

Evaluation ELISA and Rapid test for the Detection of HBsAg among chronic liver disease (CLD) patients in Bihar, India.

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Abstract

Background: Hepatitis B is a most important health trouble in India. Hepatitis B virus (HBV) is deliberation to be the principal etiological agent for chronic liver disease (CLD) worldwide. This study aims to conclude the incidence of HBV by transmission of chronic liver disease (CLD) patients. This study also aims to anticipated the sensitivity between two diagnostic tests; one step rapid test strip device (RIA) and Enzyme Linked Immunosorbent Assay (ELISA).

Materials and Methods: It is hospital based cross-sectional learning was permitted out in adult patients with liver disease attending the Hepatology OPD, tertiary care hospital in Koshi Region Bihar India. Age, gender and clinical history of the patient were recorded. Blood specimen was Evaluation for Hepatitis B surface Antigen (HBsAg) using one step rapid test strip device and Enzyme Linked Immunosorbent Assay (ELISA).

Results: Three hundred patients were enrolled in the study. The suggest age of infected patients in the learning group was 38.2 ± 1.22 years (range 20 to 72 years). Of the 300 samples tested, 18.6% were positive for HBsAg by **HEPA-SCAN HBsAg ELISA Test** and 13.2% were positive by **Hepacard** one step rapid test. Considering the results of **HEPA-SCAN HBsAg ELISA** the sensitivity, specificity, PPV and NPV of ELISA was 96.55%, 99.18%, 96.55% and 99.18% respectively. The sensitivity, specificity, PPV and NPV of Hepacard one step rapid test was 95.34%, 99.23 %, 96.55% and 99.23% respectively.

Conclusion: The occurrence of HBV is high in India and the occurrence is superior in males than the females. We also well-known that in comparing both methods (ELISA method and the rapid test strip) for assessing the presence of HBsAg. ELISA test technique was found to be extra sensitive than the rapid test strip device. Therefore, we advocate sturdily, that

ELISA method be used to validate test results obtained from the one step rapid test, when monitor for chronic liver disease (CLD).

Keywords: ELISA methods, chronic liver disease, Hepatitis B, Rapid test strip

Introduction

Liver disease recognized to hepatitis B virus (HBV) has turned into a marvelous issue broadly [1]. As per current appraisals, around 2 billion people worldwide are infected with HBV; among whom approximately 400 million with chronic disease [2]. India is at the intermediate endemic level of hepatitis B, with hepatitis B surface antigen (HBsAg) prevalence between 2% and 10% surrounded by the populations measured.[3] Of the 2 billion people who have been infected with the hepatitis B virus (HBV) in the world, extra than 360 million have chronic (lifelong) infections. In India, of the 25 million infants born every year, over one million run the lifetime risk of budding chronic HBV infection. Estimates designate that per annum over 100,000 Indians die due to illnesses related to HBV infection [3] Hepatitis B Virus (HBV) infection is a worldwide public health problem. It is predictable that around 360 million people are infected worldwide with the virus.^[4] Viral hepatitis is a universal disease primarily concerning the liver. Most of the cases of acute viral hepatitis are caused by Hepatitis A (HAV), Hepatitis B Virus (HBV) or Hepatitis C Viruses (HCV). HBV has a double standard DNA encoding for P, X, core and surface proteins. The multifarious antigen found on the surface of HBV is called Hepatitis B surface antigen (HBsAg). Antibodies adjacent to HBV proteins are other immunological markers of infection, of which Anti-Hepatitis B core antigen, Hepatitis B envelope antigen and Hepatitis B envelope antibody are also recognized abruptly after HBSAg, and are

significant markers of past or present HBV infection. In a typical Hepatitis B infection, Hepatitis B surface antigen (HBSAg) will be detected contained by 2 to 5 weeks before symptoms or jaundice develop.^[5]

Presently serological screening tests are working for the analysis amongst all HBSAg assays, ELISA techniques are the the majority numerous used because of their efficiency. In many urbanized countries, HBSAg screening is frequently ready with ELISA techniques.^[6] Blood transfusion services are a vital part of contemporary health care system, with every unit of blood there is 1% probability of transfusion connected problem include transfusion communicable diseases. Transfusing infected blood to unsuspected patients in requires is a crime. It is mandatory to test each and every unit of donor blood for antibodies to HIV-1 and HIV-2, syphilis, HBSAg and HC Virus. ELISA is suggested and favorite screening technique for blood banking.^[7] While Enzyme-linked Immunosorbent Assay (ELISA) is an enzymatic immuno-assay technique of the “sandwich” type for the detection of Hepatitis B virus in human serum or plasma. The test uses monoclonal antibodies preferred for their ability to bind themselves to the various sub-types of HBSAg now recognized by the World Health Organization (WHO) and the most part of modification HBV strains.^[8, 9] The present study was hence approved out to find out the prevalence of HBsAg in liver disease patients and to compare the regularly Central Pathology Section used HBsAg recognition kit using (**Hepacard** one step rapid test) with the detection ELISA for in-vitro qualitative decision of HBsAg test kit in human serum (**HEPA-SCAN HBsAg ELISA Test**) was used.

Materials and Methods

Study Design: It is sanatorium based cross-sectional learning was carried out in adult patients with liver

disease presence the Hematology OPD, tertiary care hospital in Koshi region (Bihar) India subsequent to obtaining institutional ethics committee consent over a period of Two years from March 2014 to March 2016. All the uninterrupted patients during the study period who were more than 18-years of age and gave on paper informed permission were included in the study. Age, gender and clinical history of the patient were recorded in the case record form after printed informed approval. This prospective study was conducted in the Department of Microbiology, Lord Buddha Koshi Medical College, Saharsa and Associated Hospital of Bihar India from March 2014 to March 2016. The 300 serum/plasma samples were screened using **Hepacard** one step rapid test for the detection of hepatitis B surface antigen (HBsAg).

Detection of HBsAg using ELISA method: - The 300 subjects were also screened using ELISA technique for hepatitis B. The ELISA test is a solid-phase micro titer plate coated with monoclonal antibodies to human IgM which is based on sandwich belief.

Statistical Analysis

The statistical examination in this study. The X2 (Chi-square) test and analyzed using the statistical software (SPSS version 18) was performed for quantitative variables to check for relationship in detecting HBV infection. Percentages were calculated directly for HBV infection. P = 0.05 was used as the accepted significance level.

Results

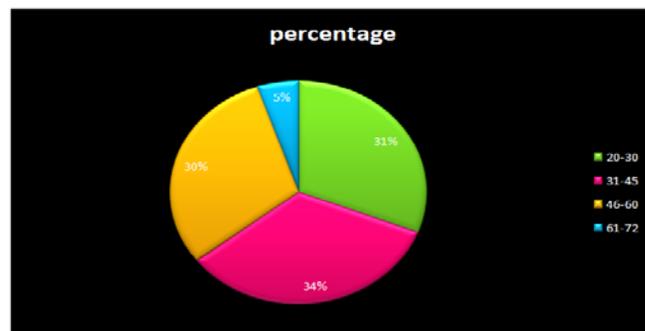
Three hundred patients were enrolled in the study. The mean age of infected patients in the study group was 38.2± 1.22 years (range 20 to 72 years). Two hundred five (68.28%) were males and Ninety five (31.72%) were females. The positivity amongst the male population was 19.14% which was higher than the

female population (17.28%) but the difference was statistically significant (p = 0.05) [Table-1].

Most of the CLD patients of HBsAg ELISA results were in the age group of 20-30 years of age 30.90% followed by 33.60 % in 31-45 years, 30.30 % in 46-60 years and 5.20% in 61-75.(Table-1) (Graph No.1) Sero prevalence of HBV among 300 CLD patients in present study was 18.8 %. HBV infection was seen mostly in age group 31-45 yrs (33.60%). (CHART TABLE No.2) Of the 300 samples tested, 18.8% were positive for HBsAg by **HEPA-SCAN HBsAg ELISA Test** [Fig-1] and 13.6% were positive by **Hepacard** one step rapid test [Fig/Table-2] [Chart Table-2] Considering the results of **HEPA-SCAN HBsAg ELISA Test** the sensitivity, specificity, PPV and NPV of ELISA was 96.55%, 99.18%, 96.55% and 99.18% respectively. The sensitivity, specificity, PPV and NPV of Hepacard one step rapid test was 95.34%, 99.23 %, 96.55% and 99.23% respectively.

Age	Number	Positive (%)
20-30	44	17 (30.90)
31-45	128	18 (33.60)
46-60	108	17 (30.30)
61-72	20	3 (5.20)
Male	205	39 (19.14)
Female	95	16 (17.28)

Table 1: Age and Gender wise distribution of patients of HBsAg results



Graph 1: Percentage of age groups

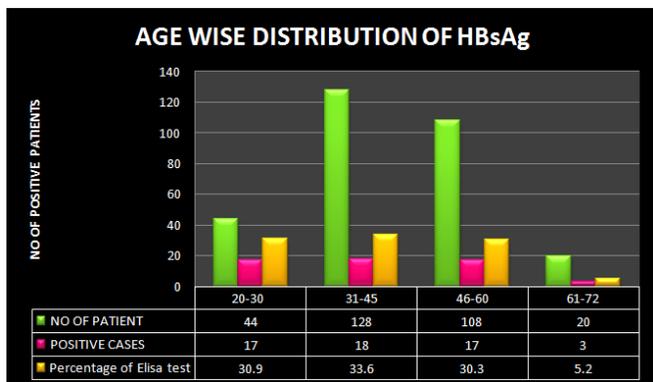


Chart Fig. 1: Age Wise Distribution of Hbsag Elisa Test Positive.

Test (n=300)	Positive	Negative
ELISA HBsAg	56	244 (18.8)
RAPID HEPACARD	41	259 (13.6)

Table 2: Comparison of ELISA results with Rapid test.

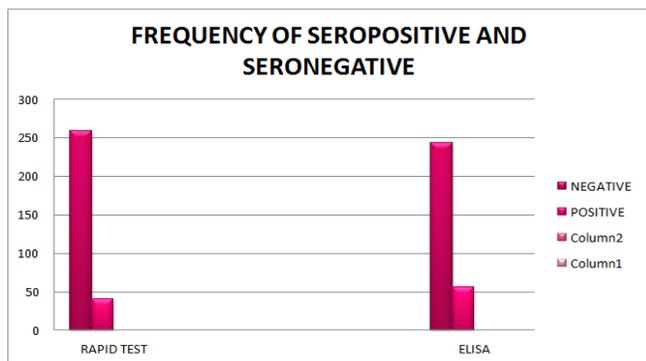


Chart Fig. 2: Incidence of Seropositivity and seronegativity according to the diverse methods use for HBsAg detection

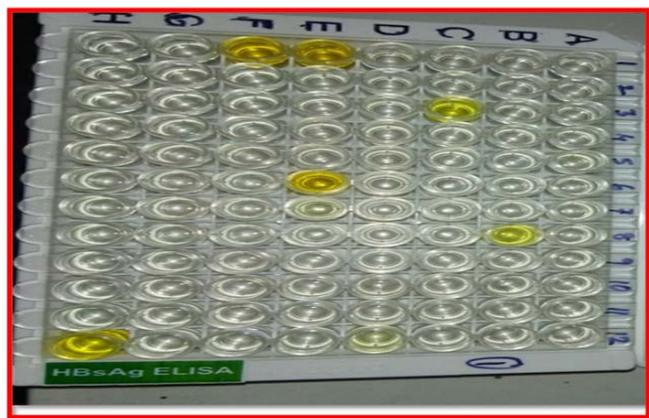


Fig.1 : HEPA-SCAN HBsAg ELISA Test



Fig.2: Hepacard one step rapid test.

Discussion

It is predictable by the World Health Organisation that there are about 350 million chronic carriers of hepatitis B allocation in all continent in Asia, America, Europe and Africa^[10,11] In this learning, two methods were second-hand (rapid strip test device and ELISA test) to check for sensitivity in communication for hepatitis B surface antigen among chronic liver disease patients. A quantity of the subjects (96.55%) experienced positive using ELISA method while 95.34% tested positive by means of the rapid test strip

Viral hepatitis is the most prevalent cause of chronic liver disease all during the world^[12, 13]. In India, HBV is reported to be answerable for 70% of chronic hepatitis cases and 80% of cirrhosis of liver cases^[14]. The mean age of the infected subjects in the near study was 43 years which is in concordance by means of a study accessible by Arora and Mann^[15]. Nayak et al., in their study have suggested that 30% of chronic carriers get infected vertically and remaining get infection directly from those who got it vertically^[16]. The probability of horizontal infection happening through close contact with carriers, use of unsafe injections, and an association with a number of socio-cultural practices increases with advancing age. Hepatitis B vaccination became available in 1981 and has been included in the

Universal Immunization program in India as late as 2007- 2008. This has contributed significantly in falling the occurrence there after ^[17]. However in the present study the incidence of Hepatitis B infection gradually decreased with age from 33.60 % in 30-45 years age group to 30.30% in patients older than 45 years of age. This possibly could be due to the other causes of chronic liver disease in older age group [Table/Fig-1]. HBsAg prevalence in men was higher (19.14%) as compared to women (17.28%) in the present study. A similar higher prevalence in men has also been reported by Ahmad I, Mishra A, Poddar CK, et al. ^[3]. Though no specific reasons can be attributed to the higher prevalence in men, it could possibly be due to a higher exposure risk in this population or because more male population seeks health care ^[3, 18]. In our study, 18.8% of patients with liver disease had hepatitis B infection which is similar to the study by Kumar et al., (17.34%) ^[19]. Other studies in India have reported HBsAg detection rate varying between 12.2-51% in liver disease patients ^[14, 15, 19, 20, 21]. The difference in prevalence rate in a given region is dependent on the degree of endemicity in that region. The higher prevalence in the present study as compared to that in the general population is expected since the patients enrolled were liver disease patients attending the Hepatology OPD with majority having symptoms like jaundice, anorexia, nausea, vomiting, and haematemesis.

Where a high frequency rate has been seen in ELISA method when compared to rapid strip test device. So, there is a great and urgent need for people in this locality and beyond to screen their blood for HBV and it should be screened using the ELISA technique which has been found to be more sensitive and accurate as compared to the rapid test strip device method.

Conclusion

This study shows that frequency of HBV is high in India and the incidence is greater in males than the females. We also noted that in comparing both methods (ELISA method and the rapid test strip) for assessing the presence of HBsAg, ELISA test method was found to be more sensitive than the rapid test strip device. Therefore, we recommend strongly, that ELISA method be used to confirm test results obtained from the one step rapid test, when screen for chronic liver disease (CLD).

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