THE CHOLESTEROL-SOLUBILIZING POWER OF BILIARY LIPIDS

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Biliary cholesterol has long excited interest, since it was shown that cholesterol was an important constituent of many gallstones and thus might be of aetiological importance in their formation.[1] Since cholesterol is present in bile in a concentration beyond that at which it is normally soluble in triglycerides at body temperature,[2] the continued state of solution of it in the oily phase of bile must depend on the presence of other factors. This problem has been approached in a number of ways. There are indications that both bile acids[1] and biliary phospholipids[3] increase the solubility of cholesterol in bile.

The ability of phospholipids to 'solubilize' other lipids in tissues has been outlined elsewhere.[4] The factors that keep serum cholesterol in the oily phase have also been investigated. The addition of very small amounts of total serum lipids to a supersaturated cholesterol solution in a triglyceride oil not only increases the stability of the solution but also depresses the rate of crystallization of the sterol.[5, 6] This property of the serum lipids could be attributed to the action of the combined serum phospholipids; consequently, it was suggested that the serum phospholipids might help to maintain the solubility of serum cholesterol in the circulating plasma lipids.[6]

The effect of the total biliary lipids on a supersaturated solution of cholesterol in a triglyceride oil has been investigated for the following reasons, namely:

(a) It appeared to be a simpler method than certain others[1, 3] for studying the stabilizing factors present in bile in health and disease.

(b) A study of the effect of the total serum lipids on the same test system has proved useful. Ischaemic heart disease,[5, 6] the administration of adrenaline,[7] and surgical stress,[8] have all been shown to reduce the stabilizing effect of the total serum lipids.

This paper deals with the results of a preliminary investigation into the effects of the total biliary lipids on the stability of a supersaturated solution of cholesterol in a triglyceride oil.

Material

Bile was obtained from the following sources, namely,
(a) from patients with postoperative T-tubes,
(b) from rabbits and cats with tubes in the common bile duct while anaesthetized with 'pentobarbitone', and
(c) from direct puncture of the gallbladder in dogs.

The bile was stored at a temperature lower than 0°C. until used.

Methods

The method of Wilkens and Krut[6] (1963), designed to study the effect of the total serum lipids on the stability of cholesterol-triglyceride solution, was used with the following changes, namely,
(a) 1 ml. of bile was used instead of 1 ml. of fasting serum, and
(b) exactly 1·25 G. of cholesterol, instead of about 1·75 G., was dissolved in the triglyceride oil.

Crystallization rate value. The maximum crystallization rate (CR value) was computed as described by Wilkens and Krut.[6] In each case the CR value produced by a sample of total biliary lipids was compared with the CR value of the standard solution to which no biliary lipid had been added.

Results

The addition of small amounts of total biliary lipids to a supersaturated solution of cholesterol in a triglyceride oil not only increased the stability of the solution but also

markedly depressed the rate of crystallization of the sterol when crystallization did occur (Table I). The CR values of different samples of bile also varied.

TABLE I. THE STABILIZING EFFECT OF BILIARY LIPIDS ON CHOLESTEROL

	Subjects		*Controls*
Animal	*Volume of bile used (ml.)*	*CR value*	*CR value*
Cat 1	1·0	2·60	4·01
Cat 2	1·0	1·96	4·01
Cat 3	1·0	2·05	3·73
Cat 4	1·0	1·43	4·01
Rabbit 1	1·0	2·16	4·01
Rabbit 2	0·8	2·45	4·01
Rabbit 3	0·5	3·49	4·01
Rabbit 4	1·0	2·75	3·49
Man 1	1·0	3·49	4·01
Man 2	1·0	1·88	3·73
Man 3	1·0	3·08	4·01
Man 4	1·0	2·75	3·73
Man 5	1·0	3·49	3·73
Man 6	1·0	1·73	4·01
Man 7	1·0	2·36	3·73
Man 8	1·0	1·00	4·01
Dog 1	1·0	1·96	4·01

DISCUSSION

The nature of the stabilizing factors present in the total biliary lipids used remains to be determined. It would seem likely, however, that the bile salts and phospholipids might account for this property.

Molecules of bile salts in solution will at certain critical concentrations aggregate to form larger molecular masses called micelles. Once these micelles are formed, the ability of the solution to dissolve cholesterol is increased.[9] Preliminary results likewise suggest that the addition of sodium taurocholate to the cholesterol-triglyceride solution increases the stability of the contained cholesterol.

The addition of lecithin to bile increases its cholesterol-holding capacity.[3] The stabilizing effect of the total serum lipids on the cholesterol-triglyceride solution used was likewise found to be attributable to the effect of the phospholipids.[5, 6]

Lecithin has been shown to increase the stability of cholesterol in the cholesterol-triglyceride solution, while inositol and lysolecithin cause a decrease in stability; however, none of these phospholipid fractions were pure.[6] Presumably the decreased stabilizing power of the total serum lipids found in patients with ischaemic heart disease,[5, 6] in animals after the injection of adrenaline,[7] and in animals during surgical operations,[8] is related to a relative and/or absolute change in these phospholipid fractions.

Until quite recently it was believed that lecithin was the only phospholipid present in bile;[10] however, since 1960, it has been shown that several other phospholipids, e.g. cephalin, sphingomyelin and lysolecithin, are also present.[2, 11, 12] This would suggest that changes in the relative and/or absolute amounts of biliary phospholipids could produce changes in cholesterol stability. Bearing this possibility in mind, a study of the effect of biliary lipids on cholesterol stability under various conditions (e.g. cholelithiasis) has been started.

Although such studies into the physicochemical properties of bile salts, cholesterol and phospholipids are no doubt important in the elucidation of the problem of gallstone formation, it has been rightly pointed out that other factors, such as biliary stasis, infection and foreign bodies, may also be of aetiological importance.[1, 9]

SUMMARY

Biliary lipids increase the stability of cholesterol in a cholesterol-triglyceride solution and decrease the rate of crystallization of cholesterol when crystallization occurs. The possible role of these findings in relation to biliary pathology is discussed.

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