Influence of flaxseed on serum and liver lipids in rats

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Rats were fed varying levels of flaxseed for three weeks. The diets were designed to provide 10%, 15%, and 20% flaxseed oil, respectively. The diets also contained 0.5% cholesterol and 0.2% cholic acid. Liver weight increased with increasing levels of fat. Serum cholesterol levels rose with increasing levels of fat, but HDL cholesterol and triglycerides were unaffected. Considering the presence of both cholesterol and cholic acid in the diet, the elevations of serum cholesterol were modest, which may reflect a potent hypocholesterolemic effect of linolenic acid. Liver total cholesterol levels fell with increasing levels of dietary flaxseed. Liver cholesteryl ester levels (%) were lower in rats fed higher doses of flaxseed, indicating reduced cholesterol deposition. Liver triglyceride levels were the same in all three dietary groups. Calculations of the serum plus liver cholesterol pool showed them to be 334 mg, 273 mg, and 252 mg for rats fed 10%, 15%, or 20% flaxseed oil, respectively. Since high levels of flaxseed reduced cholesterol deposition in liver, it might be useful to test in experimental atherosclerosis.

Keywords: cholesterol; flaxseed; linolenic acid; rats; serum lipids

Introduction

Flaxseed is a plant material rich in linolenic acid. The major fatty acids of flaxseed oil are: palmitic, 5.3%; stearic, 4.1%; oleic, 20.2%; linoleic, 12.7%; and linolenic, 53.3%. The P/S ratio of this oil is 7.25. The overall composition of whole flaxseed is: fat, 36%; protein, 24%; carbohydrate, 24%; fiber, 6%; water, 5.5%; and ash, 3.4%. Because of its high unsaturated fat content and its relatively high level of protein, we deemed it of interest to examine the effects of diets containing three levels of flaxseed on lipid levels in rats fed a hypercholesterolemic diet. The diets were designed to contain 10%, 15%, or 20% flaxseed oil, respectively.

Materials and methods

The diets were built around levels of flaxseed which would provide 10%, 15%, or 20% fat. The protein, carbohydrate, and fiber were augmented with peanut protein; corn starch-dextrose 1:1; and psyllium, respectively. The dietary compositions are presented in *Table 1*. Diets were prepared in pelleted form by Dyets, Inc. (Bethlehem, PA).

Male Wistar rats, average starting weight 206 g, were randomized into three groups of six rats each. They were maintained in individual cages in an air-conditioned room under a 12-hour dark-light schedule. Water and food were provided ad libitum. After three weeks, animals were killed by barbiturate injection. Blood was taken by heart puncture and the serum analyzed for cholesterol,[1] HDL cholesterol,[2] and triglycerides.[3] Livers were weighed and aliquots extracted in chloroform:methanol 2:1[4] and analyzed for total and esterified cholesterol[1,5] and triglycerides.[3]

Results and discussion

Our findings are summarized in *Table 2*. There were no significant differences in weight gain among the three groups, but liver weight increased as the level of fat in the diet increased. Serum cholesterol levels also increased with increasing fat levels, but the percentage of HDL cholesterol and triglycerides did not. In general, the presence of cholic acid in a cholesterol-containing diet is a powerful stimulus to hypercholesterolemia. The observation that the serum cholesterol levels in all three groups were within the normal range speaks to a strong hypocholesterolemic effect of linolenic acid. Going from low to high flaxseed oil serum

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Table 1 Composition of flaxseed-rich diets (g/100 g)

	Designation		
	Low	Medium	High
Flaxseed	27.8	41.7	55.6
Peanut protein	11.8	7.0	2.2
DL-methionine	0.3	0.3	0.3
Corn starch	23.2	19.0	14.9
Dextrose	23.2	19.1	14.9
Cholesterol	0.5	0.5	0.5
Cholic acid	0.2	0.2	0.2
Psyllium	8.3	7.5	6.7
Salt mix (AIN-76)	3.5	3.5	3.5
Vitamin mix (AIN-76A)	1.0	1.0	1.0
Choline bitartrate	0.2	0.2	0.2
Kjoules/100 g	90.0	94.5	99.0
% Fat	23.9	34.1	43.4
Protein	19.7	17.2	15.0
Carbohydrate	56.4	48.7	41.6

Table 2 Necropsy data. Rats fed special diets for 21 days (6/group)[a]

	Flaxseed Group		
	Low	Medium	High
Weight gain, g	86 ± 4	101 ± 8	97 ± 4
Liver weight, g	10.5 ± 0.44a	11.6 ± 0.41	12.2 ± 0.3a
Relative liver wt.	3.56 ± 0.11b	3.70 ± 0.11	3.96 ± 0.09b
Serum (mmol/L)			
Cholesterol	1.86 ± 0.13cd	2.28 ± 0.13c	2.35 ± 0.13d
HDL cholesterol, %	20.3 ± 1.74	18.6 ± 1.36	19.1 ± 1.57
Triglycerides	1.14 ± 0.14	1.19 ± 0.14	1.42 ± 0.19
Liver (g/100g)			
Cholesterol	3.09 ± 0.20ef	2.30 ± 0.24e	1.99 ± 0.20f
% Ester	75.7 ± 1.77gh	65.0 ± 2.73g	63.0 ± 3.65h
Triglycerides	6.89 ± 0.41	5.86 ± 0.60	6.92 ± 0.63

[a] Values in horizontal row bearing same letter are significantly different, $P < 0.05$. All values ± SEM.

cholesterol levels increased by 22% and 26% and LDL-cholesterol increased by only 2.1% and 1.5% in the medium and high flaxseed oil groups, respectively. These findings may be related to the increasing fat level of those two diets. Liver triglyceride levels were not significantly different among the three groups, but as the level of flaxseed in the diet increased, the level of liver cholesterol fell as did the amount of cholesteryl ester, an indicator of reduced cholesterol deposition. We calculated the actual levels of cholesterol present in serum and liver (*Table 3*), which show that as the level of dietary flaxseed rose, the total liver plus serum cholesterol pool fell by 18.6% going from 10% to 15% fat and by 7.7% going from 15% to 20% fat. Increasing levels of flaxseed were shown to decrease deposition of dietary cholesterol in the livers of cholesterol and cholic acid-fed rats. Flaxseed would appear to be a likely candidate for testing in experimental atherosclerosis.

Table 3 Serum plus liver pool (mg) of cholesterol in rats fed special diets[a]

	Flaxseed Group		
	Low	Medium	High
Serum	6.3 ± 0.5ab	8.1 ± 0.5a	8.7 ± 0.6b
Liver	328 ± 31	265 ± 28	244 ± 29
Serum plus liver	334 ± 32	273 ± 27	252 ± 29

[a] Values in horizontal row bearing same letter are significantly different, $P < 0.05$. All values ± SEM.

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