Persistence of Serum Antibody to Hepatitis B Core Antigen

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The purpose of the present study was to measure the amount of antibody to hepatitis B core antigen (anti-HBc) in different populations by the immunoelectroosmophoresis method. High titers of anti-HBc, up to 1/4,096, were found in the acute stage of hepatitis B virus infections and in the chronic carrier state of hepatitis B surface antigen. In cases of acute hepatitis the anti-HBc titers gradually declined to low levels but persisted for the observation time of 5 to 6 years. Individuals positive for antibodies to hepatitis B surface and core antigens selected from a Swedish "normal" population showed still lower anti-HBc titers, indicating that the hepatitis B infection had occurred earlier. The anti-HBc titers in sera drawn at intervals of 4 years from a group of hemophilia patients may indicate previous infection with replicating hepatitis B virus rather than immunization with noninfectious hepatitis B core antigen material.

The Dane particle, considered to be the hepatitis B virus, contains at least two separate antigens. The outer coat of the Dane particle as well as the 20-nm spherical particle and the tubular form found in the serum of patients with acute hepatitis B is composed of the hepatitis B surface antigen (HBsAg). The internal core of the Dane particle contains the hepatitis B core antigen (HBcAg). This antigen can also be found in the nuclei of infected hepatocytes (1, 5).

In most previous studies on antibody to hepatitis B core antigen (anti-HBc), the amount of antibody present was not quantified. Results obtained by complement fixation tests suggest that, in patients convalescing from acute type B hepatitis, anti-HBc was shorter lived than the antibody to hepatitis B surface antigen (anti-HBs) (5, 6). In studies of blood donor populations and hemophiliacs, a much lower incidence of anti-HBc has been detected by complement fixation compared with that of anti-HBc, measured by passive hemagglutination or radioimmunoassay (5, 6). On the other hand, in two previous studies dealing with a "normal" population and a group of hemophilia patients, the present author found a very good correlation between the incidence of anti-HBc and anti-HBs (3; B. G. Hansson, Scand. J. Infect. Dis., in press). Anti-HBc was determined by immunoelectroosmophoresis (IEOP), and anti-HBs was determined by passive hemagglutination and radioimmunoassay. It was, therefore, considered to be of interest to study the levels of the anti-HBc titer, measured by IEOP, in a long-term follow-up of hepatitis B infections and compare the antibody titers of chronic HBsAg carriers and those of hemophiliacs with "normal" individuals with anti-HBc.

MATERIALS AND METHODS

Hepatitis patients. From each of 20 patients with hepatitis B, one serum from the acute, HBsAg-positive stage of the disease and one serum drawn between 5 and 6 years later were tested for anti-HBc. Eight of these patients were followed up with frequently drawn blood samples, especially during the first year after the onset of illness.

Chronic HBcAg carriers. Sera from 10 HBcAg-positive blood donors and 10 dialysis patients who were carriers of HBcAg were tested for anti-HBc. One of the dialysis patients had been under continuous treatment with immunosuppressive drugs before she was first found positive for HBcAg. None of the remaining nine dialysis patients was under immunosuppressive medication at the time when the serum sample was collected.

Hemophilia patients. Changes in the anti-HBc titer over a period of time were investigated in 20 hemophiliacs by testing two sera drawn from each patient 4 years apart. One of these patients had had clinical hepatitis 13 years before the first serum sample was taken. The remaining 19 patients had no history of overt hepatitis.

"Normal" individuals with anti-HBc and anti-HBs. The amount of anti-HBc in sera from 20 persons with anti-HBc and anti-HBs selected from a Swedish "normal" population described in a previous work (3) was quantified.

Demonstration of anti-HBc. The IEOP technique previously described for HBsAg and anti-HBs detection (4) was used to test for anti-HBc by using HBcAg purified from a human liver (3). The same batch of antigen was used throughout the study. The amount of anti-HBc in serum specimens was measured by...
testing two-step dilutions of the individual sera by IEOP.

RESULTS

The ranges and geometric mean anti-HBc titers of the different groups studied are shown in Table 1.

Hepatitis patients. In 20 cases of hepatitis B the reciprocal of the mean anti-HBc titer was 630 in the acute phase. After 5 to 6 years, all patients were positive for anti-HBc, the reciprocal mean titer being 9.4. The mean antibody titer of the eight patients, who were tested frequently, showed a continuous decrease over the period studied. The decline of antibody titer during the first year was 3.5 dilution steps, and during the second year it was 1 dilution step; between the second and fifth years after the disease, the antibody titer declined 0.5 dilution step annually (Fig. 1).

Chronic HB,Ag carriers. All ten HB,Ag carriers found among blood donors had anti-HBc, the reciprocal mean antibody titer being 270. The dialysis patient who was under treatment with immunosuppressive drugs was negative for anti-HBc. The reciprocal mean anti-HBc titer of the remaining nine dialysis patients was 1,200.

Hemophilia patients. The reciprocal mean anti-HBc titer of the initial sera in hemophilia patients was 10. In sera drawn 4 years later from the same patients, the reciprocal antibody titer was 6.5.

"Normal" individuals with anti-HBc and anti-HB, . Anti-HBc titers ranging between 1/1 and 1/8 were detected among the 20 persons with anti-HBc and anti-HB, selected from a "normal" Swedish population. The reciprocal mean anti-HBc titer was 3.0.

DISCUSSION

Since it had been observed that in many patients with acute type B hepatitis anti-HBc could be detected for only a short period of time after the acute phase of the disease (5, 6, 10), it was suggested that anti-HBc would be a sensitive indicator of continued virus replication (7). However, in the present study anti-HBc could be detected by IEOP in all of the 20 patients studied when tested 5 to 6 years after onset of illness. The anti-HBc titers in the acute HB,Ag-positive phase of hepatitis were comparable to those of the chronic HB,Ag carriers, both the asymptomatic carriers detected in a blood-donor population and the HB,Ag-positive dialysis patients (P > 0.05). The HB,Ag-positive patient groups had 30 to 400 times higher anti-HBc titers than the others (P < 0.001), suggesting that continuing synthesis of HB,Ag was occurring in the HB,Ag-positive patients, including the asymptomatic HB,Ag carriers detected among blood donors, who, it has been suggested, present little risk of being contagious (8, 11). However, low titers of anti-HBc can be detected for many years after the hepatitis B virus infection, as shown in previous studies (5; Hansson, in press) and the present work. Theoretically, the anti-HBc antibodies in hemophilia patients could be a result either of infection by hepatitis B virus or of immunization by noninfectious HB,Ag contained in clotting-factor concentrates. Out of 20 children with hemophilia type A or B whose treatment with clot-
ting-factor concentrates was started in 1971 or later, three were positive for HB,Ag and anti-HB, and seven were positive for anti-HB, only, when tested after 1 to 6 years (Hansson, in press). However, although the clotting-factor concentrates thus were shown to give rise to anti-HB, synthesis, none of the 20 patients of the present study showed higher antibody titers in the second serum than in the first one. Since none of these patients showed boosting of anti-HB, one could conclude that the amount of HB,Ag, in virus particles or as free noninfectious antigen, which might be contained in the clotting-factor concentrates, is too small to cause antibody synthesis. Thus, the anti-HB, antibodies found in hemophiliacs are an indication of previous infection by hepatitis B virus rather than of immunization by noninfectious HB,Ag material.

During the 4-year study period, the low anti-HB, titers of the 20 hemophiliacs demonstrated a decline of somewhat less than one dilution step. The mean antibody titer of the first drawn sera corresponded to that of the 20 cases of clinical hepatitis 5 to 6 years after onset of illness, which might indicate that the hepatitis B infection of these hemophilia patients had occurred about 5 years before the first sera were drawn. This would agree well with the time when treatment with clotting-factor concentrates, which have been shown to transmit viral hepatitis in high frequency (2, 9), was first introduced into practice.

The anti-HB, titers in the "normal" material without any known history of hepatitis were low, about one-third of the antibody titers found in sera drawn 5 to 6 years after acute clinical hepatitis ($P < 0.001$). It could be assumed that the hepatitis B infection in most of these 20 "normal" individuals would have occurred more than 5 years ago. The anti-HB, findings of this study, together with the good correlation between the prevalence of anti-HB, and anti-HB, in different populations demonstrated in previous studies (3; Hansson, in press), suggest that, following an acute infection with hepatitis B virus, both anti-HB, and anti-HB, persist to about the same extent.

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LITERATURE CITED