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Trans Fatty Acids, HDL-cholesterol, and Cardiovascular Disease. Effects of Dietary Changes on Vascular Reactivity

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Abstract: A high consumption of *trans* fatty acids increases the risk of cardiovascular disease (CVD). We investigeted whether this increase in risk was due to the decrease in serum HDL-cholesterol by *trans* fatty acids, because low concentrations of serum HDL-cholesterol also increase risk of CVD. Flow-mediated vasodilation (FMD) was used as an endpoint in dietary interventions that were designed to change the concentration of serum HDL-cholesterol within 4 weeks in healthy volunteers. Replacement of 10% of energy from saturated by *trans* fatty acids decreased serum HDL-cholesterol by 21 % and impaired FMD. However, a replacement of monounsaturated fats by carbohydrates did not impair FMD, although it decreased serum HDL-cholesterol by 13%. Acute postprandial impairments of FMD by either *trans* fats or saturated fats were not found, suggesting that long-term effects are responsible for the detrimental effect of *trans* fats on health. However, the role of serum HDL-cholesterol appears to be less than we expected.

Key words: flow-mediated vasodilation; dietary fat; blood lipids; controlled experiment

Introduction

Consumption of *trans* fatty acids increases risk of cardiovascular disease; presumably by 25% for each additional 2% of energy from these fatty acids [1]. This increase in risk is stronger than that of saturated fatty acids, despite similar effects on LDL-cholesterol. We investigated whether the decrease in HDL-cholesterol could explain the additional risk of cardiovascular disease with consumption of *trans* fatty acids. We used flow-mediated vasodilation, a marker of cardiovascular disease risk, as principal outcome of our studies. Flow-mediated vasodilation (FMD) is the percentage increase in the diameter of an artery after a provoked local increase in blood flow [2]. An impaired FMD appears to predict future coronary events [3]. FMD was measured non-invasively with high-resolution ultrasound in the brachial artery of the right arm.

Reproducibility and Sample Size

In a pilot study with 13 healthy young volunteers measured 6 times each we found a mean FMD of 5.6% of the baseline diameter. The within-subject standard deviation was 2.8 percentage points, so the corresponding coefficient of variation was 50%. From these data we estimated that we would need 32 volunteers in future cross-over designs to detect a treatment effect of 2 percentage points with 5% probability and a power of 80%.

Replacement of Saturated by *Trans* Fatty Acids Impaired FMD

The first study was a dietary controlled study with 10 men and 19 women in a cross-over design with two diets [4]. The volunteers had a mean (± SD) age of 30 ± 16 years, a bodyweight of 69 ± 9 kg, a body mass index of 22.5 ± 2.4 kg/m², and fasting total cholesterol of 5.1 ± 1.1 mmol/L. The aim of this study was to decrease serum HDL-cholesterol with *trans* fatty acids and to measure the effect on FMD. The *Trans*-diet provided 9.2% of energy from *trans* fatty acids, these were replaced by saturated fatty acids in the Sat-diet. Energy intake was 11 MJ/d on both diets. Pre-

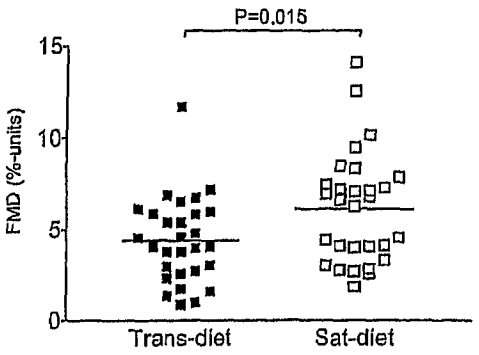

Fig. 1. Flow-mediated vasodilation of 29 subjects after the diet rich in trans fatty acids (■) and after the diet rich in saturated fatty acids (□). The subjects consumed both diets for 21-32 days in random order.

study total cholesterol was 5.1 ± 1.1 mmol/L After 4 weeks, serum HDL-cholesterol was 1.87 mmol/L after the Sat-diet and 1.48 mmol/L after the *Trans*-diet, a difference (95% CI) of 0.39 (0.28 to 0.50) mmol/L. The low HDL-cholesterol was parallelled by an impaired FMD, which was 4.4% after the *Trans*-diet; significantly lower than the 6.2% after the Sat-diet with a difference of 1.8%-points (0.4, 3.2). This result suggested that decreases in HDL-cholesterol impair FMD, and thereby increase risk of cardiovascular disease.

Replacement of Monounsaturated Fat by Carbohydrates Did Not Impair FMD

We sought confirmation for the impairment of FMD after a decrease in HDL-cholesterol in a subsequent cross-over study in which we lowered serum HDL-cholesterol by replacement of monounsaturated fat by carbohydrates [5]. The volunteers in this study were 11 men and 21 women with a mean (± SD) age of 27 ± 13 years, a bodyweight of 68 ± 9 kg, body mass index 22 ± 2.2 kg/m², and fasting total cholesterol of 4.6 ± 0.8 mmol/L. Although serum HDL-cholesterol was lowered by 0.21 (0.17, 0.26) mmol/L, FMD was not impaired. Instead, FMD was 4.1% after the diet rich in monounsaturated fat and 4.8% after the diet rich in carbohydrates, a difference of 0.7%-points (-0.6, 1.9). We checked whether the carbohydrate-rich diet, with its 124g larger fruit and 80g larger vegetable content, had reduced serum homocysteine, which may have improved FMD [6], but this was not the case. Thus, either the decrease in serum HDL-cholesterol had been too small to elicit an effect, or HDL-cholesterol was not causally related to FMD. In that case, the effect of *trans* fatty acids on FMD may have been mediated by other factors, such as serum paraoxonase activity.

Trans Fatty Acids Decrease Serum Paraoxonase Activity

We measured serum paraoxonase activity in serum samples of the first study. Paraoxonase is an HDL-bound enzyme that can hydrolyse oxidized lipids in LDL and HDL particles; and oxidized lipids may impair endothelial function as suggested by in vitro and animal studies. Moreover, serum paraoxonase activity is inversely related to risk of coronary heart disease in cross-sectional studies, and to vasoreactivity in patients with coronary heart disease. We found that serum paraoxonase activity was 196 μmol/L/min after the Sat-diet and 6% (2%, 10%) lower, or 185 μmol/L/min, after the *Trans*-diet [7]. This difference is about half that between smokers and nonsmokers, who also differ markedly in FMD. Thus, consumption of *trans* fatty acids for 4 weeks not only reduced serum HDL-cholesterol but also reduced the activity of serum paraoxonase. Whether this contributed to the impairment of FMD, however, remains to be determined. The last question we wished to answer was whether replacement of saturated fat by *trans* fat could impair FMD independent of changes in HDL-cholesterol. We did this by comparing postprandial effects of the two fats on FMD.

Postprandial FMD Not Different for *Trans* Fats and Saturated Fats

Based on reports by others we hypothesized that meals rich in saturated or *trans* fat would impair postprandial FMD, and based on the first study we expected to see a larger effect with *trans* fat. Twenty-five men aged 35 years and older were given oral fat loads (0.9-1.0 g fat/kg body weight) with saturated or *trans* fatty acids, and FMD was measured before and 3 h after the fat load [8]. In contrast to reports by others we found a vasodilation of 3.1% after the meals, which was higher than the 2.6% before the meals. Moreover, we found no differences in change in FMD between the two types of fat.

Serum paraoxonase activity parallelled the change in FMD, and was about 2% higher after the meals than before. In accordance with reports by others we found that postprandial changes in serum triacylglycerols were inversely related with changes in FMD.

Conclusion

In conclusion, we found that replacement of dietary saturated fatty acids by *trans* fatty acids for several weeks impaired flow-mediated vasodila-

Table 1. Concentration of serum lipids (mmol/L) after consumption of the two diets.

	oil diet mean	SD	carbohydrate diet mean	SD	Difference (95% CI)
total cholesterol	4.48	0.87	4.34	0.84	-0.14 (-0.27 to -0.01)
HDL cholesterol	1.66	0.39	1.44	0.35	-0.21 (-0.26 to -0. 17)
LDL cholesterol	2.45	0.65	2.42	0.67	-0.03 (-0.12 to 0.07)
triacylglycerols	0.81	0.41	1.03	0.52	0.22 (0.12 to 0.32)

Values are means ± SD. The 32 subjects consumed both diets for 3 1/2 weeks in random order. To convert values for total, HDL, and LDL cholesterol to milligrams per deciliter, multiply by 38.7. To convert triacylglycerols to milligrams per deciliter, multiply by 88.5.

tion, a marker of cardiovascular disease risk. This effect may have been caused by the concomitant decrease in serum HDL-cholesterol, but this was not confirmed in our second diet study in which we decreased HDL-cholesterol by a low-fat diet. The effects of *trans* fatty acids on FMD appeared to be long-term only: single meals rich in *trans* fatty acids or saturated fatty acids did not act differently on postprandial FMD. Serum paraoxonase activity parallelled changes in FMD in both studies with *trans* fatty acids, but whether this is causal remains to be determined. Thus, evidence for a causal role of HDL-cholesterol in coronary heart disease remains convincing but tentative.

References

1. Ascherio A, Katan MB, Stampfer MJ, Willett WC (1999) Trans fatty acids and coronary heart disease. N Engl J Med 340: 1994-1998
2. Celermajer DS, Sorensen KE, Gooch VM, Spiegelhalter DJ, Miller OI, Sullivan ID et al. (1992) Non-invasive detection of endothelial dysfunction in children and adults at risk of atherosclerosis. Lancet 340: 1111-1115
3. Neunteufl T, Heher S, Katzenschlager R, Wolfl G, Kostner K, Maurer G et al. (2000) Late prognostic value of flow-mediated dilation in the brachial artery of patients with chest pain. Am J Cardiol 86: 207-210
4. De Roos NM, Bots ML, Katan MB (2001) Replacement of dietary saturated fatty acids by trans fatty acids lowers serum HDL cholesterol and impairs endothelial function in healthy men and women. Arterioscler Thromb Vasc Biol 21: 1233-1237
5. De Roos NM, Bots ML, Siebelink E, Schouten E, Katan MB (2001) Flow-mediated vasodilation is not impaired when HDL-cholesterol is lowered by substituting carbohydrates for monounsaturated fat. Br J Nutr 86: 181-188
6. Tawakol A, Omland T, Gerhard M, Wu JT, Creager MA (1997) Hyperhomocyst(e)inemia is associated with impaired endothelium-dependent vasodilation in humans. Circulation 95: 1119-1121
7. De Roos NM, Schouten EG, Scheek LM, van Tol A, Katan MB (2002) Replacement of dietary saturated fat with trans fat reduces serum paraoxonase activity in healthy men and women. Metabolism 51: 1534-1537
8. De Roos NM, Siebelink E, Bots ML, van Tol A, Schouten EG, Katan MB (2002) Trans monounsaturated fatty acids and saturated fatty acids have similar effects on postprandial flow-mediated vasodilation. Eur J Clin Nutr 56: 674-679

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