

# Antithrombin III, plasminogen and alpha<sub>2</sub> antiplasmin in jaundice. Clinical usefulness and prognostic significance

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**SUMMARY** In this prospective study, antithrombin III, plasminogen and alpha<sub>2</sub> antiplasmin which are synthesised by the liver were measured and compared with the Normotest, Thrombotest and fibrinogen concentrations in 92 consecutive jaundiced patients. Antithrombin III appeared to be the most discriminant coagulation test in differentiating hepatocellular from cholestatic jaundice. A high correlation was observed between antithrombin III, plasminogen and alpha<sub>2</sub> antiplasmin values suggesting that the liver synthesis of these parameters was closely linked. The prognostic significance of the blood coagulation tests in patients with jaundice has been studied. In parenchymatous liver disease, antithrombin III, plasminogen and alpha<sub>2</sub> antiplasmin were superior to the Normotest, Thrombotest and fibrinogen concentrations in predicting the prognosis of the patients at the time of admission. In cholestatic jaundice, however, none of the blood coagulation tests studied had a prognostic significance.

Normotest and Thrombotest, which evaluate the vitamin K dependent coagulation factors synthesised by the liver, are prolonged in both hepatocellular and cholestatic jaundice.<sup>1-3</sup> The vitamin K independent fibrinogen concentration, however, is reduced only in severe hepatic failure.<sup>4,5</sup> Some other vitamin K independent components of haemostasis are synthesised by the liver including plasminogen, antithrombin III, the main inhibitor of thrombin, and alpha<sub>2</sub> antiplasmin, the main inhibitor of plasmin.<sup>6-9</sup> Although these parameters have been investigated in liver disease,<sup>10-16</sup> no attention has been given to jaundice.

The first aim of this prospective study was therefore to evaluate plasminogen, antithrombin III and alpha<sub>2</sub> antiplasmin in icteric patients in order to determine their clinical usefulness and their prognostic significance in jaundice. The second aim of the present investigation was to compare plasminogen, antithrombin III and alpha<sub>2</sub> antiplasmin with the classical blood coagulation tests used in jaundice – that is, Normotest, Thrombotest and fibrinogen concentration.

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Received for publication 16 December 1983

## Methods

### PATIENTS

In 92 jaundiced patients, 49 men and 43 women, ages ranging from 19–92 years (mean 61 years), hospitalised with a serum bilirubin above 2 mg/dl, antithrombin III, plasminogen, alpha<sub>2</sub> antiplasmin, Normotest, Thrombotest and fibrinogen determinations were performed at admission time. The onset of the symptoms varied from one to 45 days (mean 14 days) and the value of serum bilirubin at admission time ranged from 2 to 33.6 mg/dl (mean 7.4 mg/dl).

According to the final diagnosis established on the basis of the results of enzymatic pattern, viral serologic screening, chest radiograph, ultrasonography, endoscopic retrograde cholangiography, computed tomography, laparotomy or liver biopsy, the patients were divided into three groups without knowledge of the blood coagulation test results:

#### *Group 1 Hepatocellular jaundice, n=56*

Constituted by 34 patients with liver cirrhosis, 13 patients with acute viral hepatitis and nine patients with congestive heart failure.

**Group II Cholestatic jaundice, n=25**

Constituted by 13 patients with cholelithiasis, nine patients with cancer of the biliary tract without evidence of liver metastasis (pancreas six, cholangiocarcinoma three, two patients with primary biliary cirrhosis and one patient with sclerosing cholangitis).

**Group III Liver metastasis, n=11**

Constituted by six patients with gastrointestinal cancer, three patients with lung cancer, one patient with renal cancer, and one patient with gall bladder cancer.

**COAGULATION TESTS**

Blood samples were obtained by antecubital venepuncture using a tourniquet and a 1.2 mm steel needle. They were collected in plastic tubes containing 1/10 volume of 3.8% citrate solution for the performance of the blood coagulation tests and in a glass tube for the determination of serum bilirubin. Fibrinogen concentration was measured following the method of Clauss,<sup>17</sup> normal values: 200–400 mg/dl. Normotest and Thrombotest were determined following Owren's method,<sup>18, 19</sup> normal values: 70–100%. Antithrombin III, plasminogen, and alpha<sub>2</sub> antiplasmin were determined by chromogenic substrate (Kabi Vitrum S 2238 and S 2251) on Kemomat I (Analisis) respectively following the methods of Odegaard,<sup>20</sup> Friberger<sup>21</sup> and Edy.<sup>22</sup> The normal range of these parameters was established on 58 healthy ambulatory volunteers, free of medical or surgical disease: antithrombin III 73–129%, plasminogen 70–126%,

and alpha<sub>2</sub> antiplasmin 70–121%. Serum bilirubin was measured following the method of Routh,<sup>23</sup> normal values below 1 mg/dl.

**STATISTICAL ANALYSIS**

According to the non-Gaussian distribution of the blood coagulation tests in the different groups, the Wilcoxon's rank sum test was used for statistical analysis.<sup>24</sup> The percentage of abnormality of the tests in the different groups was analysed by the  $\chi^2$  test. The correlations were measured by the Spearman rank correlation coefficient.

**Results****VALUES OF THE DIFFERENT PARAMETERS IN THE THREE GROUPS**

The mean values of Normotest and of Thrombotest in the three groups were not significantly different. On the other hand, the mean values of antithrombin III, plasminogen, alpha<sub>2</sub> antiplasmin, and fibrinogen were lower in parenchymatous liver disease than in cholestatic jaundice with a highly significant difference ( $p < 0.01$  Wilcoxon's test) (Table 1).

As is shown in Table 2, Normotest and Thrombotest values were respectively abnormal in 40 out of 56 patients (71.4%) of group I and 13 out of 25 patients (52%) of group II. This difference was not statistically significant. The percentage of abnormal values of antithrombin III, plasminogen, alpha<sub>2</sub> antiplasmin, and fibrinogen, however, was higher in hepatocellular than in cholestatic jaundice with a highly significant statistical difference ( $p < 0.001$   $\chi^2$  test).

Table 1 Range and mean values of the different parameters in the three groups

Test	Group I Hepatocellular jaundice n=56	Group II Cholestatic jaundice n=25	Group III Liver metastasis n=11	Wilcoxon's test
Normotest				
Range (%)	12–100	10–95	34–95	
Mean (%)	52*	63*	68	*NS
Thrombotest				
Range (%)	11–100	4–100	16–100	
Mean (%)	55*	64*	66	*NS
Fibrinogen				
Range	84 mg/dl–590 mg/dl	187 mg/dl–900 mg/dl	229 mg/dl–640 mg/dl	
Mean	203 mg/dl*	446 mg/dl*	432 mg/dl	*p<0.01
Antithrombin				
Range (%)	5–117	43–129	60–115	
Mean (%)	50*	99*	84	*p<0.01
Plasminogen				
Range (%)	9–100	53–120	50–107	
Mean (%)	48*	92*	77	*p<0.01
Antiplasmin				
Range (%)	9–110	57–130	50–115	
Mean (%)	52*	96*	86	*p<0.01

Table 2 Percentage of abnormal results in the different groups

Test	Group I Hepatocellular jaundice (%) n=56	Group II Cholestatic jaundice (%) n=25	Group III Liver metastasis (%) n=11	$\chi^2$ test
Normotest < 70%	73*	52*	45.4	*NS
Thrombotest < 70%	73*	52*	45.4	*NS
Fibrinogen < 200 mg/dl	62.5*	4*	0	*p<0.001
Antithrombin < 73%	78.6*	8*	18.2	*p<0.001
Plasminogen < 70%	73.3*	8*	36.4	*p<0.001
Antiplasmin < 70%	71.5*	8*	27.2	*p<0.001

In parenchymatous liver disease, antithrombin III was the most frequently abnormal parameter (78.6% of the patients) and liver cirrhosis was more frequently associated with an abnormality of antithrombin III value than was acute hepatitis (91.1% vs 38.4% respectively).

The probability of presenting a hepatocellular jaundice according to the blood coagulation test results at admission time is shown in Table 3. Normotest and Thrombotest were poor discriminant coagulation parameters. When a low fibrinogen level was observed (39.8% of the patients with jaundice), the probability of presenting a hepatocellular jaundice was 97%. As far as a low antithrombin III value was concerned (52.7% of the patients with jaundice), the probability of presenting a hepatocellular jaundice was 92% and this probability decreased to 27% when antithrombin III value was normal.

Table 3 Probability of presenting a hepatocellular jaundice according to the blood coagulation test results at admission time

Test	Frequency observed in jaundice* (%)	Probability of hepatocellular jaundice† (%)
Normotest <70%	73.0	69.5
>70%	27.0	45.4
Thrombotest <70%	73.0	69.5
>70%	27.0	45.4
Fibrinogen <200 mg/dl	39.8	97.0
>200 mg/dl	60.2	37.0
Antithrombin <73%	52.7	92.0
>73%	47.3	27.0
Plasminogen <70%	51.7	87.0
>70%	48.3	33.0
Antiplasmin <70%	48.4	89.0
>70%	51.6	34.0

\*  $\frac{\text{Total number of patients with the test result}}{\text{Total number of patients}} \times 100$

†  $\frac{\text{Number of patients in group I with the test result}}{\text{Total number of patients with this result}} \times 100$

#### RELATION BETWEEN THE DIFFERENT COAGULATION TESTS

A close correlation was observed in jaundice between antithrombin III and plasminogen values ( $r=0.89$ ), between antithrombin III and  $\alpha_2$  antiplasmin values ( $r=0.92$ ) and between plasminogen and  $\alpha_2$  antiplasmin values ( $r=0.91$ ) ( $p<0.001$ ) (Fig. 1).

The relation between fibrinogen levels and antithrombin III values is shown in Figure 2. A low value of fibrinogen was associated with a low value of antithrombin III in 88.8% of the patients (32 out of 36) while a normal value of antithrombin III was associated with a normal value of fibrinogen in 90.7% of the patients (39 out of 43). Nevertheless, 30.3% of the patients (17 out of 56) with a normal fibrinogen level exhibited an abnormal value of antithrombin III (13 cases of group I, two cases of group II and two cases of group III) and 13 out of these 17 patients (76.4%) had an abnormality of Thrombotest.

As is shown in Figure 3, there was no correlation between antithrombin III values and Normotest results in cholestatic jaundice ( $r=0.11$ ) while a correlation between antithrombin III and Normotest results was observed in parenchymatous liver disease ( $r=0.72$ ;  $p<0.001$ ).

#### PROGNOSTIC SIGNIFICANCE OF THE BLOOD COAGULATION TESTS

In hepatocellular jaundice (group I), 16 out of 56 patients (28.6%) died in hospital, including nine patients with liver cirrhosis, five patients with congestive heart failure and two patients with acute viral hepatitis. The delay between admission time and death ranged from one to 37 days (mean 14 days). The causes of death were gastrointestinal bleeding (eight patients), cardiac failure (four patients), hepatorenal syndrome (three patients) and septicaemia (one patient). The blood coagulation test results at admission time were more impaired in these patients than in the survivors ( $p<0.01$  Wilcoxon's test) and 100% of the patients with a fatal evolution exhibited an abnormal value

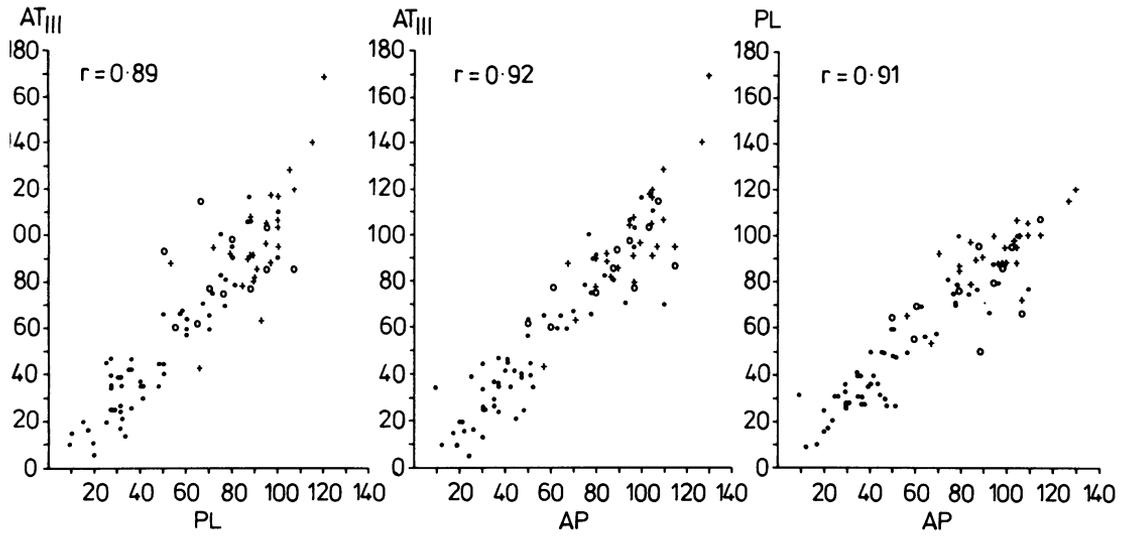


Fig. 1 Relation between antithrombin III (AT III), plasminogen (PL) and alpha<sub>2</sub> antiplasmin (AP) in jaundice. ● hepatocellular jaundice, + cholestatic jaundice, ○ liver metastasis.

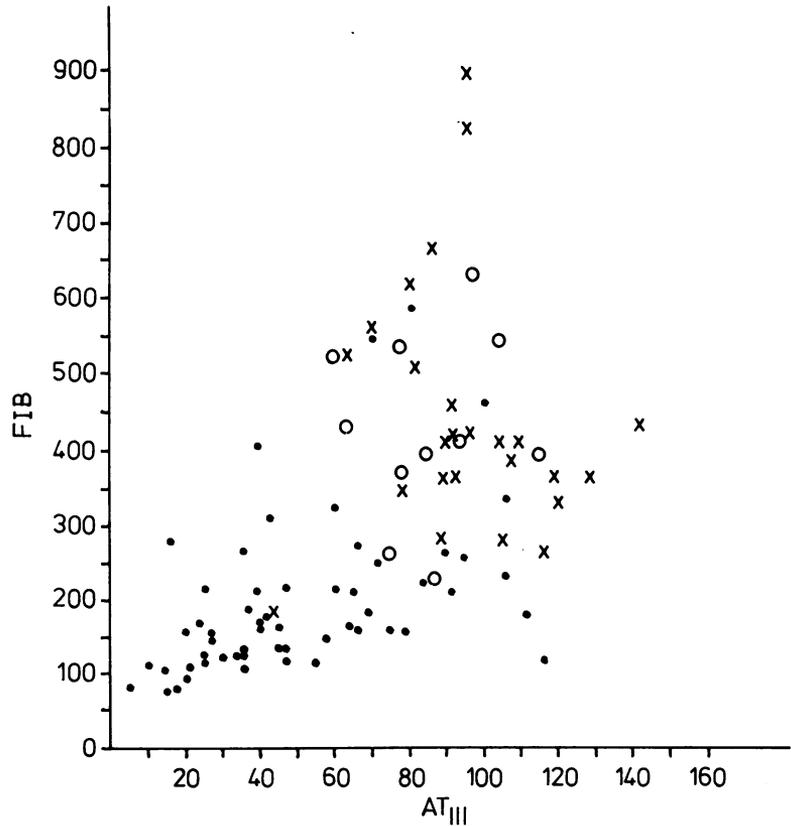


Fig. 2 Relation between antithrombin III (AT III) and fibrinogen (FIB) values.

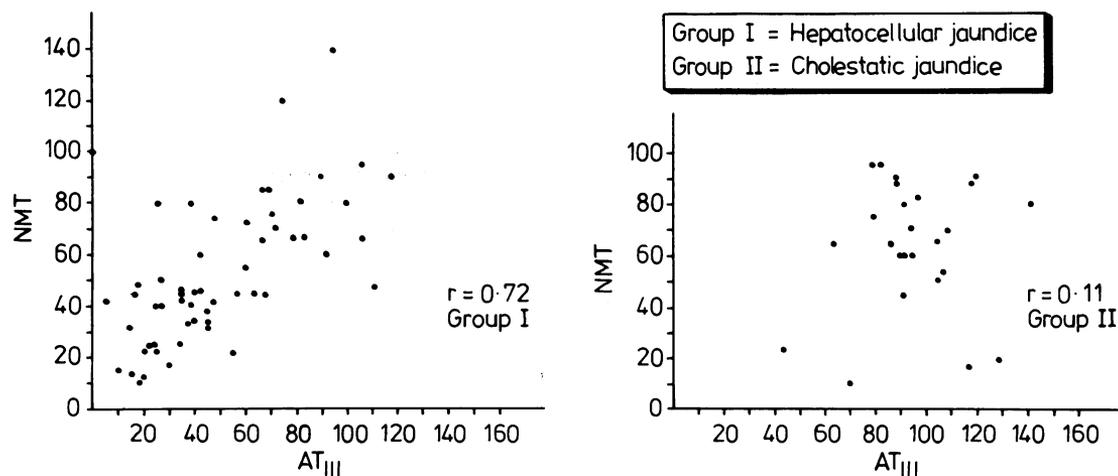


Fig. 3 Relation between antithrombin III (AT III) and Normotest (NMT) values.

of both antithrombin III, plasminogen, and  $\alpha_2$  antiplasmin at admission time. Table 4 shows the prognostic significance of the blood coagulation tests in hepatocellular jaundice. A normal value of antithrombin III, plasminogen or  $\alpha_2$  antiplasmin at admission time was never associated with death while normal values for Normotest, Thrombotest or fibrinogen were associated with a 12.5%, 12.5%, and 18.2% death rate respectively. On the other hand, when the blood coagulation test results were abnormal at admission time, the mortality rate ranged from 35–40%. In cholestatic jaundice (group II), the blood coagulation tests which were studied had no prognostic significance and abnormalities of Normotest and of Thrombotest depended upon the duration of the obstructive disease (abnormal in 90% of the cases of jaundice persisting for more

than 15 days and in only 26.6% of the cases present for less than 15 days).

In hepatic metastasis (group III), all the patients with abnormalities of the blood coagulation tests showed signs of massive infiltration of their liver at ultrasonography and died within one to 11 days (mean five days).

#### Discussion

Abnormality of haemostasis is frequently observed in patients with jaundice.<sup>1-16</sup> The Normotest and Thrombotest are prolonged in both hepatocellular and obstructive jaundice as they are vitamin K dependent while the vitamin K independent fibrinogen concentration is reduced only in parenchymatous liver disease.<sup>1-5</sup> The liver synthesis of antithrombin III, plasminogen, and  $\alpha_2$  antiplasmin has been recently shown.<sup>6-9</sup> The normal value of these parameters in cholestatic jaundice and the high correlation which existed in our series between antithrombin III, plasminogen, and  $\alpha_2$  antiplasmin values in hepatocellular jaundice suggested that their liver synthesis was closely linked and vitamin K independent. Moreover, this correlation showed that in parenchymatous liver disease, the enhanced fibrinolytic activity owing to the low concentration of  $\alpha_2$  antiplasmin was counterbalanced by an hypercoagulability state because of the low concentration of antithrombin III.

Among the blood coagulation tests which were studied, the antithrombin III was the most discriminant in differentiating hepatocellular from

Table 4 Prognostic significance of the blood coagulation tests

Test	Death (%)	$\chi^2$ test
Normotest <70%	35.0	NS
>70%	12.5	
Thrombotest <70%	35.0	NS
>70%	12.5	
Fibrinogen <200 mg/dl	35.2	NS
>200 mg/dl	18.2	
Antithrombin <73%	36.4	p<0.05
>73%	0	
Plasminogen <70%	39.0	p<0.01
>70%	0	
Antiplasmin <70%	40.0	p<0.01
>70%	0	

cholestatic jaundice. An abnormal value of antithrombin III strongly suggested the presence of parenchymatous liver disease while a normal value was in favour of jaundice due to another cause. Further investigations will be required to determine the discriminant value of antithrombin III compared with the classical enzymatic patterns in differentiating the origin of jaundice.

In our study, a relation between antithrombin III value and fibrinogen concentration was observed. Most of the patients with a low fibrinogen concentration had a low antithrombin III value and the majority of the patients with a normal antithrombin III value had a normal fibrinogen concentration. Nevertheless, one third of the patients with a normal fibrinogen value exhibition an abnormal value of antithrombin III and most of these patients presented an abnormality of Thrombotest. Arousseau has previously shown a close correlation between plasma antithrombin III and prothrombin concentrations in patients with liver cirrhosis.<sup>11</sup> These observations suggested that antithrombin III values were abnormal in both severe hepatic failure (with low fibrinogen concentrations) and in moderate liver insufficiency (with normal fibrinogen concentrations and abnormal values of Thrombotest).

The main interest of the blood coagulation tests which were studied, was in their prognostic significance in parenchymatous liver disease. Normal values of antithrombin III, plasminogen or alpha<sub>2</sub> antiplasmin were never associated with death and therefore indicated a good prognosis. On the other hand, abnormal values of antithrombin III, plasminogen or alpha<sub>2</sub> antiplasmin at admission time, were associated with a death rate of 36.4%, 39%, and 40% respectively. Normotest, Thrombotest, and fibrinogen determinations did not allow a prediction of the patients' prognosis with the same accuracy.

In conclusion, among the blood coagulation tests which were studied, the antithrombin III was the most discriminant factor in differentiating hepatocellular from cholestatic jaundice. Furthermore, antithrombin III, plasminogen, and alpha<sub>2</sub> antiplasmin appeared to provide a good index of the liver cell function and were superior to the Normotest, Thrombotest, and fibrinogen concentration in predicting the prognosis of the patients at admission time.

The authors are indebted to Professor I A D Bouchier for his critical review of the manuscript. This study was presented at the IXth International Congress on Thrombosis and Haemostasis, Stockholm, Sweden, July 1983.

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