ORIGINAL ARTICLE

SIGNIFICANCE OF TESTING ANTI-HBCIGM ANTIBODIES FOR THE SCREENING OF HEPATITIS B IN THE DONOR BLOOD

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Background: The purpose to perform this study was to screen blood donors for possible occult HBV by checking the seroprevalence of the hepatitis B antibodies in blood donors. It was a Cross-sectional study conducted at Blood Bank of Lahore General Hospital Lahore from April to June 2015 (3-months). **Methods**: In this prospective study, 180 healthy blood donors, presenting to the blood bank of Lahore General Hospital were selected. Their detailed demographic data and blood samples were collected. HBsAg testing was done by ELISA and further HBc IgM testing was also done by ELISA. Those testing positive for HBc IgM were further evaluated by real-time PCR to detect HBV DNA. **Results:** Mean duration of the life span was 26.51 years with a range of 18–61 years. Sex distribution show 93.9% (n=169) males and 6.1% (n=11) females. HBsAg was positive in 3.3% (n=6) while their HBc IgM was negative and HBc IGM was positive in 2.2% (n=4) of the healthy donors in whom HBsAg was found negative by ICT method. further qualitative HBV DNA by rt-PCR was done on those positive with anti HBc IgM and no patient had HBV DNA detected from their blood. **Conclusion:** Without routine screening of the sera for the HBc Antibody, the low-level HBV viraemia may not be detected as the nonappearance of the surface antigen in the blood of apparently healthy donors do not ensure the absence of circulating virus in the blood of these donors.

Keywords: Blood Donors; Hepatitis B virus; Screening

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INTRODUCTION

In current practice, the only screening test used in developing countries like Pakistan to evaluate the presence of Hepatitis B virus (HBV) infection in stored blood samples is HBsAg. But, it did not grantee that HBV will not transmit to the recipient, as during the "core window period", HBsAg can't be recognized in the blood, although hepatitis B disease is there.[1] In this "window period", antibody detection against hepatitis B core antigen (anti-HBc) is useful especially for countries like Pakistan which have a moderate to high frequency of hepatitis B endemicity.[2] It was observed, that it is a good indicator of the "core window" period, thereby helping to effectively remove, the blood units from inventory obtained from people contaminated with HBV in whom HBsAg is absent on testing. Similarly, individuals with acute hepatitis B who are in the window time frame following the loss of HBsAg and before the presence of anti-HBs. For this reason, some blood banks still advocate anti-HBc testing for the screening of their donor.[3]

However, due to availability of more developed strategies for Nucleic Acid Testing (NAT) like TMA (Transcription Mediated Amplification) or PCR (Polymerase Chain Reaction), as these can increase chances of detection of HBV DNA, the role of antiHBc testing is limiting. However Prior anti-HBc testing can result in the drawing of boundary lines between a large pool of donors and some effective and healthy donors.

The primary objective of the research was to assess the usefulness of anti-HBc as a screening marker of donors in a blood bank in a resource-constrained country, which falls in the middle of the zone for Hepatitis B endemicity

MATERIAL AND METHODS

The study was conducted in the Blood bank of Lahore General Hospital, Lahore, from April to June 2015 (3 months).Both male and female blood donors of 18 years to 60 years age group were included and blood donors who were known patients of Hepatitis B ,having history of close contact with hepatitis patients in last 6 month or having history of vaccination against Hepatitis B were excluded from study. We used simple random technique for sampling

The medical history and clinical examination were completed for every one of these individuals by qualified staff prepared to screen contributors for blood donation. Informed consent was taken before sampling of these volunteer blood donors. Sample for testing of infection marker was obtained during the collection of blood units in these donors. These blood samples were used to check HBs Antigen and HBc antibody by third-generation ELISA kits. Every positive specimen by ELISA for HBsAg as well HBc antibody were rechecked, utilizing a similar ELISA kit.

Only those Samples were considered positive which were positive on the repeated test. Data

was entered and analysed using SPSS 20. Quantitative parameters were represented by mean and standard deviation while frequency and percentages were used to present qualitative parameters.

RESULTS

In total, 180 patients were included in the study, among which 169 (93.9%) patients were male and 11 (6.1%) were female. The mean age and SD of patients enrolled in the study was 26.51±6.09 with a minimum age of 18 years and maximum age of 69 years.

Out of 180 patients, 174 (96.7%) had negative HBsAG while 6 (3.3%) patients were tested positive for HBsAG whereas 176 (97.8%) patients had negative HBclgM antibody and 4(2.2%) patients had positive HBclgM antibody. Interestingly all Anti HBc positive patients were male and had negative HBs Ag while all the patients who had detected HBs Ag in their serum were negative for Anti HBc IgM antibodies. (Tabl-1)

Table-1: Frequency and percentage of serum markers to detect occult Hep B

Contingency table among Gender *HBsAG *HBclGM	**HBsAG**	**Total**		
	Yes	No		
Positive Anti HBc IgM	Male	0 (0%)	4 (2.2%)	4 (2.2%)
Negative Anti HBc IgM	Male	5 (2.8%)	160 (88.9%)	165 (91.7%)
	Female	1 (0.5%)	10 (5.5%)	11 (6.1%)
Total	180 (100%)			

DISCUSSION

Due to a large number of patients suffering from Hepatitis B in Pakistan, chances of transferring this infection in individuals receiving blood products are fairly high.

Despite being tested for HBsAg, post-transfusion HBV infection is still occurring, because mostly HbsAg is at a very low or undetectable level by screening assays in these individuals.[4] Therefore, a need for a marker, that will detect hepatitis B during window period is essentially required.[4,5] Mosley et al. concluded in his study that AntiHBc screening of blood donators may inhibit HBV transmission from HBsAg-negative blood donors.[6]

Though sensitive, above mentioned serological tests have reduced the pre-seroconversion window period, they still cannot identify quite a few newly infected donors.[7] The baseline risk of blood donation in the window period has been bypassed by utilizing NAT assays which has extensively reduced the window period.[8–11]

After the disappearance of HBs Antigen in patients exposed to the Hepatitis B virus, AntiHBc alone may be seen in many such individuals due to persistence low level viremia or chronic infection[12]. These individuals can be HBV DNA positive and may transmit HBV infection.[13]

Anti HBc positivity rate in India varied from 8.4% as published by Dhawen *et al* to 18.3% in the study of Bhattacharya *et al*.[14,15] Similar high prevalence of Anti HBc in blood donors was observed at 15.9% in a study conducted at New Dehli.[16] Data presented by Chaudhuri *et al*, Anti HBc positivity rate was 10.82.[17] These figures were slightly higher than we observed in our population.

Amini *et al*.,[18] found 5.1% of patients positive for anti HBc, out of 4930 healthy blood donors with undetectable HBsAg, however, none of the positive anti HBc cases were HBV DNA positive. Behzad-Behbahani *et al*.,[5] conducted a study on 2000 healthy blood donors in Iran, confirmed 6.55% of healthy blood donors with HBsAg-negative samples to be positive for anti HBc antibody.

In our study blood donors who were positive for Anti HBc, were negative for HBV DNA on further testing. Similar finding were observed in a study from Poland that despite transfusion of HBV contaminated Red cell concentrate no evidence of infection transfer was found in recipients.[19]

But in few studies in early nineties reported transfer of HBV infection from Anti HBc positive blood donors.[20] Anti HBc screening in high endemic area would result in much smaller donor pool as discard rate of collected blood will be high.[21,22]

Many authors advocated the use of PCR in the early window period of HBV infection in blood donors[23], although it's not cost-effective for routine blood screening in developing countries like Pakistan.

We feel that in Pakistan where we have large number of individuals having HBV exposure, Anti HBc testing in blood donors will lead to a reduction of blood donor pools. The solution of this problem was found in a study done by El-Sherf *et al*[24], who suggested Anti HBc testing in blood units, which were HBs negative would identify potentially infectious units. Those AntiHBc positive blood units should further be tested by HBV DNA. This approach will not only be cost effective but also help in decreasing the rejection of blood donors positive with HBc antibody alone.

CONCLUSION

Adding anti HBc testing of blood donor screening does improve blood transfusion safety but will reduce the pool of blood donors, particularly in our country which is an intermediate endemic zone in the world. No doubt PCR testing is expensive but it is need of time to get the

blood sample screened by this due to its good screening accuracy. We recommend on basis of this study that blood donor screening should include anti HBc testing to detect occult Hepatitis B infection and positive donors should further evaluated by PCR testing to prevent the transmission of hepatitis B infection.

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AUTHORS' CONTRIBUTION

AM: Script writing, data collection, statistical analysis. SR: Data collection, statistical analysis. AJR: Data collection, statistical analysis. IUH: Script writing. GUNT: Supervision of study writing, data collection and statistical analysis

REFERENCES

1. Kumar H, Gupta PK, Jaiprakash M. The role of anti-HBcIgM in screening of blood donors. Med J Armed Forces Ind 2007;63(4):350–2.
2. Makroo RN, Raina V, Chowdhry M, Pathuppalli M. Hepatitis B Virus Infection among blood donors of North India. Apollo Med 2007;4(4):339–42.
3. Makroo RN, Raina V, Goyal N, Kaushik V. Effectiveness of screening blood for anti-HBc and anti-HCV on post transfusion hepatitis on multiply transfused patients. Indian J Hematol Blood Transfus 2001;19(1):49–50.
4. Hoofnagle JH, Seefe LB, Bales ZB, Zimmerman HJ. Type B hepatitis after transfusion with blood containing antibody to hepatitis B core antigen. N Engl J Med 1978;298(25):1379–83.
5. Behzad-Behbahani A, Mafi-Nejad A, Tabei SZ, Lankarani KB, Rashidi M, Rasouli M, *et al*. Indication of anti-HBc antibody screening and HBV- DNA detection in diagnosing latent hepatitis B virus infection. Iran J Med Sci 2005;30(1):28–33.
6. Mosley JW, Stevens CE, Aach RD, Hollinger FB, Mimms LT, Solomon LR, *et al*. Donor screening for antibody to hepatitis B core antigen and hepatitis B virus infection in transfusion recipients. Transfusion 1995;35(1):5–12.
7. Makroo RN, Choudhury N, Jagannathan L, Malhotra MP, Raina V, Chaudhary RK, *et al*. Multicenter evaluation of individual donor nucleic acid testing (NAT) for simultaneous detection of Human Immunodeficiency Virus -1 and Hepatitis B and C viruses in Indian blood donors. Indian J Med Res 2008;127(2):140–7.
8. Romano L, Velati C, Baruffi L, Fomiatti L, Colucci G, Zanetti AR, *et al*. Multicenter evaluation of a semiautomated, standardized assay for detection of hepatitis B virus DNA in blood donations. J Clin Microbiol 2005;43(6):2991–3.
9. Dodd RY, Notari IV EP, Stramer SL. Current prevalence and incidence of infectious disease markers and estimated window period risk in the American Red Cross blood donor population. Transfusion. 2002;42(8):975–9.
10. Laperche S. Blood safety and nucleic acid testing in Europe. Euro Surveill 2005;10(2):516.
11. Lelie N, Heaton A. Hepatitis B: A review of the role of NAT in enhancing blood safety. J Clin Virol 2006;36(Suppl 1):S1–2.
12. Makroo RN. Practice of safe blood transfusion: Compendium of Transfusion Medicine. 2nd ed. New Delhi, India: Kong Posh Publications Pvt; 2009.
13. Li A, Yuan Q, Huang Z, Fan J, Guo R, Lou B, *et al*. Novel double-antigen sandwich immunoassay for human hepatitis B core antibody. Clin Vaccine Immunol 2010;17(3):464–9.
14. Dhawan HK, Marwaha N, Sharma RR, Chawla Y, Thakral B, Saluja K, *et al*. Anti-HBc screening in Indian blood donors: Still an unresolved issue. World J Gastroenterol 2008;14(34):5327–30.
15. Bhattacharya P, Chandra PK, Datta S, Banerjee A, ChakrabortyS, Rajendran K, *et al*. Significant increase in HBV, HCV, HIV and syphilis infections among blood donors in West Bengal, Eastern India 2004-2005: Exploratory screening reveals high frequency of occult HBV infection. World J Gastroenterol 2007;13(27):3730–3.
16. Makroo RN, Raina V, Kaushik V. Prevalence of hepatitis markers (HBsAg, anti-HBc and anti HCV) in Indian blood donors. Vox Sang 1998;74:1142.
17. Chaudhuri V, Nanu A, Panda SK, Chand P. Evaluation of serologic screening of blood donors in India reveals a lack of correlation between anti-HBc titer and PCR-amplified HBV DNA. Transfusion 2003;43(10):1442–8.
18. Amini S, Mahmoodi MF, Andalibi S, Solati AA. Seroepidemiolo gyof hepatitis B, delta and human immunodeficiency virus infections in Hamadan province, Iran: A population based study. J Trop Med Hyg. 1993;96(5):277–87.
19. Reesink HW, Engelfriet CP, Henn G, Mayr WR, Delage G, Bernier F, *et al*. Occult hepatitis B infection in blood donors. Vox Sang 2008;94(2):153–66.
20. Manzini P, Girotto M, Borsotti R, Giachino O, Guaschino R, Lan teri M, *et al*. Italian blood donors with anti-HBc and occult hepatitis B virus infection. Haematologica 2007;92(12):1664–70.
21. Alexander J, Kowdley V. Epidemiology of hepatitis B-clinical implications. Med Gen Med. 2006;8(2):13–23.
22. Hollinger FB, Sood G. Occult hepatitis B virus infection: A covert operation. J Viral Hepat. 2010;17(1):1–15.
23. Hennig H, Puchta I, Luhm J, Schlenke P, Georg S, Kirchner H. Frequency and load of hepatitis B virus DNA in first-time blood donors with antibodies to hepatitis B core antigen. Blood 2002;100(7):2637–41.
24. El-Sherif AM, Abou-Shady MA, Al-Hiatmy MA, Al-Bahrawy AM, Motawea EA. Screening for hepatitis B virus infection in Egyptian blood donors negative for hepatitis B surface antigen. Hepatol Int 2007;1(4):469–70.

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