Scand J Clin Lab Invest 1998; 58: 11–18

Randomized controlled smoking cessation study: transient increase in plasma high density lipoprotein but no change in lipoprotein oxidation resistance

H. PRIEMÉ,* K. NYYSSÖNEN,¶ K. GRØNBÆK,† M. KLARLUND,‡ S. LOFT,* P. TØNNESEN,‡ J. T. SALONEN¶ & H. E. POULSEN*§

*Department of Pharmacology, University of Copenhagen, Copenhagen, †Department of Clinical Biochemistry and ‡Department of Pulmonary Medicine, Gentofte University Hospital, Copenhagen, §Department of Clinical Pharmacology Q7642, Rigshospitalet, National University Hospital, Copenhagen, Denmark and ¶Research Institute of Public Health, University of Kuopio, Kuopio, Finland

Priemé H, Nyyssönen K, Grønbæk K, Klarlund M, Loft S, Tønnesen P, Salonen JT, Poulsen HE. Randomized controlled smoking cessation study: transient increase in plasma high density lipoprotein but no change in lipoprotein oxidation resistance. Scand J Clin Lab Invest 1998; 58: 11–18.

Low plasma levels of high density lipoprotein (HDL) and high levels of low density lipoprotein (LDL) as well as smoking are known risk factors in coronary heart disease. It has been suggested that oxidative modification renders LDL atherogenic. We investigated the influence of smoking cessation on plasma lipid and lipoprotein levels and on the ability of lipoproteins to resist oxidation *in vitro* (lag time). A total of 182 healthy smokers who smoked more than 15 cigarettes per day were randomized to stop smoking (*smoking cessation group*, n=100) or to continue smoking for 4 weeks (*control group*, n=82). The *smoking cessation group* was followed up after 26 weeks. After 4 weeks, the HDL level had increased from mean±SD 1.36±0.34 to 1.48±0.40 mmol l^{-1} ($p<0.001$) in 62 successful quitters, while levels were unchanged in the *control group* (72 subjects in per-protocol analysis). However, after 26 weeks there was no change in HDL (1.34±0.36 vs. 1.36±0.35 mmol l^{-1}) in 29 subjects from the *smoking cessation group* who fulfilled the study. Plasma levels of very low density lipoprotein (VLDL), LDL, total cholesterol, triglycerides and oxidation resistance of VLDL+LDL did not show significant changes any time during the study for either group. Thus, plasma levels of lipids and lipoproteins as well as oxidation resistance of lipoproteins seem unaffected by smoking cessation for 26 weeks.

Key words: lag time; lipoprotein peroxidation; plasma lipids and lipoproteins; smoking

Henrik E. Poulsen, Department of Clinical Pharmacology Q7642, Rigshospitalet, Tagensvej 20, DK-2200 Copenhagen N, Denmark.

Smoking is a well-known risk factor in coronary heart disease [1, 2], but the exact mechanism remains unclear. A strong inverse correlation between the high density lipoprotein (HDL) plasma level and the risk of coronary heart disease, and a positive, but less pronounced, correlation between the low density lipoprotein (LDL) level and the risk of coronary heart disease have been found [3, 4]. In several cross-sectional studies, plasma levels of HDL have been found to be lower in smokers than in non-smokers [5–8]. Smoking cessation has been shown to change lipoprotein levels in agreement with a decreased risk for cardiovascular disease [9, 10].

Experimental evidence suggests that LDL is rendered atherogenic by oxidative modification [11]. Regnström *et al.* [12] reported an inverse relation between the ability of LDL to resist oxidation *in vitro* and the severity of coronary atherosclerosis in men. Cigarette smoke contains large amounts of oxidants [13–15]. In theory, the atherogenic effect of cigarette smoke could in part be caused by an increased oxidation rate of LDL mediated by free radicals in the smoke. However, Princen *et al.* [16] could not demonstrate any difference in oxidation resistance of LDL comparing smokers with non-smokers.

We performed a controlled randomized smoking cessation study measuring plasma concentrations of total cholesterol, triglycerides, very low density lipoprotein (VLDL), LDL and HDL, and oxidation resistance of haemin and hydrogen peroxide induced oxidation of VLDL+LDL at baseline and after 4 weeks. Subjects randomized to smoking cessation were followed up after 26 weeks.

SUBJECTS AND METHODS

Subjects

The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the local ethics committee. Signed informed consent was obtained from all subjects. Two hundred volunteers were recruited through advertisement in a local newspaper. Eligible subjects of both sexes (aged 35 to 65 years) had smoked at least 15 cigarettes per day for more than one year and declared motivation to stop. Exclusion criteria were presence of known disease; daily intake of drugs, including hormonal contraceptives; antioxidant supplements within the last month; and pregnancy or breast-feeding. At the first clinic visit, 182 subjects were included.

Randomization

After collection of baseline samples, 182 subjects were assigned by a computer-generated random number list either to stop smoking the next morning (*smoking cessation group*, n=100) or to continue usual smoking for another 4 weeks (*control group*, n=82). After the first 4 weeks, the subjects in the *control group* left the study. They were thereafter offered the chance to participate in a smoking cessation programme.

Smoking cessation

Subjects in the *smoking cessation group* received support by seven visits to the clinic during the 26 weeks of the study. Nicotine patches releasing a mean of 15 mg nicotine per 16 h (Nicorette®, Pharmacia AB, Helsingborg, Sweden) were supplied for 12 weeks, one patch per day [17], after which dosage was tapered to zero during a 4-week period. At the 26-weeks visit, only four quitters reported use of nicotine patches (2) or nicotine chewing gum (2). Only subjects from the *smoking cessation group* who stated being completely abstinent and who expired 10 ppm of carbon monoxide or less were included in per-protocol data analysis: of the 100 subjects, 85 were non-smokers after the first 4 weeks, and 41 were non-smokers *and* did show up at the 26-weeks visit.

Assessments

At the visits at baseline, after 4 weeks (both groups) and after 26 weeks (only the *smoking cessation group*), blood samples were collected after overnight fasting. The subjects' smoking history, height, body weight and blood pressure were recorded. The self-reported smoking status of the subjects was confirmed at each visit by measuring the carbon monoxide level in end-expiratory air with a carbon monoxide analyser (Bedfont Monitor, Sittingbourne, UK), levels of carbon monoxide exceeding 10 ppm indicating a recent smoker [17]. Plasma cotinine concentrations were measured by capillary gas

chromatography at the Bioanalytical Laboratory, Pharmacia AB, Helsingborg, Sweden. Plasma concentrations of total cholesterol, triglycerides, VLDL, LDL and HDL cholesterol were measured by routine clinical chemistry on a HITACHI 705 (Hitachi, Japan) at the Department of Clinical Biochemistry, Gentofte University Hospital, Copenhagen, Denmark [18, 19]. The coefficients of variation for the analyses are 1.96% for total cholesterol, 1.00% for triglycerides and 8.40% for HDL cholesterol.

Blood for VLDL+LDL oxidation resistance (lag time) measurements was collected in K_3-EDTA tubes. Plasma was immediately separated by centrifugation at 1500 *g* for 10 min at 4°C and frozen at −80°C. The analysis was effectuated at the Research Institute of Public Health, University of Kuopio, Finland within the next 3 months. Haemin and hydrogen peroxide induced VLDL+LDL oxidation has previously been described in detail [20].

Statistics

Continuous variables were tested for normal distribution using the Kolmogorov-Smirnov test. For the normally distributed variables, baseline values for the two groups were compared using the two sample Student's *t*-test. Values are given as means±SD. For variables not normally distributed, the baseline values for the two groups were compared using the Mann-Whitney U-test. These values are given as medians and ranges. Differences between values before and after 4 or 26 weeks in each group were also tested for normal distribution with the Kolmogorov-Smirnov test. When the differences were normally distributed, the changes were tested using the paired Student's *t*-test. The Wilcoxon matched pairs test was used to test the changes after 4 and 26 weeks when the distributions of the differences were not normal. As 12 paired tests of lipid and lipoprotein levels and lipoprotein oxidation characteristics (six variables tested for changes after both 4 and 26 weeks) were performed for the *smoking cessation group*, the p-values for changes in these values were corrected according to the Bonferroni method multiplying by 12. For the comparisons between the two groups and for the changes in the *control group*, the p-values were corrected by a factor of six, as differences were tested for six variables. Using normal approximations, 95% confidence intervals (CI) were calculated for the mean changes in values after 4 and 26 weeks. The difference in HDL levels between the two groups after 4 weeks was tested using ANCOVA adjusting for the baseline levels. Both per-protocol and intention-to-treat analyses were performed. A two-sided p-value of less than 0.05 was considered statistically significant.

RESULTS

Per-protocol data analysis

After 4 weeks, 62 subjects from the *smoking cessation group* and 72 subjects from the *control group* were included in per-protocol data analysis. In addition to the 15 subjects, who were excluded because they smoked, 23 subjects from the *smoking cessation group* were excluded at 4 weeks for the following reasons: Infections (9), antioxidant supplementation (9), not giving blood sample (2), treatment with non-steroid anti-inflammatory drugs (2) or other (1). Ten subjects from the *control group* were excluded from per-protocol analysis because of: Infections (3), withdrawal from the study (3), antioxidant supplementation (2), not giving blood sample (1) or other (1). At the 26-week follow-up, 29 subjects from the *smoking cessation group* were still non-smokers *and* had fulfilled all criteria throughout the study (Fig. 1).

The subjects included in per-protocol analysis in the *smoking cessation group* and the *control group* were similar with respect to baseline values of all variables (Tables I and II).

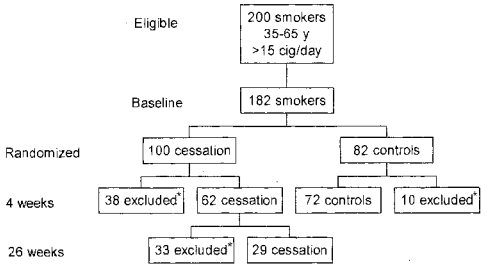

FIG. 1. Flow chart showing the study design. *Excluded because of acute disease, intake of any medicine or antioxidant supplements, or other deviations from the protocol, or for the *smoking cessation group,* resume of smoking.

TABLE I. Baseline characteristics of the subjects in per-protocol data analysis.

		Smoking cessation group	Control group
N		62	72
Gender, M/F		36/26	41/31
Age, years	Mean±SD	44.7±7.6	44.4±7.6
Pack years of cigarettes	Mean±SD	24.6±8.6	26.9±11.1
Alcohol consumption, units per week	Median (range)	11 (0–66)	8 (0–51)
Systolic blood pressure, mm Hg	Median (range)	130 (100–170)	120 (100–170)
Diastolic blood pressure, mm Hg	Median (range)	80 (70–105)	80 (60–110)

TABLE II. Values at baseline and after 4 weeks for subjects in per-protocol data analysis.

		Baseline		4 weeks	
		Smoking cessation group	Control group	Smoking cessation group	Control group
N		62	72	62	72
Cigarettes per day, #	Median (range)	20 (15–40)	23 (15–40)	0 (0–0)*	20 (4–40)
Carbon monoxide in expired air, ppm	Mean±SD	27±9	29±10	4±2*	29±12
p–Cotinine, ng ml^{-1}	Mean±SD	282±121	282±105	116±72*	289±103
Body weight, kg	Mean±SD	75.0±13.7	75.2±12.3	76.4±13.6*	75.4±12.2
Body mass index, kg m^{-2}	Mean±SD	24.7±3.6	24.3±3.1	25.2±3.6*	24.3±3.2
Total cholesterol, mmol l^{-1}	Mean±SD	5.9±1.2	6.0±0.9	5.9±1.2	6.0±1.0
Triglycerides, mmol l^{-1}	Median (range)	1.23 (0.54–3.59)	1.16 (0.44–4.39)	1.00 (0.42–5.01)	1.20 (0.46–4.94)
VLDL, mmol l^{-1}	Median (range)	0.58 (0.25–1.69)	0.55 (0.21–2.06)	0.46 (0.20–1.61)	0.56 (0.22–1.91)
LDL, mmol l^{-1}	Mean±SD	3.86±1.06	3.96±0.88	3.80±0.91	3.92±0.91
HDL, mmol l^{-1}	Mean±SD	1.36±0.34	1.39±0.39	1.48±0.40*	1.39±0.39
Lag time, min	Mean±SD	138±70	119±40	153±100	134±66

*The difference between the actual level and the baseline level is significant at the level $p<0.001$.

Body weight

There was a significant increase in mean body weight from 75.0 to 76.4 kg ($p<0.001$) in the *smoking cessation group* already 4 weeks after quitting. Body weight in the *control group* was without significant changes after 4 weeks. After 26 weeks, the 29 subjects who were still refraining from smoking and who were included in per-protocol analysis had increased their mean body weight from 75.0 to 79.3 kg (5.7%, $p<0.001$) (Table III). There were similar changes in body mass index (BMI; weight in kg divided by height in metres in second power).

Plasma lipids and lipoproteins

In the *smoking cessation group*, we found a significant increase in plasma HDL concentration of 9% (95% CI 5% to 13%, $p<0.001$,

TABLE III. Values at baseline and after 4 and 26 weeks for 29 subjects from *the smoking cessation group* who fulfilled the study.

		Baseline	4 weeks	26 weeks
N		29	29	29
Cigarettes per day, #	Median (range)	20 (15–40)	0 (0–0)**	0 (0–0)**
Carbon monoxide in expired air, ppm	Mean±SD	26±8	3±2**	1±2**
p–Cotinine, ng ml^{-1}	Mean±SD	265±115	94±55**	35±53**
Body weight, kg	Mean±SD	75.0±13.0	76.4±13.0**	79.3±12.9**
Body mass index, kg m^{-2} kg m^{-2}	Mean±SD	25.2±3.9	25.7±4.0**	26.7±4.1**
Total cholesterol, mmol l^{-1}	Mean±SD	5.5±1.0	5.6±1.0	5.4±1.0
Triglycerides, mmol l^{-1}	Median (range)	1.18 (0.63–2.52)	0.96 (0.42–2.38)	1.00 (0.51–3.08)
VLDL, mmol l^{-1}	Median (range)	0.55 (0.30–1.18)	0.45 (0.20–1.12)	0.47 (0.24–1.45)
LDL, mmol l^{-1}	Mean±SD	3.56±0.88	3.63±0.74	3.43±0.83
HDL, mmol l^{-1}	Mean±SD	1.34±0.36	1.47±0.42*	1.36±0.35
Lag time, min	Mean±SD	147±71	171±124	36±46

*The difference between the actual level and the baseline level is significant at the level p=0.024, when Bonferroni corrected with a factor 12.
**The difference between the actual level and the baseline level is significant at the level p<0.001.

Bonferroni corrected) after 4 weeks. This beneficial increase in HDL plasma concentration had disappeared after 26 weeks in the 29 subjects included in per-protocol analysis at the follow-up. The plasma HDL concentration did not change in the *control group* during the study. At 4 weeks, the difference in HDL plasma concentrations between the two groups was 0.12 mmol l^{-1} (95% CI 0.07 to 0.17 mmol l^{-1}), tested by ANCOVA and adjusted for the baseline levels (p=0.0022, Bonferroni corrected). There were no significant changes in the plasma concentrations of total cholesterol, triglycerides, LDL or VLDL for both groups throughout the study.

Oxidation resistance of VLDL+LDL

There were no significant changes in any group after 4 and 26 weeks in oxidation resistance of VLDL+LDL assessed by the lag time to the start of the oxidation reaction. After 4 weeks, there was a non-significant average change in lag time of 10% (95% CI −8% to 29%) in the *smoking cessation group* and of 13% (95% CI −0.4% to 27%) in the *control group*. The difference in change in lag time between the two groups was also non-significant (p=0.93). After 26 weeks, lag time had decreased non-significantly by 7% (95% CI −19% to 5%) in 29 successful quitters included in per-protocol analysis.

Intention-to-treat analysis

Intention-to-treat analysis at 4 weeks included 89 subjects from the *smoking cessation group* and 76 subjects from the *control group* who gave blood samples both at baseline and after 4 weeks. After 26 weeks, 55 subjects from the *smoking cessation group* gave blood samples and had done it throughout the study. The results from the intention-to-treat and the per-protocol analyses were similar.

DISCUSSION

The relative levels of the different plasma lipids and lipoproteins are considered of importance for the development of atherosclerosis [3, 4]. Smoking seems to raise plasma triglycerol and

TABLE IV. Changes in plasma lipid and lipoprotein levels and body weight following smoking cessation in five other studies (only statistically significant changes).

Reference	Subjects	Study period	Increase in body weight	Total cholesterol	Total triglycerides	HDL	LDL	VLDL
Stubbe *et al.* 1982 [23]	10 quitters	6 weeks	1.8 kg	No change	No change	29% increase	No change	
Rabkin 1984 [25]	35 quitters 72 non-quitters*	2–3 months	2.0 kg	No change	No change	9% increase	No change	
Gerace *et al.* 1991 [24]	1200 quitters 2000 non-quitters*	72 months	2.1 kg	No change		Increase	No change	No change
Allen *et al.* 1994 [9]	432 quitters 254 non-quitters†	6 weeks	2.0 kg	No change	11% increase	5% increase	3% decrease	
Terres *et al.* 1994 [10]	52 quitters 69 non-quitters*	24 weeks	3.4 kg		No change	10% increase after 12 weeks; after 24 weeks no change	6% decrease	
Nilsson *et al.* 1996 [26]	98 quitters 156 smokers (controls)	4 months	2.7 kg	No change	No change	11% increase	5% decrease, but also a significant 3% decrease in the control group	

*The non-quitters were subjects who did not succeed in quitting.
†249 other subjects withdrew prematurely.

LDL cholesterol and reduces HDL cholesterol [21]. In a meta-analysis of 54 published studies comparing plasma lipoprotein levels in smokers and non-smokers, HDL levels were on average 5.7% lower in smokers than in non-smokers [22]. Therefore it is interesting that we found a significant increase of 9% in HDL plasma levels after 4 weeks of smoking cessation while levels were unchanged in the *control group*. This finding is in accordance with the results from other smoking cessation studies summarized in Table IV [9, 10, 23–26]. The present study was designed as a randomized, controlled study thereby eliminating the possible bias that smokers who succeed in quitting may differ from those who relapse in respect to, e.g. plasma lipoprotein levels. However, in the present study, 26 weeks after smoking cessation the beneficial effect on the HDL level had disappeared. One explanation for this could be a change in diet or the increase in body fat associated with the considerable weight gain among the quitters. A corresponding rise and subsequent fall in HDL level was also seen in the smoking cessation study by Terres *et al.* [10]. After 72 months, the Multiple Risk Factor Intervention Trial Research Group found a significant increase in HDL levels among quitters but in that study diet was also a target for intervention [24].

Cross-sectional studies counting more than 10,000 subjects consistently show higher levels of HDL in non-smokers than in smokers [5–8]. However, data from these studies and the smoking cessation studies mentioned above are inconsistent in respect to the effect of smoking

on plasma levels of triglycerides, LDL, VLDL and total cholesterol [9, 10, 23–26]. In our study, these variables were not affected by smoking cessation.

The susceptibility of LDL to oxidation *in vivo* can be estimated from its susceptibility to oxidation *in vitro*. A widely accepted method is induction of LDL oxidation by copper ions [27] or induction of VLDL+LDL oxidation by haemin and hydrogen peroxide [20] and measurement of the time (lag time) that elapses before the lipid peroxidation process propagates.

We found no change in resistance (lag time) against haemin and hydrogen peroxide mediated VLDL+LDL oxidation following smoking cessation. This finding is in accordance with the work by Princen *et al.* [16], who found no difference in the resistance of LDL against copper-mediated oxidation comparing a group of smokers with a group of non-smokers. Also, Brown [28] found no change in lag time of copper-induced LDL oxidation in six smokers who stopped smoking for 84 h. However, it is possible that minor oxidative changes in LDL (minimally modified LDL), caused by cigarette smoke but not detected by the lag time measurement, play a role in accelerating atherosclerosis in smokers [16].

The outcome of the present study could possibly have been influenced by nicotine as the subjects in the *smoking cessation group* used nicotine patches during the first 16 weeks. However, in an earlier study, nicotine administration to healthy non-smokers has been shown not to influence plasma lipoprotein concentrations [29].

In conclusion, the increased risk of atherosclerosis in smokers could not be explained by an increased susceptibility to oxidation of lipoproteins in smokers using the present method. A beneficial increase in HDL levels after 4 weeks of smoking cessation disappeared after 26 weeks. During the 26 weeks, the quitters had a considerable weight gain. Other mechanisms, including endothelial dysfunction [30], seem to play a role in accelerating atherosclerosis in smokers. Still, important components of the mechanisms relating smoking to cardiovascular disease remain to be determined. Controlled and randomized smoking cessation studies are necessary tools in the future research in this field.

ACKNOWLEDGEMENTS

This study was supported by the Danish Environmental Research Programme, by the Finnish Academy and by British American Tobacco (H. Priemé and H. E. Poulsen). Nicotine patches were generously provided by Pharmacia AB, Helsingborg, Sweden

REFERENCES

1 Hammond EC. Smoking in relation to the death rates of one million men and women. National Cancer Institute Monographs 1966; 19: 127–204.
2 Doll R, Peto R, Wheatley K, Gray R, Sutherland I. Mortality in relation to smoking: 40 years' observations on male British doctors. Br Med J 1994; 309: 901–10.
3 Gordon T, Castelli WP, Hjortland MC, Kannel WB, Dawber TR. HDL as a protective factor against coronary heart disease. The Framingham Study. Am J Med 1977; 62: 707–14.
4 Miller NE, Førde OH, Thelle DS, Mjøs OD. The Trosmø Heart Study. High-density lipoprotein and coronary heart disease: a prospective case-control study. Lancet 1977; 1: 965–7.
5 Garrison RJ, Kannel WB, Feinleib M, Castelli WP, McNamara PM, Padgett SJ. Cigarette smoking and HDL cholesterol. The Framingham Offspring Study. Atherosclerosis 1978; 30: 17–25.
6 Criqui MH, Wallace RB, Heiss G, Mishkel M, Schonfeld G, Jones GTL. Cigarette smoking and plasma high-density lipoprotein cholesterol. Circulation 1980; 62 Suppl 4: 70–6.
7 Mjøs OD. Lipid effects of smoking. Am Heart J 1988; 115: 272–5.
8 Brischetto CS, Connor WE, Connor SL, Matarazzo JD. Plasma lipid and lipoprotein profiles of cigarette smokers from randomly selected families: enhancement of hyperlipidemia and depression of high-density lipoprotein. Am J Cardiol 1983; 52: 675–80.
9 Allen SS, Hatsukami D, Gorsline J and the Transdermal Nicotine Study Group. Cholesterol changes in smoking cessation using the transdermal nicotine system. Prev Med 1994; 23: 190–6.
10 Terres W, Becker P, Rosenberg A. Changes in cardiovascular risk profile during the cessation of smoking. Am J Med 1994; 97: 242–9.
11 Steinberg D, Parthasarathy S, Carew TE, Khoo JC, Witztum JL. Beyond cholesterol. Modifications of low-density lipoprotein that increase its atherogenicity. N Engl J Med 1989; 320: 915–24.
12 Regnström J, Nilsson J, Tornvall P, Landou C, Hamsten A. Susceptibility to low-density lipoprotein oxidation and coronary atherosclerosis in man. Lancet 1992; 339: 1183–6.
13 Church DF, Pryor WA. Free-radical chemistry of cigarette smoke and its toxicological implications. Environ Health Perspect 1985; 64: 111–26.

14 Pryor WA, Prier DG, Church DF. Electron-spin resonance study of mainstream and sidestream cigarette smoke: nature of the free radicals in gas-phase smoke and in cigarette tar. Environ Health Perspect 1983; 47: 345–55.
15 Kalra J, Chaudhary AK, Prasad K. Increased production of oxygen free radicals in cigarette smokers. Int J Exp Path 1991; 72: 1–7.
16 Princen HMG, van Poppel G, Vogelezang C, Buytenhek R, Frans JK. Supplementation with vitamin E but not beta-catotene in vivo protects LDL from lipid peroxidation in vitro. Effect of cigarette smoking. Arterioscler Thromb 1992; 12: 554–62.
17 Tønnesen P, Nørregaard J, Simonsen K, Säwe U. A double-blind trial of a 16-hour transdermal nicotine patch in smoking cessation. N Engl J Med 1991; 325: 311–5.
18 Bucolo G, David H. Quantitative determination of serum triglycerides by the use of enzymes. Clin Chem 1973; 19: 476–82.
19 Friedewald WT, Levy RI, Frederickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of preparative ultracentrifuge. Clin Chem 1972; 18: 499–502.
20 Nyyssönen K, Porkkala E, Salonen R, Korpela H, Salonen JT. Increase in oxidation resistance of atherogenic serum lipoproteins following antioxidant supplementation: a randomized double-blind placebo-controlled clinical trial. Eur J Clin Nutr 1994; 48: 633–42.
21 Freeman DJ, Packard CJ. Smoking and plasma lipoprotein metabolism. Clin Sci 1995; 89: 333–42.
22 Craig WY, Palomaki GE, Haddow JE. Cigarette smoking and serum lipid and lipoprotein concentrations: an analysis of published data. Br Med J 1989; 298: 784–8.
23 Stubbe I, Eskilsson J, Nilsson-Ehle P. High-density lipoprotein concentrations increase after stopping smoking. Br Med J 1982; 284: 1511–3.
24 Gerace TA, Hollis J, Ockene JK, Svendsen K. For the MRFIT Research Group. Smoking cessation and change in diastolic blood pressure, body weight, and plasma lipids. Prev Med 1991; 20: 602–20.
25 Rabkin SW. Effect of cigarette smoking cessation on risk factors for coronary atherosclerosis. Atherosclerosis 1984; 53: 173–84.
26 Nilsson P, Lundgren H, Söderström M, Fagerström K-O, Nilsson-Ehle P. Effects of smoking cessation on insulin and cardiovascular risk factors—a controlled study of 4 months' duration. J Intern Med 1996; 240: 189–94.
27 Esterbauer H, Striegl G, Puhl H, Rotheneder M. Continuous monitoring of in vitro oxidation of human low density lipoprotein. Free Rad Res Comms 1989; 6: 67–75.
28 Brown AJ. Acute effects of smoking on antioxidant status. J Nutr Biochem 1996; 7: 29–39.
29 Quensel M, Agardh C-D, Nilsson-Ehle P. Nicotine does not affect plasma lipoprotein concentrations in healthy men. Scand J Clin Lab Invest 1989; 49: 149–53.
30 Celermajer DS, Sorensen KE, Georgakopoulos D, Bull C, Thomas O, Robinson J, Deanfield JE. Cigarette smoking is associated with dose-related and potentially reversible impairment of endothelium-dependent dilation in healthy young adults. Circulation 1993; 88: 2149–55.

Received: 17 April 1997
Accepted: 15 October 1997