3 fatty acid supplementation. Scand. J. Clin. Lab. Investig. 55: 295–300.

29. National Cholesterol Education Program (1991) Report of the Expert Panel on Population Strategies for Blood Cholesterol Reduction. Circulation 83: 2154–2232.

30. Criqui, M. H. (1998) Triacylglycerol and cardiovascular disease. A focus on clinical trials. Eur. Heart J. 19: A36-A39.

31. Seman, L. J., McNamara, J. R. & Schaefer, E. J. (1999) Lipoprotein(a), homocysteine, and remnant like particles: emerging risk factors. Curr. Opin. Cardiol. 14: 186–191.

32. Headrick, L. A., Speroff, T., Pelecanos, H. I. & Cebul, R. D. (1992) Efforts to improve compliance with the National Cholesterol Education Program guidelines. Results of a randomized controlled trial. Arch. Intern. Med. 152: 2385–2387.

33. McManus, K., Roberts, C. & Manson, J. E. (1996) Appendix A, Nutrition guidelines for reducing coronary risk. In: Prevention of Myocardial Infarction (Manson, J. E., Ridker, P. M., Gaziano, J. M., Hennekens, C. H., eds.), pp. 529–540. Oxford University Press, New York, NY.