

# Effects of cholic acid on the metabolism of endogenous plasma triglyceride and on biliary lipid composition in hyperlipoproteinemia

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**Abstract** Plasma lipids, endogenous triglyceride kinetics, and biliary lipid composition were determined in 13 patients with primary hyperlipoproteinemia (HLP) before and during treatment with cholic acid (15 mg/kg body weight/day for 3 months). In patients with type IIa HLP (n = 5), no consistent effects were seen on fasting plasma lipids or triglyceride turnover determined over a 10-hour period. Plasma triglyceride concentration was decreased in six of the eight patients with type IV HLP. Apparent triglyceride production rate was not significantly changed during treatment, but a decrease was seen in the five patients with initially elevated triglyceride synthesis. Treatment with cholic acid resulted in an increased proportion of bile acids and a decreased proportion of cholesterol and phospholipids in fasting duodenal bile; bile saturation with cholesterol was not significantly reduced. The results are discussed in relation to previous studies on the integrated regulation of bile acid and triglyceride metabolism, and it is concluded that cholic acid and chenodeoxycholic acid exert different effects on plasma triglyceride metabolism and on biliary lipid composition in HLP.—**Angelin, B., and B. Leijð.** Effects of cholic acid on the metabolism of endogenous plasma triglyceride and on biliary lipid composition in hyperlipoproteinemia. *J. Lipid Res.* 1980. **21**: 1–9.

**Supplementary key-words** bile acid · cholesterol · chenodeoxycholic acid · deoxycholic acid · very low density lipoprotein

In patients with primary hyperlipoproteinemia (HLP), changes in plasma triglyceride turnover appear to be associated with changes in the metabolism of cholesterol. Hyperlipoproteinemia type IV (1) is often linked to an increased production of cholesterol (2) and bile acids, especially cholic acid (3). The formation of bile acids is positively correlated to the synthesis of endogenous plasma triglycerides in patients with type IIa and type IV HLP when studied under basal conditions (4). Furthermore, a reduction of triglyceride production in patients with type IV HLP by means of weight reduction (5), clofibrate (6, 7), or nicotinic acid treatment (7) is accompanied by a decrease in bile acid formation (3, 8, 9).

In a previous study (10) we reported that some experimental conditions primarily affecting chole-

sterol and bile acid biosynthesis also influence the metabolism of plasma triglycerides. Thus, stimulation of bile acid synthesis by treatment with cholestyramine was associated with an augmented synthesis of plasma triglycerides in HLP type IIa. Inhibition of bile acid formation by means of chenodeoxycholic acid feeding gave a decreased triglyceride synthesis in both type IIa and type IV HLP (10). In the present study, the effect of cholic acid treatment on the metabolism of plasma triglycerides in HLP type IIa and type IV was studied. In addition, the lipid composition and cholesterol-solubilizing capacity of bile were determined before and during treatment.

## METHODS

### The patients

The study comprised 13 patients, 5 with type IIa and 8 with type IV HLP. Diagnostic criteria and outlines for lipoprotein patterning according to WHO recommendations (1) have been given previously (3, 4). No evidence of intestinal, hepatic or renal disease, hyper- or hypothyroidism, or addiction to alcohol or narcotics was found in any of the subjects. An oral cholecystogram was obtained in all non-cholecystectomized subjects. Clinical diagnoses and basal data are listed in **Table 1**. Genetic analysis (11) could be carried out in some of the patients and revealed familial hypercholesterolemia in patients 1 and 3, familial combined hyperlipidemia in patient 8, and familial hypertriglyceridemia in patients 9 and 13. Patients 4 and 5 obviously had HLP of polygenetic origin. Some of the patients were being treated

Abbreviations: GBD, gallbladder disease (cholelithiasis, cholecystitis, cholecystectomy); HLP, hyperlipoproteinemia; HMG-CoA reductase, 3-hydroxy-3-methylglutaryl CoA reductase; VLDL, very low density lipoprotein.

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TABLE 1. Basal data on individual patients before and during treatment with cholic acid.

Patient, Type of HLP	Sex, Age	Body Weight			Plasma Cholesterol <sup>b</sup>	Plasma Triglyceride <sup>b</sup>	Previous History, Present Symptoms <sup>c</sup>
		Before	During	Before Relative <sup>a</sup>			
		yr	kg	%			
1. KR IIa	M 55	72	72	101	10.2	1.8	FHC, IHD
2. AL IIa	F 62	45	45	88	8.2	1.6	
3. KL IIa	M 65	81	81	110	9.5	1.6	FHC
4. AP IIa	M 65	80	80	114	8.9	1.2	IHD
5. LS IIa	F 62	71	71	103	10.0	1.8	
Mean ± SEM	58 ± 4	68 ± 6	68 ± 6	103 ± 4	9.4 ± 0.4	1.6 ± 0.1	
6. TB IV	M 50	70	72	100	6.3	2.2	GBD
7. TW IV	M 56	73	72	106	7.6	3.7	HT
8. BA IV	M 49	87	88	118	5.8	2.9	FCH
9. EC IV	F 67	59	59	95	5.8	4.3	FHT, IHD, GBD, HT
10. KS IV	M 40	77	77	115	6.5	2.8	IHD
11. NS IV	M 36	83	83	109	5.0	3.4	
12. NG IV	M 60	85	83	129	6.7	4.3	
13. HL IV	M 40	76	77	89	5.3	2.1	FHT
Mean ± SEM	50 ± 4	76 ± 3	76 ± 3	108 ± 5	6.2 ± 0.3	3.2 ± 0.3	

<sup>a</sup> Calculated as weight (kg)/[height (cm) - 100] × 100%.

<sup>b</sup> To convert mmol/l to mg/dl, multiply cholesterol concentrations by 38.7 and triglyceride concentrations by 0.1 × mol wt of triglyceride (e.g., 88.5 for triolein).

<sup>c</sup> FCH, familial combined hyperlipidemia; FHC, familial hypercholesterolemia; FHT, familial hypertriglyceridemia; GBD, gallbladder disease (cholelithiasis, cholecystitis, cholecystectomy); HT, hypertension; IHD, ischemic heart disease.

continuously with digitalis, diuretics and/or nitrate preparations; this therapy was kept unchanged during the present investigations and in the interval between them. During the preceding months, none of the patients had been treated with drugs or diets known to interfere with lipoprotein metabolism.

### Experimental procedure

The patients were hospitalized during the studies and maintained on a standardized diet of natural type (cf. 3, 4). About 40% of the energy content was supplied as fat, most of which contained saturated fatty acids. The major part of the carbohydrates, which accounted for 39% of the calories, was supplied as starch. The energy intake, calculated from standard foodstuff tables<sup>2</sup>, was adjusted to keep the body weight constant. The intake of cholesterol was about 0.5 mmol/day in each subject.

In patients without gallbladder disease (GBD), fasting duodenal bile was sampled after 4–7 days. Cholecystokinin was given intravenously, and bile was obtained through a thin polyvinyl tube on 2 consecutive days. A few days later, the patients received an intravenous injection of [<sup>3</sup>H]glycerol (40 μCi) in the morning after an overnight fast. Venous

blood samples were obtained at intervals of 30 min from 2–6 hr and then at 7, 8, and 10 hr after administration of the isotope. Plasma radioactivity was determined in all samples, and the concentration of plasma triglycerides in the samples collected at 0, 2, 4, 6, and 8 hr.

Following the first study, the patients were treated with cholic acid, 15 mg (37.5 μmol) per kg body weight per day, for three months. The cholic acid was administered in specially prepared capsules twice daily, and was tolerated without obvious discomfort. Routine indices of hepatic function (serum bilirubin, alanine aminotransferase, aspartate aminotransferase, and alkaline phosphatase) were unchanged and within normal limits in all patients. Body weights remained constant within 2 kg (Table 1).

At the end of the treatment period, the patients were again hospitalized and fed the standardized diet, and the biliary lipid composition and plasma triglyceride kinetics were reexamined during medication as described above.

### Materials

[2-<sup>3</sup>H]Glycerol (sp act, 200 μCi/μmol) was obtained from the Radiochemical Centre, Amersham, England. Prior to use, the isotope was purified from possible contamination with [<sup>3</sup>H]water by evaporation in vacuo to dryness. The residue, which was shown to be radio-

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chemically pure by thin-layer chromatography, was dissolved in 70% ethanol and diluted with saline before intravenous injection.

Cholic acid was obtained from Sigma Chemical Company, St. Louis, MO, and was shown to be >98% pure by thin-layer chromatography. A purified 3 $\alpha$ -hydroxysteroid dehydrogenase preparation, supplied as a kit (Sterognost-3 $\alpha$ ) was purchased from Nyegaard & Co. A/S, Oslo, Norway. Cholecystokinin was obtained from the Gastrointestinal Hormone Research Group, Department of Chemistry, Karolinska Institutet, Stockholm, Sweden.

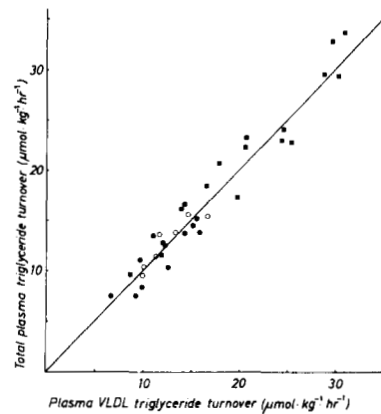
### Determination of plasma lipid levels and triglyceride kinetics

Cholesterol and triglycerides were determined with a Technicon Autoanalyzer (Technicon Instruments Corp., Tarrytown, NY). Lipoprotein phenotyping according to WHO recommendations (1) was performed as described earlier (3, 4).

Plasma triglyceride kinetics were determined by the method of Farquhar et al. (12) as described by Nikkilä and Kekki (6, 13, 14). Details of the experimental procedure have been given in a previous paper (4). In no case did the plasma triglyceride concentration show variations of more than  $\pm 5\%$  of the initial value. One patient was excluded from the study, as her triglyceride elimination curve was found to be composed of more than one exponential slope.

The apparent fractional turnover rate of endogenous plasma triglyceride ( $k$ , hr<sup>-1</sup>) was determined from the exponential descending slope of the plasma radioactivity curve. Before treatment, the standard error of the individual fractional turnover rate determinations averaged 0.008 (6%) in patients with type IIa and 0.008 (6%) in patients with type IV HLP. During medication, the corresponding figures were 0.009 (6%) and 0.007 (5%) in type IIa and type IV HLP, respectively. The correlation coefficients ranged between 0.940 and 0.992 before and between 0.954 and 0.997 during treatment with cholic acid ( $P < 0.001$  in all cases). Thus, in all cases, except the one excluded as mentioned above, single-exponential decay curves were obtained.

The apparent plasma triglyceride production rate, expressed in  $\mu\text{mol}\cdot\text{kg}^{-1}\cdot\text{hr}^{-1}$ , was calculated from the apparent fractional turnover rate and the mean plasma triglyceride concentration as described previously (4). The standard error in the calculation of triglyceride synthesis in the individual subject before treatment averaged 8% in patients with type IIa HLP and 7% in those with type IV HLP. In this calculation, both error in fractional turnover rate and triglyceride



**Fig. 1.** Apparent plasma total triglyceride synthesis as a function of apparent VLDL triglyceride synthesis (measured over 10 hr). Equation of the regression line  $y = 0.073 + 1.02x$ ,  $R = +0.973$ ,  $P < 0.001$ .  $\circ$ , controls;  $\bullet$ , type IIa HLP;  $\blacksquare$ , type IV HLP.

variability are considered. Corresponding figures during treatment were 8% in both type IIa and type IV HLP.

At the time of the present study, we did not have the technical possibilities for isolating the very low-density lipoproteins (VLDL), and the data presented thus refer to total plasma triglyceride turnover. In order to relate the kinetics of total plasma triglycerides to those of VLDL triglycerides, parallel determinations of the two parameters were performed over 10 hr in altogether 35 subjects (7 normolipidemic controls, 10 with type IIa, and 18 with type IV HLP). VLDL was isolated by preparative ultracentrifugation (15), and calculations made as for total plasma triglycerides. As expected, the apparent fractional turnover rate for total plasma triglyceride was lower than for VLDL triglyceride ( $0.174 \pm 0.010$  vs.  $0.268 \pm 0.019$ , mean  $\pm$  SEM,  $P < 0.001$ ), but the two variables were well correlated ( $R = +0.72$ ,  $P < 0.001$ ). In agreement with previous studies (16), there was a close similarity between the apparent production rate of total plasma triglycerides and that of VLDL triglycerides ( $16.9 \pm 1.1$  and  $16.3 \pm 1.1 \mu\text{mol}\cdot\text{kg}^{-1}\cdot\text{hr}^{-1}$ ). As seen in **Fig. 1**, there was a close correlation between the two determinations ( $R = +0.92$  for controls,  $R = +0.91$  for type IIa,  $R = +0.97$  for type IV, and  $R = +0.97$  for all subjects,  $P < 0.001$ ). The mean deviation of plasma total triglyceride synthesis from VLDL triglyceride synthesis was  $4 \pm 2\%$ , and in no case was the error greater than 20%.

### Determination of biliary lipid composition

The total bile acid concentration in one aliquot of the bile samples was determined enzymatically with a 3 $\alpha$ -hydroxysteroid dehydrogenase method (17). Another aliquot of the bile was extracted without delay



TABLE 2. Effect of cholic acid on plasma lipid levels and triglyceride kinetics in patients with hyperlipoproteinemia (HLP).

Patient, Type of HLP	Plasma Cholesterol Concentration <sup>a</sup>		Plasma Triglyceride					
	Before	During	Concentration <sup>a</sup>		Apparent Fractional Turnover Rate		Apparent Synthesis	
			Before	During	Before	During	Before	During
	mmol/l		mmol/l		hr <sup>-1</sup>		μmol·kg <sup>-1</sup> ·hr <sup>-1</sup>	
1. IIa	10.2	9.2	1.8	1.7	0.179	0.163	14.4	12.5
2. IIa	8.2	8.2	1.6	1.2	0.104	0.155	7.5	8.4
3. IIa	9.5	9.4	1.6	1.8	0.232	0.217	15.5	16.3
4. IIa	8.9	7.8	1.2	1.4	0.139	0.163	6.8	9.2
5. IIa	10.0	9.0	1.8	1.5	0.134	0.168	10.6	11.1
Mean ± SEM	9.4 ± 0.4	8.7 ± 0.3	1.6 ± 0.1	1.5 ± 0.1	0.158 ± 0.022	0.173 ± 0.011	11.0 ± 1.8	11.5 ± 1.4
6. IV	6.3	6.7	2.2	2.2	0.057	0.097	5.6	9.4
7. IV	7.6	7.3	3.7	3.2	0.174	0.120	27.8	16.6
8. IV	5.8	6.7	2.9	2.3	0.253	0.236	29.1	21.3
9. IV	5.8	6.6	4.3	3.8	0.114	0.097	22.1	16.6
10. IV	6.5	7.6	2.8	2.6	0.261	0.157	29.6	16.5
11. IV	5.0	5.5	3.4	2.8	0.099	0.164	14.2	19.4
12. IV	6.7	6.1	4.3	2.5	0.161	0.125	25.6	11.7
13. IV	5.3	5.2	2.1	2.0	0.201	0.248	19.0	22.3
Mean ± SEM	6.2 ± 0.3	6.6 ± 0.3	3.2 ± 0.3	2.7 ± 0.2 <sup>b</sup>	0.160 ± 0.029	0.142 ± 0.018	21.6 ± 3.1	16.7 ± 1.6
Total			2.6 ± 0.3	2.3 ± 0.2	0.159 ± 0.019	0.155 ± 0.012	17.5 ± 2.4	14.7 ± 1.3

<sup>a</sup> To convert mmol/l to mg/dl, multiply cholesterol concentrations by 38.7 and triglyceride concentrations by 0.1 × mol wt of triglyceride (e.g., 88.5 for triolein).

<sup>b</sup> Significantly different from pretreatment value,  $P < 0.05$ .

with 20 volumes of chloroform-methanol 2:1 (v/v). Cholesterol (18) and phospholipids (19) in the chloroform extract were determined. The remaining part of the bile sample was hydrolyzed with 1 M KOH in closed steel tubes for 12 hr at 110°C. The bile acids were extracted with ethyl ether, methylated with diazomethane, converted into trimethylsilyl ethers, and finally analyzed by gas-liquid chromatography using a 1% Hi-Eff 8BP column.

Lipid composition of bile was expressed as molar per cent cholesterol, bile acids, and phospholipids. The cholesterol saturation in bile was determined according to Carey and Small (20), using a biliary lipid concentration of 10 g/dl.<sup>3</sup>

#### Statistical analysis

Data are presented as mean ± SEM. The significance of differences was evaluated by Student's paired *t* test (21).

<sup>3</sup> This concentration was chosen as we have found the fasting gallbladder bile to contain about 10 g/dl of lipids in patients with, as well as without, GBD (Ahlberg, J., B. Angelin, and K. Einarsson. Unpublished results).

## RESULTS

### Plasma lipid levels and triglyceride kinetics

Individual values for plasma lipid levels as well as the apparent fractional turnover rate and synthesis of endogenous plasma triglyceride are shown in **Table 2**.

In type IIa HLP, the plasma cholesterol levels decreased slightly during treatment with cholic acid in three of the five patients studied, whereas the plasma triglycerides were essentially unchanged. There were no consistent changes in the apparent fractional turnover rate or synthesis of plasma triglycerides as determined over 10 hr.

In type IV HLP, the plasma triglyceride concentration decreased during treatment in six of the eight patients (from 3.2 ± 0.3 to 2.7 ± 0.2 mmol/l,  $P < 0.05$ ). No changes in plasma cholesterol levels were seen. Before treatment, the apparent formation of endogenous plasma triglycerides was about twice as high as that seen in type IIa HLP. Medication with cholic acid was not associated with any statistically significant changes in the apparent fractional turnover rate or production. The apparent synthesis of plasma

triglycerides was, however, clearly decreased in five of the patients, while the remaining three were essentially unchanged. The decrease in apparent synthetic rate was confined to the patients with an initial high triglyceride formation (more than  $20 \mu\text{mol}\cdot\text{kg}^{-1}\cdot\text{hr}^{-1}$ ). As seen in Fig. 2, the change in apparent plasma triglyceride synthesis was significantly related to the initial production values ( $R = -0.85, P < 0.001$ ).

### Biliary lipid composition

Cholic acid, chenodeoxycholic acid, and deoxycholic acid were the dominating bile acids in duodenal bile, and only trace amounts of lithocholic acid or ursodeoxycholic acid were occasionally observed. Treatment with cholic acid was associated with a considerable increase not only in the proportion of cholic acid but also in that of deoxycholic acid (Table 3). These changes were somewhat less pronounced in type IV than in type IIa HLP.

The concentration of bile acids, expressed as a molar percentage of total biliary lipids (bile acids, cholesterol, and phospholipids), increased during treatment with cholic acid from  $70.3 \pm 2.0$  to  $76.0 \pm 1.2$  molar % ( $P < 0.02$ ) (Table 3). Concomitantly, the proportion of cholesterol was reduced (from  $6.6 \pm 0.7$  to  $5.3 \pm 0.4$  molar %,  $P < 0.02$ ), as was the phospholipid fraction (from  $23.1 \pm 1.7$  to  $18.6 \pm 0.9$  molar %,  $P < 0.05$ ). Thus, the ratios between cholesterol and bile acids, and between phospholipids and bile acids, were reduced during treatment with cholic acid, from  $0.097 \pm 0.013$  to  $0.071 \pm 0.007$  ( $P < 0.02$ ), and from  $0.338 \pm 0.037$  to  $0.247 \pm 0.015$  ( $P < 0.05$ ), respectively, while the ratio between cholesterol and phospholipids was unchanged ( $0.295 \pm 0.033$  before and  $0.286 \pm 0.019$  during therapy).

The saturation of bile with cholesterol, using the

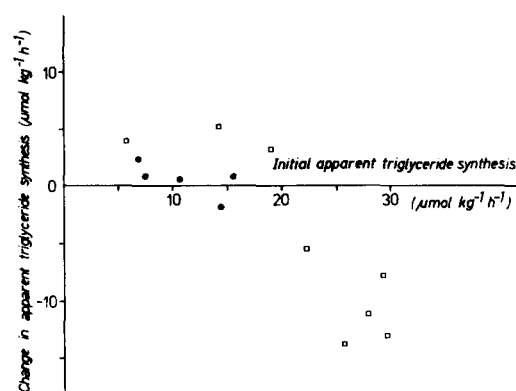


Fig. 2. Change in apparent plasma triglyceride synthesis during cholic acid treatment as a function of initial synthesis rate. ●, indicates patients with type IIa HLP; □, patients with type IV HLP.  $R = -0.846, P < 0.001$ .

solubility limits of Carey and Small (20), was not significantly reduced during cholic acid treatment. As seen in Fig. 3, four out of the five patients with initially supersaturated bile decreased their biliary cholesterol saturation, while those with low initial values were essentially unchanged. The change in cholesterol saturation was thus closely related to the initial value ( $R = -0.89, P < 0.001$ ).

### DISCUSSION

The biosynthesis of bile acids is regulated by a negative feedback control, triggered by the amount of bile acids reaching the liver via the portal vein (for a review, see ref. 22). Expansion of the chenodeoxycholic acid pool by feeding with this bile acid is associated with a decreased formation and pool size of both cholic acid and its metabolite, deoxycholic acid (23–26). Treatment with cholic acid is associated with ex-

TABLE 3. Biliary lipid composition before and during treatment with cholic acid in hyperlipoproteinemia (HLP). Means  $\pm$  SEM.

Patients (Number of Subjects)		Ratio between Duodenal Bile Acids <sup>a</sup>			Cholesterol molar %	Bile Acids molar %	Phospholipids molar %	Cholesterol Saturation <sup>b</sup>
		C	CD	D				
HLP type IIa (n = 5)	Before	1.2 $\pm$ 0.2	1	0.3 $\pm$ 0.2	6.1 $\pm$ 0.9	73.9 $\pm$ 2.2	20.0 $\pm$ 1.4	90 $\pm$ 10
	During	6.8 $\pm$ 1.2 <sup>e</sup>	1	4.5 $\pm$ 1.4 <sup>e</sup>	4.7 $\pm$ 0.3	78.7 $\pm$ 1.2 <sup>c</sup>	16.6 $\pm$ 1.0	81 $\pm$ 3
HLP type IV (n = 6)	Before	1.1 $\pm$ 0.2	1	0.8 $\pm$ 0.3	7.1 $\pm$ 1.1	67.3 $\pm$ 2.8	25.6 $\pm$ 2.5	96 $\pm$ 16
	During	4.2 $\pm$ 0.6 <sup>e</sup>	1	4.2 $\pm$ 1.4 <sup>c</sup>	5.9 $\pm$ 0.7	73.8 $\pm$ 1.4	20.3 $\pm$ 0.9	87 $\pm$ 10
Total (n = 11)	Before	1.1 $\pm$ 0.1	1	0.7 $\pm$ 0.2	6.6 $\pm$ 0.7	70.3 $\pm$ 2.0	23.1 $\pm$ 1.7	93 $\pm$ 9
	During	5.4 $\pm$ 0.7 <sup>f</sup>	1	4.6 $\pm$ 0.9 <sup>f</sup>	5.3 $\pm$ 0.4 <sup>d</sup>	76.0 $\pm$ 1.2 <sup>d</sup>	18.6 $\pm$ 0.9 <sup>e</sup>	84 $\pm$ 5

<sup>a</sup> C, Cholic acid; CD, chenodeoxycholic acid; D, deoxycholic acid.

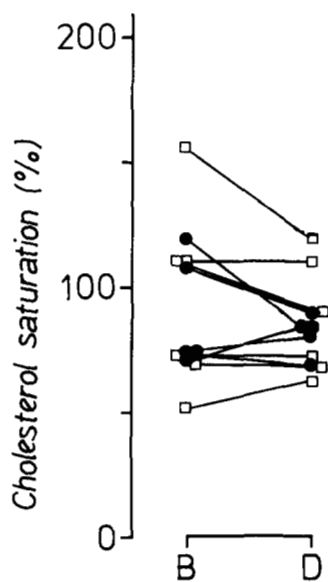
<sup>b</sup> Calculated according to Carey and Small (14) using a biliary lipid concentration of 10 g/dl.

<sup>c</sup> Significantly different from before treatment,  $P < 0.05$ .

<sup>d</sup> Significantly different from before treatment,  $P < 0.02$ .

<sup>e</sup> Significantly different from before treatment,  $P < 0.01$ .

<sup>f</sup> Significantly different from before treatment,  $P < 0.001$ .



**Fig. 3.** Cholesterol saturation (%) of duodenal bile as calculated from cholesterol solubility limits of Carey and Small (17) before (B) and during (D) treatment with cholic acid. Symbols as in Fig. 1.

pansion of the pool size of both cholic and deoxycholic acids, whereas the production and pool size of chenodeoxycholic acid are reduced (23–25, 27, 28). In agreement with previous reports, the present study showed the proportions of both cholic and deoxycholic acids to be increased in duodenal bile. Thus, the effects of cholic acid treatment on the variables measured in the present work actually reflect the net effects of both cholic and deoxycholic acids.

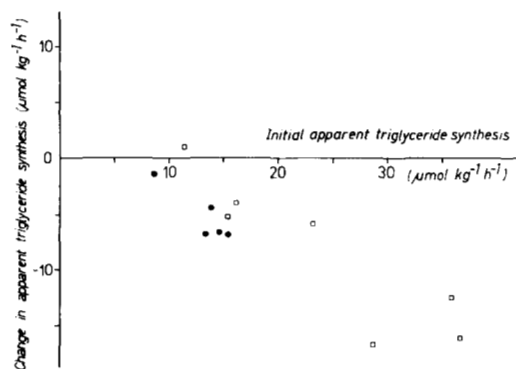
Treatment with chenodeoxycholic acid, but not with cholic acid, is reported to be associated with a reduction of the biliary cholesterol saturation (23–25, 29). In the present series of hyperlipidemic patients without GBD, feeding of cholic acid resulted in an increase in biliary bile acids and a concomitant decrease in cholesterol and phospholipids, whereas no significant change in cholesterol saturation was seen. Although some patients with HLP, viz. those with the highest pretreatment saturation, may show a decrease in biliary cholesterol saturation (Fig. 2), the slight changes in cholesterol solubility seen in the present investigation are marginal compared to the effects seen during chenodeoxycholic acid treatment in HLP (30). Thus, treatment with the two primary bile acids appears to have different effects on biliary cholesterol saturation.

The plasma concentration of triglycerides was unaffected by cholic acid therapy in type IIa HLP, whereas a significant decrease was seen in type IV. This is in accordance with a previous report (28), where triglyceride levels were reduced by about 20% in type IV HLP. In similarity with that study, the

present investigation showed a tendency to a slight reduction of plasma cholesterol levels in type IIa HLP during medication with cholic acid.

In the present work, endogenous plasma triglyceride production was estimated using the [ $^3\text{H}$ ]-glycerol labeling technique originally described by Farquhar et al. (12). As the patients were studied in the postabsorptive state, the “apparent” plasma triglyceride synthesis determined should closely reflect the formation of VLDL triglycerides, which has also been demonstrated by parallel measurements of these two parameters (cf. Methods, 16, 31). It should be pointed out, that the present studies were carried out over a 10 hr time period, and thus the emergence of a slow component of the curve after 15 hr, as has been suggested in some preliminary reports (32, 33), cannot be excluded. The possible limitations of the present method have been discussed in detail previously (4). The glycerol labeling technique, as used in the present study, has however been found to give data in good agreement with those obtained with independent techniques using lipolytic rate procedures or VLDL-apoB kinetics (31, 34).

The pretreatment values for apparent plasma endogenous triglyceride synthesis seen in the present study are in close agreement with those previously reported for a larger series of hyperlipidemic patients using a similar technique (4). Thus, the mean synthesis was about twice as high in type IV as in type IIa HLP. In contrast to the clear reduction in 10 hr triglyceride production rate generally seen during chenodeoxycholic acid medication in HLP (10; Fig. 4), treatment with cholic acid was not associated with consistent changes in plasma triglyceride metabolism. However, in the subpopulation of type IV patients with an initially elevated formation of plasma triglycerides, this was clearly reduced during cholic acid therapy (Fig. 2). Considering the apparent link between



**Fig. 4.** Change in apparent plasma triglyceride synthesis during chenodeoxycholic acid treatment as a function of initial synthetic rate (data from ref. 10). Symbols as in Fig. 1.  $R = -0.871$ ,  $P < 0.001$ .



triglyceride metabolism and bile acid biosynthesis (4), it is reasonable to assume that these patients have an increased bile acid production before treatment. The possibility of a defective intestinal uptake of cholic acid in this patient subgroup has been raised (3, 10, 28, 35), and treatment with cholic acid might thus restore the feed-back inhibition to normal. In this context it is interesting to note, that Adler et al. (36) recently reported that rhesus monkeys with a partial biliary diversion displayed an increased plasma triglyceride formation. Also this increase could be returned to normal by duodenal perfusion with cholic acid (36).

Thus, the data of the present study together with previous work (10) suggest that treatment with chenodeoxycholic acid, but not with cholic acid, generally reduces the production of endogenous plasma triglycerides in HLP. The reason for this difference between the two primary bile acids cannot be decided from the present study. The difference may be related to the different effects of the two bile acids on hepatic HMG CoA reductase, the rate-limiting step in cholesterol biosynthesis. Thus, treatment with chenodeoxycholic acid is known to suppress hepatic HMG CoA reductase in man (37, 38)<sup>4</sup>, whereas medication with cholic acid does not.<sup>4</sup> At least in the rat, HMG CoA reductase activity is related to the production of VLDL triglycerides (39), and the difference between individual bile acids in enzymatic suppression may thus be a possible explanation for the different effects on apparent plasma triglyceride production rate. Patients with type IV HLP often display an increased hepatic HMG CoA reductase activity (40); such patients may hypothetically respond more readily to bile acid feeding.

The different biological effects seen during treatment with cholic and chenodeoxycholic acids may be related to their structural differences; however, it may well be that differences in "bioavailability" are of major importance. As mentioned, cholic acid is efficiently converted to deoxycholic acid, which may itself have influences on the parameters studied, and thus never accumulates in the enterohepatic circulation to the same extent as chenodeoxycholic acid. Furthermore, we have previously presented evidence for a more efficient absorption and a more rapid enterohepatic circulation of chenodeoxycholic than of cholic acid in fasting man (41–43). In agreement with this contention, it was recently shown that the fasting portal venous concentration of bile acids was higher during chenodeoxycholic than during cholic acid treatment (44). This higher concentration may be

of importance for a more efficient suppression of hepatic HMG CoA reductase activity.

Finally, the parallel changes in plasma triglyceride turnover, bile acid metabolism, and biliary cholesterol saturation during treatment with chenodeoxycholic and cholic acids in HLP focus interest on the possible role of deranged lipoprotein metabolism as one factor in the development of supersaturated bile. Certain forms of HLP appear to carry an increased risk for development of gallstone disease (45, 46), due to an elevated biliary cholesterol saturation.<sup>5</sup> Further studies aimed at defining these apparent relationships seem to be of definite future interest. ■■

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