ORIGINAL RESEARCH PAPER

Microbiology

A COMPARISION STUDY OF DETECTION OF HEPATITIS B SURFACE ANTIGEN AND ANTIBODY TO HEPATITIS C VIRUS AT A TERTIARY CARE CENTRE IN BHOPAL

KEY WORDS: Hepatitis B virus (HBV), Hepatitis C virus (HCV), HCV TRI-DOT, Radioimmunoassay.

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ABSTRACT

Background: Hepatitis B virus (HBV) and Hepatitis C virus (HCV) are serious health problem All over the world and major causes of chronic hepatitis, liver cirrhosis and hepatocellular carcinoma. The aim of this study was to estimate the prevalence rates of HBV and HCV infections in Bhopal region.

Methods: Serum samples of inpatients and outpatients of J. K. Hospital, Bhopal (MP) were collected over a period of one and a half year. HBsAg was determined using the HBsAg One Step Hepatitis B Surface Antigen Test Device. Antibody detection of HCV was done using HCV TRI-DOT.

Results: A total of 4298 serum samples were tested for HBsAg detection and 767 serum samples were tested for Hepatitis C virus antibodies. Seropositivity for HBsAg was 2.60% whereas HCV seropositivity was 4.17 %. A higher seroprevalence of HBsAg and HCV was found in males as compared with females.

Conclusions: Attempts should be made to reduce the incidence of HCV and HBV and their unregulated spread which can be done by increasing public awareness of simple preventive measures.

Introduction

Hepatitis B Virus (HBV) and Hepatitis C Virus (HCV) are diseases characterized by a high global prevalence with complex clinical course, and limited effectiveness of currently available antiviral therapy. Hepatitis B virus (HBV) and Hepatitis C virus (HCV) are serious public health problem worldwide and major causes of chronic hepatitis, liver cirrhosis and hepatocellular carcinoma.[1] WHO estimates that approximately 2 billion people have serological evidence of past or present HBV infection. More than 350 million are chronic carriers of HBV.[2] The HBV prevalence rate is 2 to 8 % with an approximate carrier pool of 40 million. The HBV infection rate varies in different regions of the same country as well HBV prevalence is more in South India than West India.[3,4]

It has been estimated by WHO that the global prevalence of Hepatitis C virus (HCV) infection is around 2%, with 170 million persons chronically infected with the virus and 3 to 4 million persons newly infected each year.[5]

Clinically, HBV infection is indistinguishable from other viral hepatitis. Accordingly, its diagnosis relies on a specific laboratory tests for distinguishing it from such viruses.[6,7] Several viral markers are available for the detection of HBV and Hepatitis B surface antigen (HBsAg) is the major viral marker used for the detection of HBV infection.[8] HBsAg is the first marker to appear and becomes detectable during acute HBV infection. Several immunological methods such as enzyme immunoassays (EIA), radioimmunoassays (RIA), immunochromatographic assays (ICA) and haemagglutination assays are available to detect HBsAg. Among these methods, EIAs and RIAs are the most sensitive methods.[9,10] EIA methods are generally used by reference laboratories and blood banks because of its accuracy, low cost and safety in comparison with RIA methods.

For detection of HBsAg, rapid diagnostic tests based on immunochromatographic principles are widely used in most developing countries, including India. These methods are considerably cheaper than EIA methods and can generate results within 30 minutes, therefore being less time consuming when compared to EIA. Also in comparison with EIA, expert training is not required to perform an ICA.

A significant percentage of individuals suffering from HCV infection are asymptomatic and are detected only on random check-ups for various purposes. The presence of hepatitis C antibody in the serum or plasma is an indication of HCV infection, although this does not indicate whether the infection is acute, chronic or resolved.[11]

The purpose of this study was to estimate the prevalence rates of HBV and HCV infections in this part of country. To assess the magnitude of transmission of a disease in a community and for its control and prevention, the assessment and study of its prevalence is very important. This study was therefore conducted since sufficient data of prevalence rates in this part of country is not available.

Material and Methods

This study was carried out at the Microbiology Department of L.N. Medical College & J. K. Hospital, Bhopal (MP) from September 2013 to February 2015 after getting approval from the Institutional Ethical Committee. Inpatients and outpatients who were advised hepatitis B screening and HCV antibody testing on the basis of clinical findings, risk factors, socioeconomic, demographic, and on few occasions as part of preoperative evaluation and antenatal screening of HBsAg status were included in the study. Exclusion criteria included the subjects who had prior history of HBV immunization. 5 ml of blood sample was collected using standard collection procedure[12] and was transported to the microbiology laboratory for testing. In case of delay, serum sample was separated & stored in refrigerator at 2-8°C till further testing. The blood was allowed to clot & after centrifugation, serum samples were separated in clean test tubes and were subjected to the requested tests. All the test were performed with accordance to the manufacturer's instructions with adequate controls.

HBsAg was determined using the HBsAg One Step Hepatitis B Surface Antigen Test Device (Serum/Plasma) [ACON, Rapid Diagnostics Pvt. Ltd, India]. This Hepatitis B Surface Antigen test is a qualitative, solid phase, two-site sandwich immunoassay for the detection of HBsAg in serum or plasma. The kit has sensitivity of 99.8% and specificity of 99.2%.

Antibody detection of HCV was done using HCV TRI-DOT (Diagnostic Enterprises, Parwanoo, HP), a rapid, visual, sensitive and qualitative method for detection of antibodies to Hepatitis C virus in human serum/plasma. This is a fourth generation HCV TRI-DOT test designed with increased sensitivity for core and NS3, NS4, NS5 antibodies using a unique combination of modified HCV antigens. The kit has sensitivity of 100% and specificity of 100%.

Results

A total of 4298 serum samples were tested for HBsAg detection and 767 serum samples were tested for Hepatitis C virus antibodies over a period of one and a half years. Seropositivity for HBsAg was 2.60% whereas HCV seropositivity was 4.17%. Table 1 and 2 shows age and sex wise distribution of patients with Hepatitis B and Hepatitis C seroprevalence respectively. The seroprevalence of HBsAg among males and females was 3.14% and 2.34% respectively. The seroprevalence of HCV among males and females was 3.65% and 4.30% respectively. Among males highest prevalence of HBsAg was found in age group of 31-40

years. Age group of 41-50 years showed the highest prevalence of HBsAg among females. The highest prevalence of HCV among males and females was found to be in the age group of 41-50 years.

Table 1 : Age-wise and sex-wise distribution of HBsAg in patients (n=4298)

Age group (yrs)	Males	Females	HBsAg positive Males (%)	HBsAg positive Females (%)	Total HBsAg positive cases (%)
0 – 10	50	30	00 (0)	00 (0)	00 (0)
11 – 20	38	05	04(10.52)	1 (6.60)	05(11.62)
21 – 30	610	1000	10 (3.27)	16 (1.60)	26 (1.61)
31 – 40	800	300	30 (3.75)	10 (3.33)	40 (3.63)
41- 50	600	150	20 (3.35)	10 (6.66)	30 (4.00)
51 - 60	300	50	06 (2.10)	1 (2.00)	07 (2.00)
61 - 70	190	85	02 (1.05)	1 (1.17)	03 (1.09)
Above 71	50	30	01 (2.00)	00 (0)	01 (1.25)
Total	2638	1660	83 (3.14)	39 (2.34)	112(2.60)

Table 2: Age-wise and sex-wise distribution of anti HCV Ab in patients (n=767)

Age group (yrs)	Males	Females	Anti HCV Ab positive Males (%)	Anti HCV Ab positive Females (%)	Total anti HCV Ab positive cases (%)
0 – 10	06	02	00 (0)	00 (0)	00 (0)
11 – 20	13	08	00 (0)	00 (0)	00 (0)
21 – 30	60	42	00 (0)	00 (0)	00 (0)
31 – 40	120	96	04 (3.33)	04 (4.16)	08 (3.70)
41- 50	166	65	10 (6.02)	06 (9.23)	16 (6.92)
51 - 60	40	43	01 (2.5)	02 (4.65)	05 (6.02)
61 - 70	30	30	01 (3.33)	01 (3.33)	02 (3.33)
Above 71	30	16	01 (3.33)	00 (0)	01 (2.17)
Total	465	302	17 (3.65)	13 (4.30)	32 (4.17)

Discussion

Hepatitis B virus and Hepatitis C virus are serious public health problem worldwide & major cause of Chronic Hepatitis, Liver Cirrhosis & Hepatocellular Carcinoma. In our study, the seropositivity rate of HBsAg was 2.60%. In India, Lodha et al[13] has reported a true prevalence of 1-2%. The prevalence of HBsAg in different regions of our country varies widely, and the highest prevalence has been reported from the aborigine population of Andaman as well as from Arunachal Pradesh.[14] The prevalence of hepatitis B varies from country to country and depends upon a complex mix of behavioral, environmental, and host factors. Sood et al[15] has noted 0.87% prevalence in a study of HBsAg prevalence in hospital based population. Batham et al[16] in their review of 54 studies on HBsAg prevalence in India have reported that prevalence in non tribal population is 2.4%, whereas a very high prevalence was observed among tribal population (15.9%). Srikrishna et al[17] have reported 1.86% prevalence among blood donors of Bangalore.

In our study, a higher prevalence of HBsAg is seen in males (3.14%) compared with females (2.34%). Most of the studies have reported higher prevalence among males which is also true in our study. Sood et al[15] has reported the prevalence of 1.04% and 0.58% respectively for males and females. Dutta et al has reported prevalence of 35.3% in males and 19.3% in females.[18] Singh et al has noticed prevalence to be 0.65% and 0.25 % respectively in males and female subjects.[19] It is hypothesised that females probably clear the HBV more efficiently in comparison to males.[20] Seropositivity was found higher in 3rd, 5th and 6th decade of life. Similar findings were found by Sood et al.[15]

Many developing countries widely use ICA based rapid diagnostic tests to detect HBsAg for both diagnosis and screening of acute and chronic HBV infections, although ideally, more advanced and accurate methods such as EIA or ELISA should be used for screening. Differences in the prevalence of HBV infection in a given population can occur because different ICA based rapid assays used for HBsAg detection in the serum may not have the same accuracy index in every region. Also, in a region, the circulating subtype/s of HBV can also be different. In such cases ICA that does not cover this particular subtype/s will not detect this type when testing.[21] There are no approved rapid assays by the food and drug administration (FDA) and CE mark for European Union for HBsAg detection although several rapid tests for screening for HIV have been approved.[22] Rapid assays must be used with caution. ICAs need regular validation with an accepted EIA for the detection of HBsAg if rapid ICAs are used for diagnostic and screening purposes in resource poor settings.

In our study, the seroprevalence of HCV was found to be 4.17%. Sood et al[15] has also reported a low prevalence of 0.28%. Other studies from Cuttack (Orrisa)[23] and Pondicherry[24] have reported a higher prevalence of 1.57% and 4.8% respectively. In this study, three cases which were seropositive were between 41-50 years age group. Sood et al[15] has also reported that the four reactive HCV cases belonged to the age groups 41-50 years and >60 years. A study on the seroprevalence of hepatitis C in urban areas of Madagascar by Ramarokoto et al[25] suggested that the prevalence did not differ significantly according to gender but it increased with age.

Conclusion

Prevalence of HBV and HCV in our study was 2.60% and 4.17%, respectively. Attempts should be made to reduce the incidence of HCV and HBV and their unregulated spread which can be done by increasing public awareness of simple preventive measures. HBV infection is vaccine preventable, complications due to chronic HBV infection such as cirrhosis, liver cancer can be totally prevented by compulsory immunization programmes and good awareness about the availability of effective vaccine. An effective childhood immunization programme will reduce the burden of HBV infection in this country. All high risk groups should be compulsory vaccinated, blood samples should be effectively screened before transfusion. Due to the high cost of treatment of HCV infection and the unavailability of a vaccine against HCV, efforts should be focused on preventive aspects like regular screening.

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