Unsolved Problems 3/-6
PP 51-56
Proceedings of a conference sponsored by the Bureau of Biologics (1986)

Viral Antigens and Antibodies in Hemophiliacs

ROBERT J. GERETY and LEWELLYS F. BARKER

Division of Blood and Blood Products
Bureau of Biologics
Food and Drug Administration
Bethesda, Maryland 20014

INTRODUCTION

The emergence of data showing a very high prevalence of asymptomatic liver disease in hemophiliacs prompted the assessment of the status of detectable viral antigens and antibodies among these patients in view of the possibility that the observed liver disease results from chronic infection with one or more hepatitis viruses. Viral hepatitis has traditionally included types A and B, but recent studies have provided considerable evidence for a third category, termed non-A, non-B hepatitis, which is presumably caused by an as yet unidentified agent or agents. Serologic studies of viral antigens and antibodies in hemophiliacs have concentrated on hepatitis B surface antigen (HB_sAg) and antibodies to the surface antigen (anti-HB_s) and core antigen (anti-HB_c) associated with the hepatitis B virus (HBV). A better understanding of chronic liver disease in hemophiliacs will probably require future efforts to evaluate their experience with other agents capable of causing liver damage, including hepatitis A virus (HAV), members of the herpes virus family including the cytomegaloviruses (CMV) and Epstein-Barr virus (EBV) and the identification of the agent or agents of non-A, non-B hepatitis.

HEPATITIS B VIRUS SEROLOGY

The discovery of HB_sAg, originally termed Australia antigen, was made using sera from hemophiliacs as antibody sources in Ouchterlony agar gel diffusion during a search to detect alloantigens in human sera [1]. Early studies using this very insensitive method for antibody detection revealed anti-HB_s in 30 percent or less of moderate and severly effected hemophiliacs, but the application of much more sensitive passive hemagglutination (PHA) and radioimmunoassay (RIA) techniques revealed that anti-HB_s can be identified

BAYP0000020_004_0001

in the sera of more than 80 percent of patients with this disease [2,3]. These results are not surprising in view of the high risk of exposure to HBV incurred by hemophiliacs who require treatment over many years which entails massive exposure to blood and blood products. To obtain information about the risk of HBV infection to hemophiliacs from single donor cryoprecipitate and from commercial concentrates, sera from three groups of patients were studied. One group of hemophiliacs (Group I) was treated almost entirely by single donor cryoprecipitate, the second by a combination of single donor cryoprecipitate and commercial concentrates (Group II) and the third almost entirely by commercial concentrates (Group III). These patients varied widely in age but in general, required repeated hospital based therapy. To determine the prevalence of HBV infection among these groups, the prevalence of HB_sAg, antibody to HB_sAg (anti-HB_s) and antibody to the hepatitis B core antigen (anti-HB_c) was determined. A summary of these results appears in Table 1. The prevalence of hepatitis B carriers among the three groups was 3% in Group I, 7% in Group II and 3% in Group III while the prevalence of anti-HB_s was 87%, 75%, and 66%, respectively; 21% of individuals in all three groups possessed anti-HB_c. Despite these data suggesting that the prevalence of HBV infection is independent of the use of cryoprecipitate or commercial concentrates in heavily treated groups, mild hemophiliacs who require infrequent treatment should probably receive single donor products to lessen the risk of exposures to HBV and to the agent or agents of "non-A, non-B" hepatitis.

Considering the large percentage of hemophiliacs with serologic evidence of past or ongoing HBV infections, the relatively low prevalence of chronic type B hepatitis as well as a history of clinically recognizable hepatitis in this group is somewhat surprising. Although data regarding overt clinical hepatitis among hemophiliacs are inadequate to assess the real incidence of type B hepatitis, some figures are available. Those for chronic, type B hepatitis range from 2.5 to 7.8% and those for clinically recognizable hepatitis range from 6 to 26% [4]. The patients with hemophilia who are at particularly high risk of developing clinical hepatitis and of becoming chronically infected appear, respectively, to be those with mild disease who require infrequent treatments and those who receive commercial concentrates for the first time as older children and adults. In a study by Kasper and Kipnis [5] reported in 1972, 16 of 29 patients with hemophilia A or B who developed clinical hepatitis over a 10-year period of study had either never received concentrate before or had received it more than six months prior to the most recent treatment implicated in the development of hepatitis.

Although liver enzyme elevations are rather common in these patients, the significance of these biochemical changes vis-a-vis chronic liver disease is not entirely clear. Briefly, elevated liver enzymes appear to be present in similar numbers of patients among the three treatment study groups outlined above irrespective of the modes of therapy, although all, as mentioned earlier, required repeated hospital based therapy.

BAYP0000020_004_0002

TABLE 1. Evidence of HBV Exposures Among Groups of Patients with Hemophilia

Group	Treatment	HB_sAg*	Anti-HB_s	Anti-HB_c	One or More (HB_sAg anti-HB_s, anti-HB_c)
I	Single donor cryoprecipitate	3% (1/32)	87% (28/32)	21% (7/32)	90% (29/32)
II	Single donor cryoprecipitate and commercial concentrates	7% (7/100)	75% (75/100)	21% (21/100)	82% (82/100)
III	Commercial concentrates	3% (9/268)	66%**(178/268)	21% (32/147)	69% (187/268)
	TOTAL	4% (17/400)	70% (281/400)	22% (60/279)	75% (298/400)

*HB_sAg detected by RIA. Anti-HB_s detected by passive hemagglutination (PHA)** or RIA. Anti-HB_c detected by CF.

HEPATITIS RISK FROM PLASMA DERIVATIVES

The risk of HBV infection associated with the administration of Factor IX complex concentrates and Factor VIII (AHF) is very high. The use of large plasma pools from paid donors no doubt contributes to the risk of HBV infection from these products. The use of single donor units of cryoprecipitate to treat Factor VIII deficiency presumably carries the same risk of HBV infection as single units of whole blood. Over a period of time, however, the cumulative hepatitis risk of single donor cryoprecipitate could be equivalent to the high risk associated with commercial concentrates as suggested by the similar prevalence of a positive HBV serology among patients in different treatment groups.

Clinical type B hepatitis in recipients of AHF has not been a major problem almost surely because of the residual immunity among older hemophiliacs from prior exposure to hepatitis B virus evidenced by the high prevalence of anti-HB_s. Administration of commercial AHF products to young hemophiliacs, however, may more often result in asymptomatic disease and chronic HBV infections [6]. In contrast, Factor IX complex concentrates have an unenviable record of transmitting type B hepatitis to recipients [7,8]. These cases, however, are largely confined to situations where the use of Factor IX complex occurred in patients other than those with a congenital deficiency of this factor or in hemophilia B patients with minimal previous transfusions.

The high risk of type B hepatitis following the administration of Factor IX complex or AHF is reflected in the prevalence of HB_sAg in each of these products as shown in Table 2 and in the absence of treatment of these heat-labile products likely to inactivate viruses. The dramatic decrease in the prevalence of HB_sAg positive lots from 67% to 2% for Factor IX complex and from 25% to 3% for AHF over the period from prior to 1972 through 1975 almost surely represents the effect of HB_sAg screening of donors [9,10]. Since

BAYP0000020_004_0003

TABLE 2. Prevalence of Detectable HB_sAg by RIA in Lots of Factor IX Complex and AHF Submitted to the Bureau of Biologics

	Prior to 1972	1973	1974	1975
Prevalence in Factor IX Complex	67% (10/15)	53% (19/36)	52% (41/79)	2% (2/91)
Prevalence in Factor VIII (AHF)	25% (58/232)	22% (51/228)	7% (41/580)	3% (20/606)

July 1975 no lots of either Factor IX Complex or AHF submitted to the Bureau of Biologics contained HB_sAg detectable by either RIA or reversed passive hemagglutination. These results are encouraging, but despite the exclusion of HB_sAg positive donors and the absence of detectable HB_sAg in AHF and Factor IX complex, the problem of HBV infections following the administration of these factors will most likely remain serious.

APPROACHES TO HBV PREVENTION

Several measures have been undertaken with the hope of conferring greater safety for hemophiliacs from the hepatitis B virus. Since September 15, 1975 all blood, plasma and serum for transfusion as single donor products or further manufacture into plasma derivatives has been required to be tested by one of the most sensitive tests available to detect HB_sAg, the radioimmunoassay (RIA) or reversed passive hemagglutination (RPHA) tests [10]. An all voluntary blood donor system is being pursued as a result of the known increased risk of PTH from blood derived from commercial donors [11]. Transfusion practices utilizing single donor products are being encouraged, particularly for those hemophiliacs not requiring frequent treatment. Research in a number of laboratories has been directed towards developing means of removing HBV from final products while maintaining the labile clotting factors. Pilot studies have been undertaken at the Bureau of Biologics in conjunction with Drs. Stanley Charm and Bing Wong of the Tufts University School of Medicine and Dr. Alan Johnson of the New York University School of Medicine to evaluate two such methods (solid-phase immunoadsorption and polyethylene glycol precipitation) and their ability to reproducibly remove HB_sAg and HBV infectivity from high risk plasma derivatives. Evaluation of products treated by both methods and then injected into susceptible chimpanzees have yielded somewhat equivocal results. Each is clearly capable of removing HB_sAg and low levels of HBV as judged by chimpanzee inoculations with treated products known to be infectious prior to treatment.

Studies using high titer anti-HB_s globulins (HBIG) to prevent HBV infections are at the moment being evaluated with suggestive evidence favoring efficacy

BAYP0000020_004_0004

in certain specified settings [12]. A summary of a number of clinical trials was presented at a HBIG Workshop held at the National Institutes of Health on December 17, 1975.

The need for repeated treatment would appear to make hemophiliacs prime candidates for active immunization against HBV by vaccine. Several such hepatitis B vaccines are now undergoing preliminary testing in the United States [13,14].

OTHER VIRAL AGENTS AS CAUSES OF HEPATITIS

No data appear in the literature concerning the risk to hemophiliacs of hepatitis A virus infections following the administration of blood products. The hepatitis A virus (HAV) is spread mainly by the fecal-oral route and does not appear to be commonly spread by transfusion of blood and blood products or to be associated with a carrier state. Therefore HAV probably does not represent a significant risk to hemophiliacs from replacement therapy. A limited number of post-transfusion, "non-B" hepatitis cases in hemophiliacs investigated by the Bureau of Biologics have also been found to be "non-A" hepatitis based on the absence of antibodies to HAV in the serum of these patients during convalescence. Information is also lacking about the more likely candidates in hepatitis amoung hemophiliacs, namely CMV and EBV.

The diagnosis of "non-A, non-B" hepatitis is at the moment one of exclusion, accomplished by eliminating HBV and HAV by appropriate serologic tests. Little data are available regarding the risk of this agent or agents for hemophiliacs from treatment [15]. From the available rates of post-transfusion hepatitis (PTH), the agent or agents of "non-A, non-B" hepatitis appear to be blood borne, perhaps to be associated with a form of chronic hepatitis, and to represent a considerable risk to recipients who repeatedly require the administration of blood products. Much information is needed regarding this category of hepatitis with respect to risk to hemophiliacs of chronic hepatitis, identification of the agent or agents, viability in plasma derivatives and means of treatment and prevention.

The standard association of the presence of anti-HB_s with immunity to HBV infection leads one to the conclusion that the majority of hemophiliacs are immune to HBV. Whether repeated exposures to HBV result in multiple, sequential HBV infections with recovery or merely antigenic stimulation with boosts in anti-HB_s titers is not known. In the former case, re-infections could lead to chronic liver disease. In the latter case it is difficult to implicate HBV as the cause of the chronic elevations of liver enzymes seen in hemophiliacs except on an immunological basis. An added benefit of acquired immunity as a result of HBV vaccination to prevent type B hepatitis could be the prevention of chronic liver disease if it proves to be related to HBV.

BIBLIOGRAPHY

1. Blumberg BS, HJ Alter and S Visnich: A "new" antigen in leukemia sera. JAMA 191:541-546, 1965.

BAYP0000020_004_0005

2. Peterson MR, LF Barker and DS Schade: Detection of antibody to hepatitis-associated antigen in hemophilia patients and in voluntary blood donors. Vox Sang 24:66-75, 1973.

3. Lander JJ, HJ Alter and RH Purcell: Frequency of antibody to hepatitis-associated antigen as measured by a new radioimmunoassay technique. J Immunol 106:1166-1171, 1971.

4. Nordenfelt E and IM Nilsson: Antibodies against Australia antigen in hemophilia. Europ J Clin Biol Res 16:151-155, 1971.

5. Kasper CK and SA Kipnis: Hepatitis and clotting factor concentrates. JAMA 221:510, 1972.

6. Gerety RJ, JA Markenson, JH Hoofnagle and LF Barker: Exposure to hepatitis B virus and development of the chronic HB_sAg carrier state in children. J. Peds 84:661-665, 1974.

7. Faria R and NJ Finmara: Hepatitis B associated with Konyne. N Engl J Med 287:358-359, 1972.

8. Oken MM, L Hostkin and RL Dejager: Hepatitis after Konyne administration. Am J Digest Dis. 17:271-274, 1972.

9. Hoofnagle JH, RJ Gerety, J Thiel and LF Barker: The prevalence of hepatitis B surface antigen in commercially prepared plasma products. J Lab Clin Med In Press.

10. Rules and Regulations, Federal Register, Volume 40, No. 136, p. 29706-29712, July 15, 1975.

11. Seefe LB, EC Wright, HJ Zimmerman and RW McCollum: VA cooperative study of post-transfusion hepatitis 1969-1974: Incidence and characteristics of hepatitis and responsible risk factors. Am J Med Sci 270:355-362, 1975.

12. Redeker AG, JW Mosley, DJ Gocke, AP McKee and W Pollack: Hepatitis B immune globulin as a prophylactic measure for spouses exposed to acute type B hepatitis. N Eng J Med 293:1055-1059, 1975.

13. Purcell RH and JL Gerin: Hepatitis B subunit vaccine: a preliminary report of safety and efficacy tests in chimpanzees. Am J Med Sci 270: 395-399, 1975.

14. Hilleman MR, EB Buynak, RR Rochm, AA Tytell, AU Bertland and GP Lamson, Purified and inactivated human hepatitis B vaccine: progress report. Am J Med Sci 270:401-404, 1975.

15. Craske J, N Dilling and D Stern: An outbreak of hepatitis associated with intravenous injection of Factor-VIII concentrate. Lancet 2:221-225, 1975.

BAYP0000020_004_0006