DETECTION OF ANTI-HBC AND ANTI-HBS ANTIBODIES IN BLOOD DONORS NEGATIVE FOR HEPATITIS B VIRUS SURFACE ANTIGEN

Saad S.Hamadi1[1], Hayder A. Al-Hmudi[2] & Maha M. Hameed[3]

ABSTRACT

The Objectives: **safety of blood products is one of the major issues in the area of transfusion medicine. In previous study we found that hepatitis B virus was epidemic in some of Basrah medical institutions to assess the prevalence of anti-HBc and anti-HBs antibodies in serum samples of healthy blood donors negative for both HBsAg and anti-HCV in the Central Blood Bank, Basrah-Iraq.**

Results: **204 serum samples negative for both HBsAg and anti-HCV collected from healthy blood donors were tested for the presence of HBsAg, anti-HBs, HBeAg, anti-HBe and anti-HBc by using rapid chromatographic immunoassay. Of the 204 samples tested, 13 (6.37%) of the HBsAg negative blood samples were found to be positive for anti-HBc. 3(1.47%) out of 13 anti-HBc positive were found positive for anti-HBs.**

Conclusion: **detection of anti-HBc and anti-HBs antibodies were more important than other markers in detection of latent Hepatitis B virus infection.**

INTRODUCTION

Hepatitis B (HBV) is an indolent disease that seldom causes symptoms until complications of cirrhosis and liver cancer occur.[1] It is estimated that 40% of the world's population have had contact with or are carriers of the HBV. This corresponds to an estimated 350 million HBV carriers and around one million persons die of HBV-related causes annually.[2] The genes of HBV comprise genetic codes that create numerous protein products including Hepatitis B surface Antigen (HBsAg), Hepatitis B core Antigen (HBcAg),Hepatitis B e (soluble) Antigen (HBeAg) and DNA polymerase.[3] Blood is the most important vehicle for HBV transmission.[4] Though sensitive screening assays for detection of HBsAg are available, occasional cases of post-transfusion HBV infection still occur.[5] The safety of blood products is one of the major issues in the area of transfusion medicine.[6] Anti-HBc antibodies are markers of acute, chronic, or resolved HBV infection and remain detectable for life. These can be present in the absence of both HBsAg and anti-HBs antibodies, during the convalescent period following acute hepatitis B before the appearance of anti-HBs antibodies, or in patients who have resolved infections but lost detectable anti-HBs antibodies. Anti-HBc antibodies are, therefore, detected in anyone who has been infected with HBV.[7] In previous study[8] we found that the hepatitis B virus was really society-threatening and epidemic in some of the Basra medical institutions particularly in dialysis and oncology units. Furthermore, at present, HBsAg detection is the only diagnostic screening test for HBV infection in blood transfusion centers in Iraq. Unfortunately, the absence of HBsAg in the blood of apparently healthy individuals may not be sufficient to ensure the lack of circulating HBV. So, this study was undertaken to assess the rate of prevalence anti-HBc and anti-HBs antibodies in serum samples of healthy blood donors negative for both HBsAg and anti-HCV in the Central Blood Bank, Basra-Iraq.

MATERIALS AND METHODS

The study included 204 blood donors (200 males and 4 females with age ranging from 18 to 61 years). The negative serums samples for both were HBsAg and anti-HCV were collected from healthy blood donors in the Central Blood Bank, Basrah-Iraq. All serological tests, HBsAg, anti-HBs, HBeAg,

[1]Department of Medicine, College of Medicine, University of Basrah, Iraq

[2]College of Science, University of Basrah,

[3]Basrah Central Blood Bank

anti-HBe and anti-HBc, were studied using rapid chromatographic immunoassay (one step Hepatitis B virus Combo Test Device) according to manufacture instruction (ACON Laboratories, Inc. USA)

RESULTS AND DISCUSSION

As shown in Table (1,2),the present study showed the vast majority of blood donors were non-immunized persons (92.16%) with significantly elevated (P<0.001). Also 13 out of 204(6.37%) of the HBsA negative blood samples were found to be positive for anti-HBc. Overall, 3(1.47%) out of 13 anti-HBc positive were found positive for anti-HBs. This may explain the large numbers of infected patients with HBV in the wards of dialysis and oncology after a period of transfusion. So we divided our blood donor population into4groups with the following percentages: Non-immunized persons (92.16%), post infection (4.9%), recovered persons (1.47%) and immunized persons (1.47%).

The present study showed that mean age of the blood donors was 32±8 years 98% of them were males. This can be attributed to the fact that people with these age ranges are the most active individuals in the society and the more likely to be blood donors.[8] Furthermore, the age ranges of the past infection and recovered persons groups varied between (20-29) and (30-39) years. Also, the age period (<19) and (≥50) years was the least in blood donors with significantly decreased (P<0.001). Unfortunately, exact data is difficult to generate as many cases will remain undetected due to the asymptomatic nature of many acute and chronic infections.

Table 1. Distribution of HBV markers among blood donors.

Studied groups	No.	%	Markers				
			HBsAg	*Anti-HBs*	*HBeAg*	*Anti-HBe*	*Anti-HBc*
Non-Immunized Persons	188	92.16	-	-	-	-	-
Past Infection	10	4.9	-	-	-	-	+
Recovered Persons	3	1.47	-	+	-	-	+
Immunized Persons	3	1.47	-	+	-	-	-
P < 0.05							

Table 2. Distribution of age ranges with gender among studied groups.

Age range	Total	*Non-immunized persons*		*Past infection*		*Recovered persons*		*Immunized persons*	
		Male	*Female*	*Male*	*Female*	*Male*	*Female*	*Male*	*Female*
< 19	3	3	0	0	0	0	0	0	0
20-29	45	42	0	0	0	1	0	2	0
30-39	138	125	4	6	0	2	0	1	0
40-49	15	11	0	4	0	0	0	0	0
≥ 50	3	3	0	0	0	0	0	0	0
P < 0.05									

HBsAg is the first serologic marker to appear and can be detected 1-2 weeks or as late as 11-12 weeks following exposure. It is clear within six months of onset in 90% of people who acquire Hepatitis B.HBsAg positivity persists beyond 6 months in 10% of infected individuals and is indicative of chronic hepatitis B.[9,10] Thimme, *et al* [11] showed that after inoculation, HBV does not immediately start to replicate efficiently. HBV-DNA and HBV antigens are not detectable in serum or in the liver until 4-7 weeks post-infection. [12] All viral markers were important in the diagnosis of the disease and in giving a clear indication of the cases tested. So using HBsAg marker only for detection of the virus in the medical institutions is not enough to detect all HBV infections.[8] In approximately 50% of patients with self-limited hepatitis B virus infection, there is a time interval of up to several months between the disappearance of detectable HBsAg and the appearance of anti-HBs. During this time, only the anti-HBc is detectable; this period is referred to as the **"core window"** or "window phase".[9] The anti-HBc positive subjects consisted of 6.37% of blood donor population. They can be further divided into two groups, with (1.47%) and without anti-HBs (4.9%) individuals, respectively. These individuals may have recovered from previous infection but have persistent low level of HBV. In addition the absence of anti-HBs can be due to its undetectable levels. Very few patients with chronic HBV infection become HBsAg negative in the natural course of infection. The annual rate of HBsAg clearance has been estimated to be less than 2% in western patients and even lower (0.1-0.8%) in patients of Asian origin.[13] However, clearance of HBsAg does not exclude development of cirrhosis or liver cancer in some patients. This phenomenon is thought to be linked to the fact that HBV DNA may still be detectable by PCR assay both in serum and liver.[14] The clearance of the covalently closed circular form of HBV DNA (cccDNA) has been difficult, and clearance of HBsAg rarely occurs after 1 year of treatment.[15] Also 3(1.47%) of the individuals were found positive only for anti-HBs and were considered as immunized persons. Because HBV vaccine is so effective, the Centers for Disease Control and Prevention recommends initiation of the booster vaccination if completion cannot be guaranteed. Booster vaccination is recommended only for immuno compromised persons who do not respond after the initial series.[4] Also there was a significant decline in the levels of antibody over time after vaccination, and the losses of protective levels of antibodies were quite evident by 3-4 years after the primary doses.[16] Furthermore, HBeAg and anti-HBe antibody were not determined in all samples. HBeAg appears few days after HBsAg becomes detectable and typically disappears before HBsAg disappears, although it might persist for years in a chronic carrier of HBV.[10] While anti-HBe is not a protective antibody, its appearance usually coincides with a significant immune change associated with lower HBVDNA replication.[17] The clinical and laboratory features and the outcome at 6 months of HBV acute hepatitis in Vietnam are similar in HBeAg positive and negative patients.[18]

In conclusion, this study shows the insufficient effectiveness of HBsAg screening in protecting blood recipients from HBV infection. Inclusion of anti-HBc testing should be considered in the screening of blood donors.

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