Similar serum lipoprotein reductions by lipid-lowering diets with different polyunsaturated:saturated fat values

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1. Lipid-lowering diets enriched in polyunsaturated fat decrease the serum cholesterol in hyperlipoproteinaemia, usually by reducing both the low-density-lipoprotein (LDL) and high-density-lipoprotein (HDL) cholesterol concentrations. The aim of the present study was to investigate whether the effects on LDL could be maintained but those on HDL cholesterol be diminished by reducing the ratio, polyunsaturated:saturated fat (P:S) of the diet.

2. Twenty hyperlipoproteinaemic patients (six with type IIa, eight with type IIb and six with type IV) in a metabolic ward were given two fat-modified diets during two consecutive 3-week periods in a randomized order. The diets were identical with regard to nutrient composition but differed with regard to the P:S values, which were 2·0 and 1·3 respectively.

3. The lipoprotein-lipid composition and serum apolipoprotein concentrations were similar at the end of the two dietary periods in type IIa and type IV patients but in type IIb patients a more pronounced reduction of the LDL-cholesterol concentration by 9% ($P < 0.05$) was achieved on the diet with the higher P:S value. The HDL-cholesterol did not differ significantly.

4. The results indicate that increasing the P:S value of lipid-lowering diets from 1·3 to 2·0 does not offer a great advantage with regard to the lipoprotein-lipid reductions achieved in moderate hyperlipoproteinaemia.

Increased serum lipid levels can be reduced by treatment with lipid-lowering diets. Recent studies (Shepherd *et al.* 1978; Ernst *et al.* 1980; Shepherd *et al.* 1980; Vessby *et al.* 1980*a*, *b*; Lewis *et al.* 1981; Schaefer *et al.* 1981) have indicated that the serum cholesterol reduction achieved corresponds not only to a reduction of the concentration of cholesterol in the low-density-lipoproteins (LDL), but also often to a reduction of the level of cholesterol in the high-density-lipoproteins (HDL). The reduction of LDL-cholesterol is presumably beneficial. The decrease of the HDL-cholesterol concentration is probably less desirable as judged from epidemiological findings indicating that a high HDL-cholesterol concentration is associated with a low incidence of coronary heart disease (Gordon *et al.* 1977).

In a study by Shepherd *et al.* (1978), healthy men were given a lipid-lowering diet with a very high value for the ratio between the contents of polyunsaturated and saturated fatty acids (P:S). This was associated with a reduction of the HDL-cholesterol which was more pronounced than in other studies (Ernst *et al.* 1980; Vessby *et al.* 1980*a*, *b*; Lewis *et al.* 1981; Schaefer *et al.* 1981) involving diets with more moderately elevated P:S values. This suggested that the reduction achieved might be proportional to the P:S value of the diet.

We have demonstrated in earlier studies (Vessby *et al.* 1980*a*) that the reductions of HDL-cholesterol and LDL-cholesterol during treatment with lipid-lowering diets show different time-courses. This indicates that the decreases of HDL-cholesterol and LDL-cholesterol are due to different mechanisms and thus may show different responses to diets with high and low P:S values respectively.

In the present study the patients, in a metabolic ward, were treated with two diets with high and moderate contents of polyunsaturated fat in a randomized order. The aim of the study was to investigate whether the effects on LDL-cholesterol could be maintained but

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Table 1. *Composition (energy %) of lipid-lowering diets*

Dietary polyunsaturated: saturated fat value	Protein	Carbohydrates	Fat (mg/10 MJ)				
			Total	S	M	P	Chol
2·0	20	45*	35	8·0	10·0	16·0	228
1·3	20	45*	35	11·0	10·0	14·0	230

S, Saturated; M, monounsaturated; P, polyunsaturated; Chol, cholesterol.
* Refined sugar restricted.

Table 2 *The relative fatty acid composition of the lipid-lowering diets with polyunsaturated:saturated fat (P:S) values of 1·3 and 2·0*

(Mean values with standard errors were calculated from the average value of each day of the week. For each daily menu, five to eight determinations were done)

Fatty acids	P:S value of diet					
	1·3			2·0		
	Mean	SEM	Range	Mean	SEM	Range
12:0	6·2	0·3	5·7–7·2	1·0	0·1	0·9–1·4
14:0	3·8	0·3	3·3–4·3	1·9	0·2	1·5–2·6
16:0	15·6	0·2	15·1–16·5	14·2	0·2	13·4–15·3
18:0	6·0	0·2	5·6–6·2	7·0	0·1	6·6–7·3
Total saturated	31·6	0·5	30·4–33·5	24·2	0·4	22·7–25·9
16:1ω7	1·0	0·1	0·8–1·3	1·1	0·1	1·0–1·3
18:1ω9	26·3	0·2	25·5–26·9	27·4	0·2	26·7–27·8
Total monounsaturated	27·3	0·2	26·7–28·0	28·5	0·2	27·7–29·2
18:2ω6	37·4	0·5	35·4–39·1	44·3	0·5	42·7–46·5
18:3ω3	3·7	0·2	3·1–4·3	2·8	0·2	2·2–3·3
Total polyunsaturated	41·1	0·6	38·5–43·0	47·3	0·5	45·9–49·4
P:S	1·3	0·0	1·2–1·4	2·0	0·0	1·8–2·2

those on HDL-cholesterol be diminished by reducing the P:S value from 2·0 to 1·3. An optimal lipid-lowering diet would presumably cause a reduction of LDL-cholesterol without a concomitant decrease (or even better an increase) of the HDL-cholesterol concentration.

MATERIALS AND METHODS

Design of the study

During two consecutive 3-week periods in a metabolic ward the patients were treated with two lipid-lowering diets with P:S values of 1·3 or 2·0 in a random order. The serum lipoprotein patterns were studied at the end of the two dietary periods. Three or four times weekly the fasting serum triglyceride and cholesterol concentrations, the body-weights and the fatty acid compositions of the serum lipid esters were determined.

Diets

The lipid-lowering diets used in this study were based on ordinary Swedish food and planned as a 1-week menu. Their compositions are presented in Table 1 and have been described

in detail elsewhere (Boberg *et al.* 1981). The compositions of the two diets were identical with regard to the proportion of fat, carbohydrates and protein and cholesterol content, but different with regard to the fat quality. The difference in fat quality was achieved by using different margarines. Otherwise the two diets were the same. The P:S values of the two diets were 1·3 and 2·0. The relative fatty acid compositions of the two diets are given in Table 2. The calculated fibre contents of the two diets (mean and SD) were identical (25·3 (3·4) g/10 MJ) as were the contents of vitamin C (191 (58) mg/10 MJ). The energy intake was calculated individually to assure a steady body-weight.

Patients

Twenty patients took part in the study: six patients with hyperlipoproteinaemia (HLP) type IIa, eight patients with HLP type IIb and six patients with HLP type IV according to the World Health Organization criteria (Beaumont *et al.* 1970). The type IIa group consisted of three men (age range 30–60 years) and three women (age range 50–59 years), the type IIb group of four men (age range 51–59 years) and four women (age range 41–65 years) and the type IV group of four men (age range 51–59 years) and two women (58 and 66 years old). The patients were admitted to the clinic because of hyperlipoproteinaemia or atherosclerotic disease or both. No patient had secondary hyperlipoproteinaemia.

Laboratory methods

The serum triglyceride and the serum cholesterol concentrations were determined in propanol extracts by semi-automatic methods in a Technicon AutoAnalyzer type II (Rush *et al.* 1971) in whole serum and in the top fraction (corresponding to the very-low-density lipoproteins (VLDL)) and the bottom fraction at density 1·006 after preparative ultracentrifugation (Havel *et al.* 1955). LDL were precipitated from the bottom fraction, using a heparin–manganese chloride solution and the HDL-lipid concentrations were determined in the supernatant fraction (Burstein & Samaille, 1960). A detailed description has been presented elsewhere (Vessby *et al.* 1980*a*).

The concentrations of apolipoprotein (apo) B, A-I and A-II were determined by electroimmunoassay as presented elsewhere (Vessby *et al.* 1980*a*). The apolipoprotein concentrations in serum are expressed as arbitrary units (A.U.) relative to the concentration in a reference serum, 100 A.U., obtained from a pool of healthy blood donors.

The fat content of the food was determined after extraction with chloroform–methanol and measured by weighing as described elsewhere (Boberg *et al.* 1981). The fatty acid compositions of the food and the blood plasma lipids were determined by gas–liquid chromatography (Boberg *et al.* 1981).

Statistics

Means and standard errors were calculated by ordinary methods. Confidence-interval tests of the hypothesis of equal means for the difference between the two treatments are, for each variable, based on the standard error for the difference and the *t* distribution.

RESULTS

Effects of 3-weeks dietary treatment on serum lipid concentrations

After the first 3 weeks on lipid-lowering diets with P:S values of 1·3 or 2·0, the serum triglycerides had decreased by 21, 33 and 23% in the type IIa, IIb and IV patients respectively. During the same period the serum cholesterol concentrations were reduced by 18, 16 and 21% in patients with HLP types IIa, IIb and IV respectively.

Table 3. *Comparisons of serum lipid concentrations in patients with hyperlipoproteinaemia (HLP) during treatmemt with lipid-lowering diets with polyunsaturated : saturated fat (P : S) values of 1·3 and 2·0*

(Mean values were calculated from the average value of serum lipid concentrations taken on three different days at the end of the diet periods for each patient; no. of patients in parentheses)

		Serum lipid concentrations (mmol/l)			
			P:S value of diet		Mean difference between treatments and 95% confidence interval
	HLP type	At admission	1·3	2·0	
Triglycerides	IIa	1·60	1·59	1·60	−0·01 ± 0·23 (6)
	IIb	4·07	2·66	2·69	−0·04 ± 0·28 (8)
	IV	5·35	3·98	4·19	−0·21 ± 0·34 (6)
Cholesterol	IIa	9·07	7·27	7·24	0·03 ± 0·64 (6)
	IIb	8·67	7·15	6·74	0·41 ± 0·56 (8)
	IV	7·92	6·20	6·08	0·12 ± 0·42 (6)

Comparisons of the effects of treatment with lipid-lowering diets with P : S values of 1·3 and 2·0

Body-weight. The mean (with SEM) body-weight of the patients with HLP type IIa at admission was 71·0 (2·9) kg, of the type IIb patients 71·3 (3·3) kg and of the type IV patients 69·2 (4·4) kg. On average the body-weight did not change significantly in any of the three groups during the study.

Serum lipid concentrations (*Table 3*). The serum cholesterol and the serum triglyceride concentrations were not significantly different when compared after treatment with diets with P:S values of 1·3 and 2·0.

Serum lipoprotein concentrations (*Table 4*). The concentrations of the serum lipoprotein lipids and apolipoproteins were very similar after treatment with the diet with a P:S value of 1·3 and that with a P:S value of 2·0. The LDL-cholesterol concentration in the type IIb patients only showed a further significant reduction of 9% ($P < 0·05$) when the P:S value was 2·0.

Fatty acid composition (*Table 5*). Similar changes in the fatty acid composition of the plasma lipid esters were seen in types IIa, IIb and IV. A significant increase in the polyunsaturated fatty acids (linoleic and arachidonic acids), and a decrease in the saturated fatty acids and oleic acid were observed in most of the lipid-ester fractions when the values at admission were compared with those obtained after 3 weeks on the lipid-lowering diets. Slight but significant differences in the fatty acid compositions were observed between the results after 3 weeks on the diet with a P:S value of 1·3 and 3 weeks on the diet with a P:S value of 2·0. Thus, increasing the P:S value from 1·3 to 2·0 caused a slight further increase in linoleic acid content in the cholesterol esters and the triglycerides and a decrease of the myristic and oleic acids in all plasma lipid esters.

DISCUSSION

The reduction in serum cholesterol and serum triglycerides during treatment was similar to that reported earlier with lipid-lowering diets (Shepherd *et al.* 1978; Shepherd *et al.* 1980; Ernst *et al.* 1980; Vessby *et al.* 1980*a*, *b*; Lewis *et al.* 1981; Schaefer *et al.* 1981; Chait *et al.* 1974) indicating an adequate composition of the diets. Only in the type IIb patients (Table 4) was a more pronounced reduction in the LDL concentration achieved on the diet with a P:S value of 2·0 than on the diet with a P:S value of 1·3. Thus is seems that varying the

Table 4. *Comparison of serum lipoprotein concentrations (mmol/l) and serum apolipoprotein concentrations (arbitrary units) in patients with hyperlipoproteinaemia type IIa* (n 6), *IIb* (n 8) *and IV* (n 6) *with lipid-lowering diets with polyunsaturated: saturated fat (P: S) values of 1·3 and 2·0*

(Mean values, mean difference between the two diets and 95% confidence interval for the differences are given)

	P:S value of diet		Mean difference between treatments and 95% confidence interval
	1·3	2·0	
Type IIa			
VLDL TG	0·71	0·73	−0·02±0·11
VLDL Chol	0·42	0·33	0·09±0·13
LDL TG	0·55	0·51	0·04±0·18
LDL Chol	5·51	5·49	0·02±0·26
HDL TG	0·22	0·21	0·01±0·03
HDL Chol	1·22	1·22	0·00±0·13
Apo B	175	173	2±42
Apo A-I	111	114	−3±7
Apo A-II	117	118	−1±8
Type IIb			
VLDL TG	1·66	1·86	−0·19±0·33
VLDL Chol	0·75	0·80	−0·05±0·12
LDL TG	0·64	0·58	0·06±0·08
LDL Chol	5·35	4·86	0·50±0·46*
HDL TG	0·20	0·22	−0·02±0·06
HDL Chol	0·89	0·88	0·01±0·08
Apo B	182	175	7±12
Apo A-I	97	96	1±5
Apo A-II	107	106	1±4
Type IV			
VLDL TG	3·03	2·91	0·11±0·45
VLDL Chol	1·20	1·21	−0·01±0·02
LDL TG	0·53	0·53	−0·00±0·16
LDL Chol	4·14	4·14	−0·00±0·45
HDL TG	0·25	0·24	0·01±0·12
HDL Chol	0·84	0·79	0·05±0·13
Apo B	162	153	9±29
Apo A-I	95	92	3±8
Apo A-II	96	93	3±4

VLDL, very-low-density-lipoprotein; LDL, low-density-lipoprotein; HDL, high-density-lipoprotein; TG, triglycerides; Chol, cholesterol; Apo, apolipoproteins.

* Significant change, $0{\cdot}01 < P < 0{\cdot}05$.

dietary P:S value had no major effect on the extent of serum lipoprotein reduction during treatment. In particular, our hypothesis that the HDL reductions might be less pronounced with a diet with a lower P:S value was not confirmed.

Interestingly enough, it was shown that the linoleic acid contents of the cholesterol esters and phospholipids were very similar at the end of the two different dietary periods (Table 5). A somewhat greater difference was seen with regard to the composition of the triglycerides. While the fatty acid composition of the triglycerides merely reflects the dietary composition during the last days before testing, the fatty acid composition of the cholesterol esters and phospholipids changes more gradually during a 2–3 week period and thus reflects the average dietary composition during a longer time-period (Boberg *et al.* 1978). Compared with those at admission, the linoleic acid contents of cholesterol esters and phospholipids

Table 5. *Fatty acid composition (mmol/mol) of serum triglycerides (TG), cholesterol esters (CE) and phospholipids (PL) of twenty patients, on admission to hospital and on lipid-lowering diets with polyunsaturated:saturated fat (P:S) values of 1·3 and 2·0*

Fatty acid		Fatty acid composition			
		At admission	P:S value of diet		Mean difference between treatments and 95% confidence interval
			1·3	2·0	
14:0	TG	2·54	1·96	1·66	0·30±0·17**
	CE	1·08	0·74	0·60	0·15±0·08**
	PL	0·44	0·37	0·30	0·06±0·03***
16:0	TG	27·03	22·63	21·76	0·87±0·71*
	CE	12·37	10·89	10·52	0·37±0·29*
	PL	33·94	31·74	31·68	0·06±0·51
16:1ω7	TG	6·22	5·39	5·21	0·19±0·29
	CE	4·61	2·56	2·22	0·34±0·27*
	PL	1·53	1·36	1·30	0·06±0·10
18:0	TG	4·36	3·60	3·45	0·15±0·12*
	CE	1·18	0·99	1·05	−0·05±0·11
	PL	16·64	16·68	17·09	−0·41±0·46
18:1ω9	TG	39·29	33·99	32·52	1·47±0·93*
	CE	19·59	13·48	12·97	0·51±0·45*
	PL	13·18	10·76	10·41	0·36±0·31*
18:2ω6	TG	18·35	30·04	33·33	−3·29±1·53***
	CE	55·56	65·63	66·93	−1·30±0·64***
	PL	24·95	29·14	29·72	−0·57±0·46*
18:3ω3	TG	1·29	1·32	0·99	0·32±0·17***
	CE	0·64	0·35	0·30	0·05±0·14
	PL	0·34	0·14	0·11	0·03±0·06
20:4ω6	TG	0·92	1·07	1·09	−0·01±0·08
	CE	4·98	5·35	5·42	−0·07±0·27
	PL	9·07	9·82	9·40	0·42±0·58

Mean values were significantly different from those for a P:S value of 1·3: *$P < 0·05$, **$P < 0·01$, ***$P < 0·001$.

were highly significantly increased after giving the diet with a P:S value of 1·3 for 3 weeks (Table 5). There was a significant, but very slight, increase in the cholesterol ester content of linoleic acid when comparing the fatty acid compositions at the end of treatment with the diet with a P:S value of 2·0 with that on the diet with a P:S value of 1·3. There was no significant increase of the content of 18·2ω6 in the phospholipids.

It has been suggested that the fatty acid composition of the LDL, as reflected by the fatty acid composition in the cholesterol esters in plasma, might affect the elimination rate of the lipoproteins from plasma (Deckelbaum *et al.* 1977). The reduction in LDL-cholesterol during treatment with polyunsaturated fat has been suggested to be due to decreased synthesis (Deckelbaum *et al.* 1977; Gordon *et al.* 1977; Boberg *et al.* 1981). An increased catabolic rate of LDL may be due to an altered membrane fluidity of LDL (Morrisett *et al.* 1977) or changed properties of the receptors (Jackson *et al.* 1978). If we assume that the fatty acid composition of the lipoproteins is related to the metabolic fate of these particles the very modest increase in the polyunsaturated fat content of plasma lipid esters when increasing the P:S value from 1·3 to 2·0 may explain the lack of major differences in the LDL concentration. There seems to be a saturation point, at least during relatively

short-term studies, with regard to the possible incorporation of polyunsaturated fatty acids into plasma lipoproteins.

In conclusion, the present study indicates that increasing the P:S ratio of lipid-lowering diets from 1·3 to 2·0 does not offer any great advantages with regard to lipoprotein-lipid reduction in patients with moderate hyperlipoproteinaemia. The situation may be different in familial hypercholesterolaemia with massively-increased LDL-cholesterol concentrations. The effects on LDL- and HDL-cholesterol were not dissociated by decreasing the amount of polyunsaturated fats in the present study. It is possible that advice given to patients with moderate hyperlipoproteinaemia may be modified with regard to the amount of polyunsaturated fat, thus giving the patients a greater freedom in the choice of foodstuffs which would make it easier to adhere to the prescribed diet.

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