accompanied by the diminished solubility and mobility of the bone salt. The fluoroapatite crystals, therefore, are more stable and less reactive in surface exchange reactions, since larger crystals offer less surface area for a given weight of bone. It may therefore be assumed that these changes increase the resistance of bone to the actions of the parathyroid hormone and may cause lowering of the plasma ionized calcium, so stimulating the compensatory parathyroid activity.

However, it seems unlikely that the reduced resorption of fluoride bone alone is the factor exciting parathyroid stimulation, particularly in the light of certain animal experiments. Yates *et al.* (1964) showed evidence of parathyroid stimulation on a short-term basis using intraperitoneal lavage in rats. Faccini (1969) performed immunoassay of parathyroid hormone in sheep and demonstrated a significant increase in circulating hormone levels only a week after starting fluoride administration. The possibility, therefore, that fluoride might induce secondary hyperparathyroidism by interfering with the calcium equilibrium between bone and serum by accelerating crystal growth or producing a more rapid ion exchange has to be considered. We also feel that further investigations to exclude the direct effect of fluoride on the parathyroids are necessary.

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Addendum.—Since preparing this manuscript we have determined serum immunoreactive parathyroid hormone levels in our patients and found the expected high levels of 1,250, 2,800, 1,075 1,050, and 1,225 pg/ml respectively (in the assay used parathyroid hormone was unmeasurable in normal subjects). Immunoreactive parathyroid hormone levels correlated positively with the biochemical, radiological, and histological findings in all five cases.

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Fibrinolysis in Cholestatic Jaundice

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Summary

The fibrinolytic system was studied in primary biliary cirrhosis (16 patients) and large bile duct obstruction (10 patients, nine of whom had carcinoma). Plasma fibrinolysis (plasminogen activator activity) was decreased and fibrinogen increased in both groups of patients, particularly in those with large duct obstruction. These changes were related to the degree of cholestasis. Plasminogen activator activity was inversely related to serum triglyceride levels in patients with primary biliary cirrhosis. Urokinase inhibitors were decreased in both groups and antiplasmins increased in patients with large duct obstruction; fibrin/fibrinogen degradation products were normal in primary biliary cirrhosis and moderately increased in large duct obstruction. None of these fibrinolytic indices was related to the degree of cholestasis. Fibrinolytic activity and fibrinogen returned almost to normal levels after palliative surgery in the three patients with large duct obstruction who were studied. The decreased plasma fibrinolysis and increased fibrinogen may be due to altered lipid metabolism in cholestatic jaundice. In patients undergoing surgery for large duct obstruction there may be an increased risk of thrombosis.

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Introduction

Little is known about fibrinolysis in cholestatic jaundice. Ratnoff (1949), Nilehn and Nilsson (1964), and van de Loo and Schmiesing (1965) found normal fibrinolytic activity in patients with this condition. Few patients were studied, however, and the methods used were usually designed to detect normal or increased rather than decreased fibrinolysis. We therefore investigated fibrinolysis in two groups of patients: one group with primary biliary cirrhosis and the other with large bile duct obstruction.

Patients and Controls

Sixteen patients (aged 35-66 years) with primary biliary cirrhosis were studied. The diagnosis was based on clinical, immunological, and histological criteria (Scheuer, 1968; Sherlock, 1968). There were 10 patients (aged 28-62 years) with obstruction to large bile ducts; nine had carcinoma of the pancreas or bile ducts and one a stricture of the common bile duct. The control group (12 men and nine women, aged 21-62 years) were healthy medical and laboratory personnel.

Methods

Blood samples were taken from resting, fasting, non-smoking patients and controls at about 9 a.m. Blood was obtained from an antecubital vein, almost always without venous occlusion, and was transferred to plastic tubes on ice. Plasminogen activator activity was determined by measuring the euglobulin lysis time (Januszko and Dubinska, 1965) and euglobulin lysis on bovine fibrin plates (Astrup and Müllertz, 1952). Plasminogen was measured by a caseinolytic method (Alkjaersig *et al.*, 1959), and fibrinogen by a thrombin clotting method (Quick, 1959). Serum urokinase inhibitor (Januszko *et al.*, 1966) and antiplasmin (Aoki and von Kaulla, 1969) were measured. Serum fibrin/fibrinogen degradation products were assayed on microtitre plates, using Burroughs Wellcome reagents, by a modification of the tanned red cell haemagglutination inhibition technique described by Das *et al.* (1967). Serum triglycerides (Marzo *et al.*, 1971) were measured in patients with primary biliary cirrhosis, and liver function tests were performed.

Statistical Analysis.—Wilcoxon's (1964) rank sum test was used to determine the statistical significance of the results.

Results

The results of fibrinolytic studies in patients and controls are shown in table I; liver function test results (and serum triglycerides in primary biliary cirrhosis) are shown in table II. Plasma plasminogen activator activity (measured by euglobulin clot lysis time and fibrin plate lysis) was decreased in both groups of patients, particularly in those with large bile duct obstruction (table I). Results of the euglobulin lysis test were inversely related to bilirubin levels (fig. 1), but the correlation between fibrin plate lysis and bilirubin was not statistically significant ($0{\cdot}05 < P < 0{\cdot}1$). There was no significant correlation between plasminogen activator activity and serum cholesterol. However, in patients with primary biliary cirrhosis plasminogen activator activity was inversely related to serum triglyceride levels (fig. 2).

Fibrinogen was increased in both groups, particularly in patients with large bile duct obstruction (table I). This increase was related to the levels of bilirubin ($P < 0{\cdot}02$), cholesterol ($P < 0{\cdot}05$), and alkaline phosphatase ($P < 0{\cdot}05$). Fibrinogen levels were inversely related to plasminogen activator activity in the euglobulin test ($P < 0{\cdot}01$), but not in the fibrin plate lysis test.

As shown in table I, plasma plasminogen was decreased in patients with primary biliary cirrhosis and increased in those with large bile duct obstruction, but the results were not statistically significant. Serum urokinase inhibitor levels were decreased in both groups of patients. Serum antiplasmins were increased in both groups but the results were statistically significant only in patients with large bile duct obstruction. There was no correlation between the inhibitors of fibrinolysis and liver function tests or triglycerides. Serum fibrin/fibrinogen degradation products were within the normal range in patients with primary biliary cirrhosis and increased in those with large bile duct obstruction.

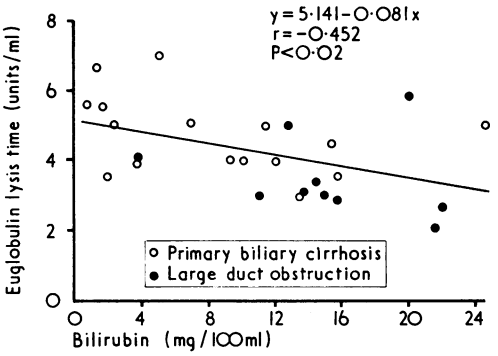

FIG. 1—Relation of euglobulin lysis time to serum bilirubin.

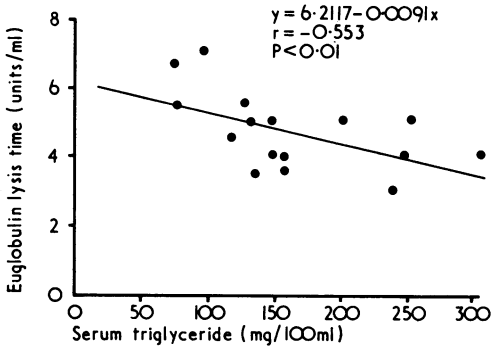

FIG. 2—Relation of euglobulin lysis time to serum triglyceride levels.

Fibrinolysis after Relief of Biliary Obstruction.—In three patients fibrinolysis was studied before and 15-20 days after palliative surgery to relieve large bile duct obstruction. None of these patients had wound infections at the time of the study. The results of fibrinolytic studies are shown in table III.

TABLE I—*Results of Fibrinolytic Studies*

		Euglobulin Lysis Time (units/ml)	Fibrin Plate Lysis (mm^2)	Fibrinogen (mg/100 ml)	Plasminogen (units/ml)	Urokinase Inhibitor (units/ml)	Antiplasmin (%)	Fibrin/ Fibrinogen Degradation Products (μg/ml)
Control (n = 21). Mean ± S.D.		9·5 ± 2	157 ± 38·6	280 ± 38	3·3 ± 0·8	1,779 ± 376	99 ± 47·4	4·4 ± 2·9
Primary biliary cirrhosis (n = 16)	Mean ± S.D.	4·7 ± 1·07	65 ± 34·2	371 ± 100	2·8 ± 1·05	1,162 ± 484	143 ± 73·6	6 ± 3
	P	<0·01	<0·01	<0·01	N.S.	<0·01	N.S.	N.S.
Large bile duct obstruction (n = 10)	Mean ± S.D.	3·5 ± 1·14	46 ± 18·8	564 ± 150	4·2 ± 1·7	1,307 ± 448	278 ± 183	11 ± 6
	P	<0·01	<0·01	<0·01	N.S.	<0·02	<0·01	<0·01

N.S. = Not significant.

TABLE II—*Results of Liver Function Tests*

	Cholesterol (mg/100 ml)	Albumin (g/100 ml)	gamma-globulin (g/100 ml)	Thrombotest (%)	Bilirubin (mg/100 ml)	Alkaline Phosphatase (K.A. units/ 100 ml)	Aspartate Transaminase (IU/l)	Serum Triglyceride (mg/100 ml)
Normal Range	130–260	3·6–5·2	0·5–1·5	70–100	0·3–1·0	3–13	5–17	30–175
Primary biliary cirrhosis Mean ± S.D.	327 ± 196·8	3·1 ± 0·66	1·89 ± 0·55	80 ± 28·9	8·1 ± 7·03	62 ± 32·9	44 ± 18·1	166 ± 67
Extrahepatic biliary obstruction Mean ± S.D.	426 ± 174·1	3·0 ± 0·69	1·3 ± 0·50	61 ± 29·7	15·1 ± 5·40	106 ± 62·5	78 ± 72·4	—

TABLE III—*Fibrinolysis Before (B) and After (A) Relief of Biliary Obstruction*

Patient No.	Euglobulin Lysis Time (units/ml)		Fibrin Plate Lysis (mm^2)		Fibrinogen		Plasminogen	
	(B)	(A)	(B)	(A)	(B)	(A)	(B)	(A)
1	2·1	6·3	40	66	523	406	6	4·2
2	2·9	6·0	47	38	823	459	2·6	2·1
3	3·1	4·8	36	33	632	382	4	2·9

Plasminogen activator activity increased after operation; fibrinogen and plasminogen levels fell.

Discussion

Ratnoff (1949), Nilehn and Nilsson (1964) and van de Loo and Schmiesing (1965) reported normal fibrinolysis in cholestatic jaundice but we have found decreased fibrinolysis. There may be two reasons for the discrepancy between our findings and those of previous authors. Firstly, there were differences in methodology and in some studies the clots were not observed long enough to establish the time of lysis, so that a prolonged lysis time could have been overlooked. Secondly, the selection of patients in this study (primary biliary cirrhosis and neoplastic biliary obstruction) differs from that in other studies, in which most patients had obstruction due to gall stones.

There is an association between decreased fibrinolysis (low plasma plasminogen activator levels) and hyperfibrinogenaemia in some diseases (Gillman and Naidoo, 1958; Edward *et al.*, 1964) and, as hyperfibrinogenaemia is present in cholestatic jaundice (Gram, 1922), decreased fibrinolysis might also be expected to occur in this condition. The significance of the association between hyperfibrinogenaemia and impaired fibrinolysis is not certain, although hyperfibrinogenaemia may be a result of impaired fibrinolysis since inhibition of fibrinolysis with aminocaproic acid increases fibrinogen levels (Niewiarowski and Wolosowicz, 1966).

Plasma fibrinolytic activity (plasminogen activator) was decreased in both groups of patients, particularly in those with large bile duct obstruction. These patients had higher bilirubin levels than those with primary biliary cirrhosis (table II). The decrease in fibrinolysis was related to the degree of jaundice (fig. 1) and in three patients with large duct obstruction plasminogen activator returned to normal levels after surgical relief of the obstruction.

Cholestasis may impair fibrinolysis by increasing serum lipids, some of which are known to inhibit fibrinolysis. Beta-lipoprotein (J. Picard, personal communication, 1972) and triglycerides (Phillips, 1960) are raised in cholestatic jaundice and both these substances affect fibrinolysis. Beta-lipoprotein exerts antiplasmin activity (Riding and Ellis, 1964) and raised beta-lipoprotein levels are associated with decreased fibrinolysis (Nestel, 1960). Recently, impaired fibrinolysis has been associated with raised triglyceride levels (Sweet *et al.*, 1966; Wardle *et al.*, 1972), and our findings in patients with primary biliary cirrhosis are similar.

There was no correlation between the serum inhibitors of fibrinolysis and liver function tests or serum triglyceride levels. The two types of inhibitor behaved differently from one another. Urokinase levels, of which we found no reports in cholestatic jaundice, were decreased in both groups, particularly in those with primary biliary cirrhosis. The significance of these findings is not certain. Antiplasmin levels were raised in patients with large duct obstruction, possibly because of increased levels of betalipoprotein (see above) and α_2-macroglobulin, the main physiological antiplasmin (Ganrot, 1967). Raised levels of α_2-macroglobulin in cholestatic jaundice have recently been reported by Bogdat *et al.* (1972). They found the highest levels of α_2-macroglobulin and antiplasmin activity in three patients with neoplastic biliary obstruction; those with choledocholithiasis had normal or slightly increased levels. Moriau (1969) reported increased antiplasmin activity in cholecystitis and cholelithiasis but did not relate his findings to the degree of cholestasis. The significance of raised antiplasmin levels remains uncertain, but they may play an important part in thrombotic disorders (Naye, 1961).

Serum fibrin/fibrinogen degradation products were within the normal range in patients with primary biliary cirrhosis, which suggests that local fibrinolytic activity was not impaired despite decreased plasma fibrinolysis and increased antiplasmin levels. The moderately increased levels of fibrin/fibrinogen degradation products in patients with large bile duct obstruction may be due to increased local fibrinolytic activity associated with malignant disease. Nilehn and Nilsson (1964) did not find any in the serum of five patients with cholestatic jaundice. However, their method was less sensitive than haemagglutination inhibition assay (Merskey *et al.*, 1966).

Our findings indicate that cholestasis impairs fibrinolysis. This impairment may be due to altered lipid metabolism which occurs in cholestasis. As decreased plasma fibrinolysis is associated with thromboembolic disease (Naye, 1961; Nilsson *et al.*, 1961) patients undergoing surgery for biliary obstruction may be particularly liable to develop postoperative venous thrombosis. This problem merits further investigation.

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