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# LIVER DYSFUNCTION IN HAEMOPHILIA

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**Summary.** *Liver function was studied in 139 of 291 haemophiliacs known to a single Regional Haemophilia Centre including patients with classical haemophilia, Christmas disease and von Willebrand's disease. In 57 patients, six-monthly liver function tests over a five year period were also available. Thirty-nine of the 139 patients had had jaundice or hepatitis and 56 had a positive test for HBsAb in the blood although few of these had had an identifiable clinical illness. Fifty-eight haemophiliacs had elevated serum aminotransferases at the time of study, but the five year review revealed only six patients who had had persistently abnormal results, although none had clinically evident liver disease. Liver dysfunction was unrelated to a history of hepatitis, to a positive HBsAb test, or to age, type of haemophilia, factor level or frequency of factor replacement treatment. Abnormalities of liver function in haemophilia appear to be unrelated to past or present hepatitis B infection in most cases and may not be related to any single transmitted infectious agent.*

Key words: Haemophilia, liver dysfunction, hepatitis.

A MAJOR problem associated with the repeated transfusion of donor blood products into patients with hereditary bleeding disorders is the high prevalence of liver dysfunction (1). A high proportion of haemophiliacs have abnormally increased liver enzymes in the blood (1-11). In a small proportion of cases liver enzyme abnormalities are related to clinical episodes of jaundice and in some of these the hepatitis B virus (HBV) can be positively implicated (1, 8, 10). In some patients multiple episodes of jaundice, negative on testing for HBV, encouraged the search for other infectious agents and the existence of a non A, non B virus was suggested (12, 13). Only a proportion of icteric episodes have however been positively related to non A, non B hepatitis due to difficulties in identifying a single infectious agent and obtaining reliable serological diagnostic tests (14). It has recently been suggested that in some instances liver dysfunction may be a hypersensitivity reaction to the foreign protein contained in the factor replacement treatment (15) rather than a viral infection.

Sensitive tests for evidence of previous HBV infection are now available and a high prevalence of HB surface antibody (HBs Ab) indicating previous HBV infection is seen in most haemophiliac populations (1, 2, 5, 7-10, 16-18). Since most patients have not had an overt icteric illness they are presumed to have had subclinical infection (1, 7, 8) and only a few remain infectious carriers of HBV.

Clinical evidence of liver disease has been uncommon in haemophilia but recent liver biopsy studies (5, 6, 19-22) suggested changes similar to chronic persistent and chronic active hepatitis in a proportion of cases. This would suggest that cases of chronic liver disease will be seen more frequently in the haemophiliac population in future. We have therefore examined the current prevalence of liver abnormalities in the haemophiliac population of the West of Scotland and reviewed the liver function of 57 patients prospectively for five years and related the results to the type and severity of haemophilia and the frequency of factor replacement treatment. In addition we have

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carried out immunological tests and related the results to those of liver function to examine the possible relevance of immune factors in haemophiliac liver dysfunction.

### Patients and methods

One hundred and thirty nine, out of 291 patients, known to the Regional Haemophilia Centre at Glasgow Royal Infirmary and the Royal Hospital for Sick Children with hereditary bleeding disorders were seen and examined by a single clinician (Group 1) as part of a study on the prevalence, severity and pathogenesis of haemophilic arthritis. Details of type and severity of haemophilia [Factor level in one stage Factor VIII or IX assays (23)], age, annual factor requirement and presence of haemophilic arthritis were recorded. Definite haemophilic arthritis was recorded if there was significant joint restriction (loss of more than 10% of the full normal range of joint movement) and or contracture (loss of more than 10° of extension) with significant radiological changes (narrowing or irregularity of the joint space with erosions of cysts in the juxtaarticular bone) in one or more joints. Possible haemophiliac arthritis was designated if there were lesser clinical and radiological changes in an individual with severe haemophilia (factor level <1% normal) or moderately severe haemophilia (factor level 1-5% normal). Included in the information obtained were details of previous hepatitis or jaundice and blood samples were tested for evidence of past or present hepatitis B infection (HBsAg by Ausria II, HBs Ab by AUsab and HBcAB by Corab, Abbott Laboratories, Basingstoke). Liver function tests including aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and albumin, were estimated by automated methods (Technicon), and related to the normal levels cited by the laboratory. Limited details were available from the case sheets of the remaining 152 patients (Group II) but where possible the same data were also collected. A review of case sheets revealed 57 patients in whom liver function testing had been carried out at least every six months over a five year period (53 from Group I and four from Group II). Eighteen of these patients had been diagnosed as having 'hepatitis' over the review period. These 57 patients were the subject of a more detailed analysis of liver function. Patients with persistently abnormal aminotransferases for three or more years were classed as chronic hepatitis (CH) and those with periodic abnormalities as intermittent hepatitis (IH). Immunological studies performed included those for rheumatoid factor (Latex agglutination test, Wellcome Laboratories and Rose Waaler with sheep red cell labelled with rabbit anti-sheep antibody, Nordic), complement components (including CH50, C3, C4 and C3d measured as previously described) (24) and immune complex assays (Raji and Ciq tests which were carried out on 42 randomly selected patients: see acknowledgements).

Liver biopsies were not carried out but post mortem histology was available on patients who died of unrelated causes after the clinical study. Statistical analysis was carried out by the appropriate non parametric test (25).

### Results

No patients had hepatomegaly, splenomegaly, or other evidence of chronic liver disease. Thirty-nine of the 134 (29%) Group I patients and 13 of 44 (30%) Group II in whom information was available gave a definite history of jaundice or hepatitis (Table I). In a few cases there had been proven hepatitis B infection with seroconversion from HBsAb negative to positive status and a relationship to blood product transfusions. In some patients multiple episodes of jaundice had occurred. People with a history of jaundice were older and more frequently had definite haemophiliac arthritis but there was no significant relationship to type or severity of factor deficiency, presence of inhibitors or frequency of factor replacement treatment. The majority of patients had serological evidence of previous HBV infection (Table I) as evidenced by a positive HBs Ab but in few cases had there been a corresponding clinical episode of jaundice. The HBs Ab results were confirmed by HBcAb in 53 of 70 patients tested. There was no significant correlation ($X^2$) between a history of jaundice and a positive HBs Ab (Table II). Only three patients were positive for HBS Ag and had been chronic carriers for two to five years. One adult, had HBe Ab indicating low infectivity. Two Group 2 patients were noted to have positive HBs Ag. Patients with a positive HBsAb had more severe haemophilia and had received treatment more frequently than those who were HBsAb negative (Table I). Forty-eight Group I patients had serum aminotransferases above the normal laboratory reference range at the time of review (Table I), and five of 33 Group II patients in which information was available also had elevated aminotransferases. The elevation in enzymes was not correlated significantly ($X^2$) with HBs Ab carriage or previous jaundice (Table II) nor with age, severity of disease, frequency of treatment, recent factor therapy, presence of arthritis or with any laboratory abnormality. Serum levels of ALP and albumin were within the normal range in all patients.

The five year review of liver function showed only six patients to have persistently normal results over this period (Table III).

**Table I.** Liver function in haemophilia.

| | History of Jaundice/hepatitis | | AST | | HBsAb | |
|---|---|---|---|---|---|---|
| | Positive (39) | Negative (95) | Elevated (48) | Normal (82) | Positive (56) | Negative (48) |
| Age (median) | 32 | 19*** | 30 | 24 | 30 | 27.5 |
| Factor level (median %) | 3 | 4 | 2.5 | 5 | 1 | 5** |
| Treatments/Year (median) | 2 | 4 | 7.5 | 2 | 10 | 1** |
| Haemophilic arthritis | | | | | | |
| — Definite | 25 | 32 | 22 | 35 | 33 | 18* |
| — Possible | 0 | 20 | 8 | 9 | 6 | 5 |
| — None | 10 | 36 | 14 | 31 | 12 | 20 |
| — Other arthritis | 4 | 7 | 4 | 7 | 5 | 5 |

*p<0.05
**p<0.05 } Mann Whitney U Test
***p<0.001 } $X^2$ TEST

**Table II.** Correlation between a history of jaundice, elevated transaminases and positive HBsAb tests.

| | History of jaundice | | HBsAB | |
|---|---|---|---|---|
| | Positive | Negative | Positive | Negative |
| AST elevated | 15/38 | 29/88* | 21/55 | 19/46* |
| Positive history of jaundice | — | — | 20/54 | 17/47* |

*$X^2$ all n.s.

**Table III.** Liver function tests over five years in 53 haemophiliacs.

| | Normal | Intermittent hepatitis | Chronic hepatitis |
|---|---|---|---|
| Number | 6 | 30 | 17 |
| Median age (years) | 48 | 30 | 36 |
| Factor deficient: VIII | 3 | 25 | 13 |
| : IX | 2 | 5 | 3 |
| Median factor level (%) | 5 | 1 | 1 |
| Median treatments/year | 7 | 30 | 19 |
| Median no of products | 2 | 4 | 3 |
| Mean ALT level* | 1 | 1.7 | 3.1 |
| Median % ALTs abnormal | 0 | 29 | 100 |
| Median no ALT 6 × normal | 0 | 0.6 | 0.9 |
| History of hepatitis | 0 | 8 | 5 |

*1 = normal
2 = 1 – 2 × normal
3 = 2 – 3 × normal
etc.

Thirty patients (including nine with a history of clinical hepatitis) had IH over the five years and 17 (including five with a history of clinical hepatitis) had CH. Patients with CH or IH could not be distinguished from each other on the basis of age, type or severity of factor deficiency, frequency of treatment or by the number of different factor concentrate preparations used. Both groups of patients were younger, had more severe disease, more frequent treatment and a greater variety of cencentrate preparations than the six patients with persistently normal liver function.

During the five-year review period 18 of the 57 patients (including 14 of the above) had episodes which were diagnosed as 'acute hepatitis'. In 15 cases there were typical clinical pictures of acute hepatitis associated in three cases with positive blood test for HBs Ag and in a further case with the development of higher titre HBs Ab. No infectious agent was implicated in the remaining 11 cases despite extensive testing and the three HBs Ag positive cases subsequently developed HBs Ab. Two further cases acquired HBs Ag without clinical illness and the last patient was a known chronic carrier of HBs Ag.

Consideration of possible non infectious causes for liver dysfunction included extensive immunological testing. Rheumatoid factors were detected in 24 patients but only four had a titre greater than 1/32. Low titre rheumatoid factors were more common in those with a history of jaundice (11/32 *vs* 11/84, $p<0.05$, $X^2$ test) and in those carrying HBs Ab (16/54 *vs* 5/46, $p<0.05$, $X^2$ test). The complement breakdown component C3D (elevated in 78 of 115 patients tested) was increased in patients with elevated AST levels (16.3 *vs* 12.6, median units/ml, $p<0.01$; Mann Whitney 'U' test) but depressed

Table IV. Prevalence of abnormal transaminases and of markers of hepatitis B virus infection in haemophiliacs.

| Author | Country | Year | No. | HBsAg+% | %HBsAB+ | % with elevated transaminases |
|---|---|---|---|---|---|---|
| Blumberg | USA | 1965 | 30 | – | 9 | – |
| Forbes | UK | 1972 | 78 | 5 | 13 | – |
| Yannitsiotis | Greece | 1975 | 131 | 25 | 65 | – |
| Mannucci | Italy | 1975 | 91 | 8 | 66 | 45 |
| Webster | USA | 1976 | 261 | – | – | 57 |
| Hilgartner | USA | 1976 | 89 | – | 60 | 41 |
| Levine | USA | 1977 | 98 | – | – | 68 |
| Spero | USA | 1978 | 120(87) | 11 | 88 | (38) |
| Preston | UK | 1978 | 36 | – | – | 77 |
| McVerry | UK | 1979 | 42 | 0 | 83 | 50 |
| Gerety | USA | 1980 | 136 | – | 90 | – |
| Kim | USA | 1980 | 103 | 12 | 77 | 79 |
| Cederbaum | USA | 1982 | 1205(506) | 5 | 70 | (20*) |
| Rickard | Australia | 1982 | 234 | 5 | 63 | 34 |
| Thomas | UK | 1982 | 76 | – | – | 92 |
| Steven | UK | 1983 | 104(130) | 2 | 55 | 37 |

*Persistently abnormal for 18 months.

CH50, C3, or C4 seen in a few patients did not relate to liver dysfunction. Twenty of 42 patients tested had positive tests for immune complexes (Raji and or Ciq binding) but the results did not correlate with those of any liver function test studied. Liver histology was examined in two patients who died; one from septicaemia following emergency surgery for diverticular disease and the other from a cerebral haemorrhage. The former had shown mild elevation of serum aminotransferases and a positive HBsAb test at the time of original review but the liver histology was normal at post-mortem. The latter patient, a known hypertensive, treated for many years with methyldopa, had shown only slight elevation of serum aminotransferases and a negative HBsAb at the review clinic but the liver showed steatosis, inflammatory cell infiltrate and piecemeal necrosis round the portal tracts at autopsy. The picture was considered a chronic active hepatitis and was consistent with the pattern described in haemophilic liver biopsies or with a methyldopa induced picture.

**Discussion**

An early consequence of the increased use of blood donor products to treat bleeding episodes in haemophiliacs was the increased incidence of transfusion associated hepatitis (1). Some cases were discovered to be due to HBV infection transmitted from the blood product donor but sensitive testing for HBV infection in donors reduced but did not stop this problem (10, 26). These episodes of hepatitis were more often associated with commercial blood products prepared from large pools of donors. Overt clinical jaundice followed transfusion in only a minority of cases (5, 9, 16) and hepatitis B has been positively implicated in few cases (1, 8 10). Sensitive assays of HBs Ab however, indicate a high rate of subclinical HBV infection (1, 2, 5, 7, 9, 10, 16-18) (Table IV) in haemophiliacs. In some cases serum from patients with jaundice or the implicated blood products produced hepatitis in animals (27, 28, 33) and a new infectious agent called non A, non B hepatitis was suggested. The varying incubation and features of non A, non B hepatitis (12, 13) suggested that a number of different agents could be responsible for the episodes of jaundice and chronically abnormal liver enzymes in haemophiliacs. Not all episodes of 'hepatitis' may be attributable to infectious agents (15), however, and disordered liver function is a feature of a variety of non-infectious illnesses. Our results would

confirm previous reports that the abnormal liver function seen in a high proportion of haemophiliacs (Table V) appear unrelated to concurrent hepatitis B Infection (30). Abnormal transaminases were unrelated to past or present HBV infection, previous jaundice, age, severity of disease or amount of treatment. The presence of HBs Ab however indicating past, often subclinical, HBV infection was, as previously noted (10), related to severity of disease and amount of treatment. Although the prevalence of HBs Ab is considered to preclude current active HBV infection it is noteworthy that HBV was demonstrable in eight of 12 liver biopsies from patients with HBs Ab in the blood (5). It might be suggested that low grade inflammation rather than an icteric illness might result from frequent exposure of such patients to HBV (7) or alternatively the presence of both antigen and antibody could lead to immune complex deposition (31). Although this could also theoretically cause joint disease (32) and we did find joint disease more frequently in those with a history of jaundice or positive HBs Ab, we could not relate the presence of circulating immune complexes to liver dysfunction. This is in keeping with the findings of McVerrey *et al.* (31). The more recent emphasis on non A, non B hepatitis as a cause of liver dysfunction is of importance as studies suggest a frequent progression to persistently abnormal liver function tests and severe abnormalities on liver biopsy (13, 33). Although similar results have been predicted for haemophiliacs (6) the studies of Manucci *et al.*, who repeated liver biopsies after a period of three years, showed no deterioration in the pathological findings (20, 21). Furthermore, clinical cases of serious liver disease has been reported infrequently in haemophiliacs.

An alternative explanation for the large number of patients affected by liver dysfunction is that a non-infectious agent is responsible. A variety of non-infectious conditions including toxicity from alcohol and chemicals, adverse drug reactions, and a number of immunologically mediated diseases (34) may all cause liver dysfunction. The Kupffer cells of the liver, as part of the reticulo-endothelial system are responsible for the clearance of much undegradable antigenic material from the circulation (34). The burden placed by continued, repeated infusion of foreign protein could conceivably directly excite a toxic or immunologically mediated inflammatory reaction. One convincing case of recurrent hepatitis with an allergic basis has been reported (15). This pattern of illness was not seen however in any of the present cases. Although rheumatoid factors, immune complexes and complement consumption were seen frequently the poor correlation with the different abnormalities of liver function would argue against a primary pathogenetic role for the immunological findings in haemophiliac liver dysfunction.

Despite increased care in the screening of factor concentrates for infectious agents elevated transaminases and HBs Ab are apparent in a high proportion of young patients treated by the best modern therapies (30). It remains to be seen whether one or several transmissible infectious agents may be identified or whether alternative mechanisms of hepatotoxicity can be positively implicated. Long term studies are also required to study the clinical significance of the frequent laboratory abnormalities.

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