PREVALENCE OF ANTIBODIES AGAINST HEPATITIS B ANTIGEN (HB Ag) IN GENERAL POPULATION

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Abstract

The objective of the present study was to estimate the exposure rate to Australia (Hepatitis-B) antigen among the general population, consisting of 250 contacts of hepatitis patients and 250 Naval recruits. The parameter included

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detection of antibodies against the antigen and the frequency of antibodies measured by the complement fixation in a micro-system. Among the contacts, three (1.2%) were found to carry antibodies to hepatitis B antigen and among recruits, eight (3.2%) were antibody positive. This rate most probably reflects only a fraction of the actual prevalence. The low rate of detection is due in part to the anticomplementary activity accruing in most of the sera.

The frequency of antibodies in low age group suggests that hepatitis-B is a disease of childhood. However, the actual predilection regarding age and sex could not be determined because of the high anticomplementarity of the sera.

On the basis of these observations it is suggested that in order to assess the actual exposure rate to the antigen of hepatitis B among general population, experiments should be performed on fresh sera, obtained from a comparatively larger number of cases. (JPMA 29:113, 1979)

Introduction

The connection between hepatitis B antigen and acute viral hepatitis is well established. The antigen is also associated with chronic active hepatitis, persistent viral hepatitis and other forms of hepatitis (Blumberg and Melartin, 1970; Elling et al., 1970). Some of the individuals who are repeatedly exposed to the antigen, may develop antibodies (Goke et al., 1970; Szmuness, 1975). Hepatitis B antibody has been isolated from the blood of the haemophilics (Blumberg and Alter, 1965) and other persons receiving multiple transfusions (Okochi and Murakami, 1968) as a result of repeated exposure to the antigen present in donor's blood (Schmidt and Lennette, 1970).

The frequency of anti-HB Ag is also directly related to the age and inversly related to socio-economic status of the individual (Cherubim et al., 1972).

Hepatitis B antigen can be transmitted by transfusion of blood (Kattamis et al., 1974) injection needles, (Alkan and Fanaru, 1973) mosquitoes (Tin et al., 1973), through placenta (Schweitzer, 1975), oralfecal route (Krugman et al., 1967), and sexual contact (Heathcote et al., 1974), leading either to hepatitis, antigenaemia or antibody formation in the recipient.

As in our country patients suffering from various contageous diseases including hepatitis are not treated in a proper way and strict preventive measures are never implemented in a true sense, chances of exposure of healthy individuals to the infective agent are more common. The present study has, therefore, been planned to carry out investigations to detect the number of individuals exposed to the infective agent among the general population, and this was achieved by detecting antibodies against the antigen by the complement fixation test in a Microsystem.

Material and Methods

The study was conducted on sera obtained from 250 apparently healthy Naval recruits and 250 house-hold contacts of hepatitis patient. Data of all the individuals, regarding their age, sex, place of residence, income, family history of hepatitis and their relation to the hepatitis patients, was obtained from the Research Centre, Pakistan Medical Research Council, Jinnah Post-graduate Medical Centre, Karachi. Sera were tested to detect the presence of antibody to hepatitis-B antigen (Anti-HB Ag) by the complement fixation test.

Sera used in this study were obtained from the Research Centre, Pakistan Medical Research Council, Jinnah Postgraduate Medical Centre, Karachi. None of these individuals had evidence of liver disease, however, 13 of the contacts and 7 of the recruits were positive for HBs antigen (Zuberi 1976).

The serum samples from the contacts were collected between 1.1.1976 and 16.12.76 and those from the recruits between 23.1.1974 and 23.3.1975. All the sera were stored at —20°C and had undergone thawing at different intervals before the commencement of the present study.

The complement fixation test was performed by the microtitre methods as described by Bradstreet and Taylor (1962). All the sera were heated undiluted at 56°C for 30 minutes to destroy the endogenous complement. Sera were tested at dilutions of 1:2-1:256. Three units of complement were used. The serum antigen/diluent and complement were mixed and incubated overnight at 4°C; the sensitized sheep cells were then added. The test was

performed with acrylic microtitre plates and 0.025 ml droppers (Khadim 1977).

Results

In the present study 387 (77.4%) of the serum samples had anticomplementary activity. The frequency of anti-HB Ag in serum samples from the contacts and the recruits has been shown in Table I. Sera from the 3 of the 250 contacts (1.2%) and 8 of the 250 recruits (3.2%) were found to harbour antibody to hepatitis-B antigen. Among the contacts one was below the age of 20 years, whereas, two were over 20 years of age. Generally the incidence of antibody was more among males and was common in younger age group (Table II). The titres of the antibody ranged between 1:2 to 1:8 (Table III) Sera from the contacts had (CF antibody titres at 1:2, whereas those of the recruits had a CF titre of 1:2 to 1:8.

None of the individuals with detectable HB Ag developed antibodies. Even in those individuals who developed anticomplementary activity, free anti-HB Ag could not be detected after anticomplementary activity disappeared (Table IV).

Table I: Frequency of HB-antibody in Contacts and Recruits

Age (years)	*Contacts*		*Recruits*	
	No. serum sample tested	*No. (%) Positive for Anti-HB Ag*	*No. serum samples tested*	*No. (%) positive for Anti-HB Ag*
1 — 10	56	1 (1.8)	—	—
11 — 20	41	—	250	8 (3.2)
21 — 30	82	2 (2.4)	—	—
31 — 40	37	—	—	—
41 — 50	21	—	—	—
51 — 60	7	—	—	—
60 onwards	6	—	—	—
Total	250	3 (1.2)	250	8 (3.2)

Table II: Distribution of the Antibody-positive Subjects According to Age and Sex.

Age (years)	*Males*		*Females*		*Total*	
	Number	*Percentage*	*Number*	*Percentage*	*Number*	*Percentage*
1 — 10	—	—	1	9.1	1	9.1
11 — 20	8	72.7	—	—	8	72.7
21 — 30	—	—	2	18.2	2	18.2
Total:	8	72.7	3	27.3	11	100.0

Table III: Distribution of CF Activity of HB Ab in Sera of Contacts and Recruits

Reciprocal of CF titres	*Number of sera with indicated titre*	
	Contacts	*Recruits*
2	3	4
4	—	3
8	—	1
Total	3	8

Discussion

The frequency of anti-HB Ag as measured by complement fixation test has been reported for patients who have received transfusions but not for the general population. Shulman and Barker (1969) found antibody to HB Ag by complement fixation (FC) in 5 per cent in patients who had been given transfusions.

Table IV: Distribution of Anticomplementary Activity

Group	*Serum samples tested*	*Sera showing AC activity*	*Sera without AC activity*	
			Antibody positive	*Antibody negative*
Antigen positive	20	14	—	6
Antigen negative	480	373	11	96
Total:	500	387	11	102

The incidence of anti-HB Ag detected by CF in patients, such as hemophilics, who have received multiple transfusions has ranged from 0 to 34 per cent (Holland et al., 1969; Cossart, 1969; Hirschman et al., 1969; Mathews and Mackay, 1970). Lander and associates (1971) have reported a similar proportion (21.7%) of 23 patients given multiple transfusions who had anti-HB Ag detectable by complement fixation.

The present study shows that only 8 of 250 recruits (3.2%) were found to carry anti-HB Ag. This rate most probably reflects only a fraction of actual prevalence. The low rate of detection was due in part to the anti-complementary activity occurring in high proportion (77.4%) of sera. On the other hand studies carried out on the sera of donors and apparently healthy subjects from Karachi, revealed that of the 449 blood-donors tested by passive haemagglutination technique, 81 (18.04%) were anti-HB Ag positive; 50 apparently healthy subjects tested by radio-immunoassay and 17 tested by passive haemagglutination. 17 (34%) and 6 (35.3%) were shown to have anti-HB Ag respectively (Zuberi 1976).

Since naval recruits belong to a selected population of healthy subjects with negative history of previous hepatitis, the likelihood of parenteral exposure in this group is thought to be low and would seem to reflect non-parenteral, sub-clinical exposure to hepatitis-B antigen. This type of transmission has been shown to occur for HB Ag-positive hepatitis by Krugman and associates (1967).

Experiments performed on the sera of 250 contacts indicate that only 3 (1.2%) serum samples had anti-HB Ag. In an epidemiological report, Mitch and his colleagues (1974) have described the detection of antibodies to hepatitis-B antigen in 22 per cent of the contacts of hepatitis patients, measured by haemagglutination technique, whereas Irwin and his colleagues (1974) have reported 4 per cent of antibody positive contacts of asymptomatic HB-Ag carriers.

Regarding HB-Ab positivity in these contacts (mother and sisters) it has been assumed that they developed the antibodies in response to sub-clinical infection (in case of mother from her son and in case of sisters from their brothers). The prevalence of both HB-Ag and HB-Ab among family contacts of donor carriers and the tendency to clustering of individuals with HB-Ag and HB-Ab within families suggest that the virus associated with HB Ag can be transmitted by person to person contact (Szmuness et al., 1973). Moreover, most of the contacts in the present study came from a low socio-economic class. Since antibody is more prevalent in lower socio-economic group (Cherubin et al., 1972), these individuals must also have an increased risk of non-parenteral exposure because of poor sanitation, overcrowding and inadequate medical facilities, an association found in the case of viruses such as poliomyelitis (Paul et al., 1952).

The age and sex distribution of anti-HB Ag (82% of those under 20 years of age and 18% of those over 20 years of age and more in males, i.e., 73%) suggests that HB Ag positive hepatitis is primarily an infection of childhood and more common in males. This is in agreement with the age specific detection of HB-Ag among persons with hepatitis in the general population. HB Ag has been detected in many cases of lower age group (less than 20 years) hepatitis (Blumberg et al., 1972), and mostly in males (Banke et al., 1971).

Failure to detect antibodies in persons with known antigenaemia is perplexing and suggests that HB Ag is not as sufficiently potent antigen to stimulate the production of antibodies detectable by the complement fixation technique after primary exposure (Purcell et al., 1971). It has also been suggested that inability to demonstrate antibody might be attributable to the fact that the small quantities of antibody produced are bound to antigen (Shulman and Barker, 1969). This concept has been strengthened by the electron microscopic studies of Almeida and Waterson (1969), who observed complexes of hepatitis-B, antigen and antibody in the sera of a patient with chronic hepatitis and one with fatal hepatitis. In the later case the immune complexes appeared to contain a low proportion of antigen and an excess of antibody, but no mention was made of the demonstration of free antibody by complement fixation.

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