plasma cholesterol levels were 817±28 (mean±SEM) mg/dL at 12 month-old and 750±55 mg/dL at 20 month-old, and were increased by about 250 mg/dL compared to the conventional WHHL rabbits.

Conclusions: Results of a selective breeding of WHHL rabbits, (1) the plasma cholesterol levels at the aged rabbits was increased; (2) the coronary atherosclerosis was advanced; (3) the coronary lesions was changed to macrophage-rich type; and (4) myocardial infarction was spontaneously developed. Newly developed WHHL rabbit is a valuable animal model for studies of myocardial infarction.

THE COFFEE DITERPENE CAFESTOL DECREASES BILE ACID SYNTHESIS BY DOWN-REGULATION OF CHOLESTEROL 7α-HYDROXYLASE IN APOLIPOPROTEIN E*3-LEIDEN MICE

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Cafestol, a diterpene present in unfiltered coffee brews, markedly increases serum cholesterol levels in humans. So far, no suitable animal model has been found to study the mechanism of action of this coffee diterpene. We determined the effects of cafestol on serum cholesterol, bile acid synthesis, liver lipids, VLDL triglyceride production and VLDL composition in female apoE*3-Leiden mice.

We fed 16 mice a diet containing 0.05% (w/w) or no cafestol. After 3 weeks of cafestol treatment, serum cholesterol was increased with 5.25±1.22 mmol/L ($p < 0.01$) compared to placebo treatment. This rise was mainly due to a rise in VLDL and IDL cholesterol. Activity and mRNA levels of hepatic cholesterol 7α-hydroxylase were suppressed by 57% and 58% (both $p < 0.05$), respectively. In addition, the amount of total bile acids in faeces was decreased with 37% ($p < 0.05$). Hepatic free and esterified cholesterol were not affected, but hepatic triglycerides were decreased by 52% ($p < 0.05$). The VLDL-triglyceride production rate was decreased compared to placebo (35.1±13.8 μmol/h/kg versus 63.1±17.5 μmol/h/kg). The number of VLDL particles was not affected but VLDL contained significantly more cholesteryl esters ($p < 0.05$). LDL receptor mRNA levels were decreased with 35%.

In conclusion, cafestol suppresses bile acid synthesis by downregulation of cholesterol 7α-hydroxylase in apoE*3-Leiden mice. This might be the cause of the decreased LDL receptor mRNA levels and an increased hepatic secretion of cholesteryl esters. Therefore, suppression of bile acid synthesis might provide an explanation for the cholesterol-raising effect of cafestol in humans.

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EFFECT OF DIETARY STANOLESTERS ON HEPATIC LIPID METABOLISM IN APOE*3-LEIDEN TRANSGENIC MICE

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Dietary plant sterols lower serum cholesterol (C) levels in humans and in hyperlipidemic rodents, mainly by inhibition of the intestinal cholesterol absorption. We used APOE*3-Leiden transgenic mice to investigate the consequences of this effect on plasma lipid levels and hepatic lipid metabolism. Five groups of 6 to 8 female and male mice, received a diet containing 0.25% cholesterol and 0.0, 0.25, 0.5, 0.75 or 1.0%(w/w) stanolesters for 9 weeks. Serum C values decreased dose-dependently, reaching maximal decreases of −37% in females and −24% in males ($P < 0.05$) after 4 weeks. There was no effect on triglyceride (TG) levels. The decrease in C was found in intermediate density lipoprotein (IDL)- and very low density lipoprotein (VLDL). Compared to the controls, the liver cholesterol ester (CE) contents of the 4 stanolester groups were significantly lowered by 28%, 31%, 48% and 50% for the females and by 29%, 49%, 41% and 52% for the males ($P < 0.05$). Hepatic TG contents were maximally decreased to 60% (females) and to 53% (males). The hypocholesterolemic mechanism of stanolesters was investigated in females and showed that stanolesters did not change the liver VLDL-TG production, but decreased the amounts of C incorporated in the VLDL particle (C −15%, CE −75%). Stanolesters increased the liver mRNA levels of the low density lipoprotein receptor (1.8 fold), cholesterol 7α-hydroxylase (1.9 fold) and HMGCoASynthase (1.7 fold) respectively. In conclusion, consumption of dietary stanolesters: (i) Lowered serum C in atherogenic lipoproteins in a dose-dependent way, but had no effect on TG. (ii) Lowered the hepatic CE pool dose-dependently, resulting in a reduced availability for VLDL production. (iii) May increase the hepatic uptake of lipoproteins and bile acid synthetic capacity. These changes may contribute to the hypolipidemic effects of stanolesters.

A COMPARATIVE STUDY OF POLICOSANOL VERSUS LOVASTATIN ON INTIMAL THICKENING IN RABBIT CUFFED CAROTID ARTERY

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Policosanol is a cholesterol-lowering drug. It have been demonstrated inhibitory effects on smooth muscle proliferation in rabbit cuffed carotid artery and in damaged arterial wall induced by forceps in rabbits. The present study was undertaken to compare the effects of policosanol and lovastatin on smooth muscle cell proliferation in the cuffed carotid artery of rabbits. Collars were placed around the left carotid for 7 and 15 days. The contralateral artery was sham operated. We studied eight experimental groups: two controls received vehicle for 7 and 15 days respectively, four other groups received policosanol at 5 and 25 mg Kg until sacrificed. Another group received lovastatin at 20 mg kg. Samples of arteries were examined by light and electron microscopy. To evaluate intimal thickening the cross-sectional area of intima and media were measured. Neointima was significantly reduced in policosanol-treated animals compared with controls. The reduction of neointima in lovastatin-treated animals was significantly lower than in policosanol-treated groups. The smooth muscle cell proliferation was studied by the immunohistochemical detection of proliferating cell nuclear antigen and an increase significant reduction was observed in policosanol-treated rabbits with respect to lovastatin-treated rabbits. It is concluded that policosanol has a protective effect moderately better than lovastatin on the neointima formation in this experimental model

GINKGO BILOBA EXTRACT INHIBITS SMOOTH MUSCLE PROLIFERATION IN VITRO AND REDUCES INTIMAL HYPERPLASIA AFTER BALLOON INJURY OF AORTA IN CHOLESTEROL-FED RABBITS

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Intimal hyperplasia, mainly smooth muscle cell proliferation and macrophage-derived foam cells accumulation, is one of major mechanisms responsible for postangioplasty restenosis. Ginkgo biloba extract has free-radicals scavenging properties and has been shown to exhibit antioxidant effect and platelet activating factor antagonism. In the present investigations, the effects of ginkgo biloba extract on proliferation of cultured smooth muscle cells and intimal hyperplasia after balloon injury of aorta in cholesterol-fed rabbits were studied. Ginkgo biloba extract at dose of 50, 100, 200, 500 and 1000g/ml were added to cultured smooth muscle cells. Radioactivity of thymidine uptake was determined. Male New Zealand white rabbits were fed with 2% cholesterol diet together with daily oral ingestion of ginkgo biloba extract (12 mg/Kg B.W.; n=8), or probucol (80 mg/Kg B.W.; n=9) or none (n=9) as control, for a total of 6 weeks. A balloon injury of abdominal aorta was performed at the end of 3rd week The animals were sacrificed and aortas were harvested at the end of 6th week. Intimal hyperplasia in the aorta was determined morphometrically. The results showed that proliferation of cultured smooth muscle cells was inhibited by ginkgo biloba extract at different concentrations. When compared with control group, intimal hyperplasia was significantly reduced by treatment with ginkgo biloba extract [intima(I)/media(M) thickness ratio: 1.070.06 vs 0.720.11, $p<0.05$; I/M area ratio: 0.780.05 vs 0.570.07, $p<0.05$] as well as by treatment with probucol [I/M thickness ratio: 1.070.06 vs 0.610.03, $p<0.01$; I/M area ratio: 0.780.05 vs 0.450.02, $p<0.01$]. In conclusion, ginkgo biloba extract significantly inhibited cultured smooth muscle cell proliferation and reduced intimal hyperplasia of the aorta after balloon injury in cholesterol-fed rabbits. These findings might have clinical implications in the prevention of postangioplasty restenosis.