

The Prevalence of Hepatitis B Surface Antigen (HBsAg) Among Blood Donors in Some Hospitals of Thamar, Yemen

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ABSTRACT

Viral hepatitis is a general term meaning inflammation of the liver and can be caused by a variety of viruses such as hepatitis B virus. Hepatitis B is a serious transfusion-transmissible disease affecting millions of people throughout the world.

This study was designed to evaluate the prevalence of HBsAg and the risk factors for HBV infection among blood donors in Thamar governorate/Yemen, and to focus upon the importance of blood transfusion in spreading the hepatitis B virus.

A total of 200 blood donors at the blood bank of Thamar hospital and Al-Wehda teaching hospital, during the period of January 2009 to October 2009, and January 2010 to July 2010, were screened for the presence of hepatitis B surface antigen (HBsAg). Each patient filled a questionnaire including age, sex, dental visit, blood transfusion ..etc. The donors were all male adult (age: 17-45 years).

Four out of 200 (2%) donors were found to be HBsAg positive by an enzyme-linked immunosorbent assay (ELISA) using monoclonal antibodies to HBsAg. Two of the four (50%) HBsAg-positive donors have relative symptoms of HBV (i.e jaundice) while the other two HBsAg-positive donors were asymptomatic. Questionnaires revealed that: The four positive donors were all married. The two asymptomatic HBsAg-positive donors were army soldiers. The other two symptomatic donors were civil workers.

One of the two asymptomatic donors has donated a blood a year ago and found to be HBsAg negative.

One of the symptomatic positive donors has had two previous surgical operations and received several pints of blood.

This study emphasizes the need for an accurate detection of this virus, and the need for using confirmatory tests for HBsAg detection and also the need to undertake serious measures to control the spread of HBV among Yemeni people.



INTRODUCTION

Viral hepatitis is a general term meaning inflammation of the liver and can be caused by a variety of different viruses such as hepatitis A, B, C, D and E. Since the development of jaundice is a characteristic feature of liver disease, a correct diagnosis can only be made by testing patient's sera for the presence of specific anti-viral antigens or antibodies^(1, 2, 3).

Hepatitis B is a serious and common infectious disease of the liver, affecting millions of people throughout the world.^(2, 3, 4, 5, 6)

The severe pathological consequences of persistent HBV infections include the development of chronic hepatic insufficiency, cirrhosis, and hepatocellular carcinoma (HCC). In addition, HBV carriers can transmit the disease for many years^(2, 3, 5). Infection occurs very often in early childhood when it is asymptomatic and often leads to the chronic carrier state⁽⁷⁾. More than 2000 million people alive today have been infected with HBV at some time in their lives. Of these, about 350 million remain infected chronically and become carriers of the virus^(2, 3, 5, 6). Three quarters of the world's population live in areas where there are high levels of infection. Every year there are over 4 million acute clinical cases of HBV, and about 25% of carriers, 1 million people a year, die from chronic active hepatitis, cirrhosis or primary liver cancer⁽⁸⁾.

Following HBV infection, HBV DNA is the first detectable marker in acute HBV infection (i.e. before HBsAg)⁽⁹⁾, then HBsAg, which is evident 2 to 8 weeks before the appearance of jaundice and biochemical evidence of liver damage. Next to appear are the markers of the virion, such as the soluble antigen, HBeAg⁽¹⁰⁾. In acute infections, clearance of the virus is marked by the disappearance of HBeAg and the appearance of anti-HBe. Later during convalescence, HBsAg also disappears with the appearance of anti-HBs⁽¹¹⁾.

Levels of several hepatitis markers found in the blood can confirm hepatitis B infection and differentiate acute from chronic infection. The diagnosis of acute hepatitis B is based upon the presence of the hepatitis B surface antigen (HBsAg) and hepatitis B core IgM antibody (HBcIgM). The diagnosis of chronic hepatitis B is based on the presence of the HBsAg marker for at least six months; HBcIgM antibody is usually negative.

Based on the prevalence of HBV chronic carriers (individuals positive for hepatitis B surface antigen, HBsAg) amongst adults in the general population, countries are classified as having low endemicity (< 2%), intermediate endemicity (2%–5%), or high endemicity (> 5%) of infection⁽¹²⁾. In most areas of the world (East and South Europe, South America, the Middle East, Middle Asia, Japan, and Turkey), HBV infection is of intermediate endemicity with HBsAg carriage rate of 2% to 7%^(7, 13, 14, 15, 16). Studies in the Middle East show the prevalence of HBsAg to range from 3% to 11% in Egypt, 4% to 5% in Iraq, 2.6% to 10% in Jordan, 2% to 6% in the Libyan Arab Jamahiriya, 2.3% to 10% in Oman, 5% to 6% in Palestine, 7.4% to 17% in Saudi Arabia, 16% to 20% in Sudan, 6.5% in Tunisia, 2% to 5% in UAE and 12.7% to 18.5% in the Republic of Yemen⁽¹⁷⁾. These surveys have been conducted on a variety of different population groups with the intention of giving a representative estimate of the prevalence of carriers in the general population. The endemicity of infection is considered high in Yemen where prevalence of positive HBsAg ranges from 8% to 20%, and up to 50% of the population generally have serological evidence of previous HBV infection^(18, 19, 9).

The hepatitis B virus can be transmitted in many ways. The main route of HBV transmission is via blood and blood products. Needle-stick injuries in health personnel, haemodialysis, shared needles in drug abusers, dental surgery, receiving blood or blood products, ear and nose piercing practices, tattooing, sexual contact and contact with body

fluid or mucosa of HBV carriers (e.g., workers in clinical laboratories) have been associated with increased risk of transmission⁽⁷⁾.

Aims

The aim of this study is to:

- 1- evaluate the prevalence of HBsAg and risk factors for HBV infection among blood donors in Thamar hospital and Al-Wehda teaching hospital /Yemen
- 2- Focus upon the importance of blood transfusion in spreading the hepatitis virus and the importance of accurate detection of this virus.

MATERIAL & METHODS

Material

The blood of the donors were collected in white tubes (without anticoagulant). Sera collected by centrifugation and subjected to this study. Blood collected by venipuncture, allowed to clot naturally and completely. Care was taken to ensure that the serum samples are clear. Any visible particulate matters in the sample were removed by centrifugation at 3000 RPM for 20 minutes at room temperature or by filtration. Highly lipaemic or haemolysed samples were excluded as they can give false results in the assay. Blood donors were also asked to answer questionnaires, which were used to obtain informations about risk factors associated with HBV infection.

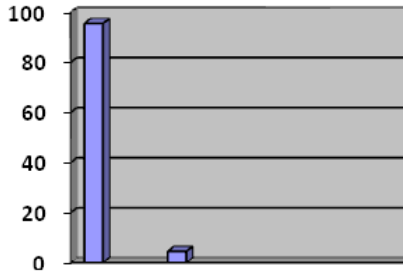
Methods

The blood samples of 200 donors were subjected to ELISA technique (sandwich method) for the detection of HBsAg using PARAMAX LABO diagnostic kit (USA). Controls were included in each test run. With monoclonal antibodies specific to HBsAg. Patient's serum sample is added to the microwell together with a second antibody conjugated with horseradish peroxidase (HRP) and directed against a different epitope of HBsAg. During incubation, the specific immunocomplex formed in case of presence of HBsAg in the sample, is captured on the solid phase. After washing to remove sample serum proteins and unbound HRP-conjugate, Chromogen solutions containing tetramethylbenzidine (TMB) and urea peroxide are added to the wells. In presence of the antibody-antigen-antibody (HRP) "sandwich" immunocomplex, the colorless Chromogens are hydrolyzed by the bound HRP-conjugate to a blue colored product. The blue color turns yellow after stopping the reaction with sulfuric acid. The amount of color measured and is proportional to the amount of antigen in the sample. Wells containing samples negative for HBsAg remain colorless.

RESULTS

The donors were all male adult (age: 17-45 years). Of the 200 blood donors subjected to this study, four were found to be HBsAg positive i.e , 2% of the blood donors have HBsAg. The results are presented in the following figure (figure; 1).

Figure (1): HBsAg positive donors (95.5%) & HBsAg negative donors (4.5%)



All positive samples were within age group 26-35 years (Table; 1).

Table (1): No. of HBsAg positive blood donors according to age

Age	No. Examined	No. positive	%
17-25	50	0	0
26-35	110	4	4
36-45	40	0	0
Total	200	4	2

Two of the four (50%) HBsAg-positive donors have relative symptoms of HBV (i.e jaundice) while the other two HBsAg-positive donors have no symptoms (asymptomatic).

Questionnaires revealed that:

1-The four positive donors were all married (age 26-35 years).

2- The two asymptomatic HBsAg-positive donors were army soldiers. The other two symptomatic donors were civil workers.

3- One of the two asymptomatic donors has donated blood a year ago and found to be HBsAg negative .

4- One of the symptomatic positive donors has had two previous surgical operations and received several pints of blood.

The results are tabulated in the following table:

Table (2): Results of the questionnaires of the HBsAg positive donors

Patients	Age	Occupation	Symptoms	History
1	26	army soldier	asymptomatic	nothing significant
2	29	army soldier	asymptomatic	blood donation
3	32	civil worker	jaundice	2 surgical operations & received blood
4	35	civil worker	jaundice	nothing significant

DISCUSSION

While transfusion-transmissible diseases, including AIDS and viral hepatitis, continue to spread especially in developing countries, the issue of safeguarding the community's blood

supply is of paramount importance. Blood banks at Tamar hospital and Al-Wehda teaching hospital receive many donations daily, and thus their blood supply has the potential to affect the community.

Four out of 200 donors were found to be positive for HBsAg. Two of the four positive donors were asymptomatic.

The two asymptomatic donors are either in the early stage of the infection or may be chronic carriers. Their condition is unknown because no further investigations were made (e.g. anti-HBcIgM, HBeAg) as these investigations are not available.

One of the asymptomatic donors had donated blood a year ago at Taiz medical center and found to be HBsAg negative using chromatography method. This indicates that either this donor was not infected at that period or that the negative result of Taiz medical center was false as the screening method (chromatography) is not as sensitive as ELIZA. In addition, no confirmatory test used. Researchers in Turkey ⁽²⁰⁾ recommend that asymptomatic HBsAg positive blood donors should be followed-up for signs of hepatitis. They found, after long follow up of asymptomatic HBsAg positive blood donors, that 15% of HBsAg positive blood donors have chronic hepatitis.

Our data showed that the two asymptomatic donors were Army soldiers. Hepatitis B is an expanding health problem in the Army^(23, 24). Military personnel are one of many core populations at increased risk for acquiring sexually-transmitted diseases and HBV infection^(25, 26). Further studies are needed to clarify associations between Army soldiers and HBV infection.

The other two positive cases in this study were symptomatic donors (jaundice). One of them has had two previous surgical operations suggesting that he might have been infected during operation (using contaminated tools) or from blood transfusion during or after operation.

In this study 98% of blood donors were HBsAg negative. Transmission by blood components negative for HBsAg can still occur in the acute phase of infection during the seronegative window period, or during chronic stages of infection ("occult" HBV infection, OHB). OHB is defined as the presence of HBV DNA in blood or liver tissues in patients negative for HBsAg, with or without any HBV antibodies^(9, 27).

Acute HBV infection is self-limiting in approximately 97% of immunocompetent adults leading to recovery and immunity. However, in 3% of cases, individuals remain chronically infected (i.e. carrier state), usually testing HBsAg positive and anti-HBc positive. Some of these chronically infected individuals can, however, have very low and/or intermittently detectable levels of HBV DNA and, on testing, are found to be HBsAg negative^(9, 27). In a study by El-Sherif, et al ⁽²¹⁾, HBV DNA was detected in 3.3% of blood donors negative for HBsAg and HBsAb. Because of limitations in current blood screening practices, OHB is an overlooked source of HBV transmission. Liu et al, ⁽⁹⁾ indicated that nucleic acid amplification test (NAT) or new HBsAg tests of enhanced sensitivity would be effective in the screening of blood donors for OHB in highly endemic countries.

In this study 2% of blood donors at Tamar hospital and Al-Wehda teaching Hospital were found to be HBsAg positive. This incidence is lower than that reported in Egypt, for instance, as the frequency of HBsAg in Egyptian blood donors was found to be 4.3%⁽²¹⁾ and much lower than that reported in Saudi Arabia's blood donors (13.9%)⁽¹⁴⁾. We attributed the differences in the carrier rates to the use of different measurement techniques. Also the results of this study is much lower than the carrier rates of the middle east countries ⁽¹⁷⁾.

These surveys have been conducted on a variety of different population groups. Higher rates may be observed in community-based studies than in studies conducted amongst blood donors⁽¹⁷⁾. Blood donors may not be representative of the general population. In some settings, blood donors tend to be healthier than individuals in the general population and consequently are more willing and capable of donating blood. In Saudi Arabia, blood donors appeared to have a lower prevalence of HBsAg (13.9%) when compared to the general population (16.7%)⁽¹⁴⁾. On the other hand, in Yemen, Scott et al.⁽²²⁾ found blood donors to have a significantly higher prevalence of HBsAg (20.6%) than non-donor (12.1%) apparently healthy individuals. Therefore, one cannot generalize findings based on studies conducted on blood donors. The 2% of the HBsAg positive cases presented here underestimate the number of hepatitis B infections transmitted by transfusion as infection is often asymptomatic and not all acute infections are diagnosed. Nevertheless, these data show that transmission of hepatitis B by transfusion does occur and emphasizes the need for improving the detection of HBV in all blood banks to reduce the chance of getting hepatitis B from a blood transfusion. Also mandatory reporting of the disease allows state health care workers to track people who have been exposed to the virus. Giving the hepatitis B vaccine or a hepatitis B immune globulin (HBIG) shot to those who have not yet developed the disease may help prevent hepatitis B infection if it is given within 24 hours of exposure. In recent years, blood centers in Yemen have tried to improve the nation's blood safety. Although a progress has already been made, many difficulties remain. Other developing countries face the same challenges as Yemen.

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فايروس الكبد (ب) بين متبرعي الدم في بعض مستشفيات ذمار- اليمن

مها الالوسي، عبدالسلام المقداد و عرفات الجرفي

كلية الطب والعلوم الصحية - جامعه ذمار

ملخص

يعتبر التهاب الكبد الفايروسي من أخطر الأمراض المعدية التي تنتقل عن طريق الدم والذي يؤثر على ملايين الناس في جميع أنحاء العالم. ويسبب هذا الالتهاب أنواع مختلفة من الفايروسات ومنها فايروس ب. أجريت هذه الدراسة للتعرف على مدى انتشار فايروس الكبد ب بين متبرعي الدم في بعض مستشفيات محافظة ذمار وكذلك لمحاولة معرفه الأسباب التي أدت للإصابة بهذا المرض. تمت هذه الدراسة على 200 متبرع للدم تتراوح أعمارهم 17-45 سنة في بنك الدم لمستشفى ذمار العام ومستشفى الوحدة التعليمي في محافظه ذمار خلال الفترة من يناير 2009 إلى أكتوبر 2009 ومن يناير 2010 إلى يوليو 2010 ولقد تم الطلب من كل متبرع بعض المعلومات مثل العمر، الجنس، زيارات لأطباء الأسنان، عمليات جراحية، نقل دم ... الخ.

المتبرعين كانوا كلهم ذكور تتراوح أعمارهم بين 17 إلى 35 سنة. ولقد تم فحص المتبرعين لوجود فايروس الكبد ب بواسطة اختبار الامتصاصية المناعية المرتبطة بالانزيم (تقنيه الأليزا) ولقد وجد ان أربعة منهم (2%) ايجابيين لفايروس الكبد ب. اثنين من الحالات الموجبة ابدوا بعض الأعراض (مثل الصفار)، البقية بدون أعراض. كل المرضى المصابين كانوا متزوجين، اثنان منهم جنود(أحدهم تبرع بالدم قبل سنة وكان فحصه سالب. الاثنان الاخران الذي كان فحصهم موجب مدنيين، احدهم خضع لعمليتين جراحيين في السابق وأستلم عدة قناني من الدم. هذه الدراسة تؤكد الحاجة لاستخدام فحوصات أكثر دقة لكشف هذا الفايروس والحاجة الى فحوصات توكيدية للفايروس واتخاذ اجراءات مشدده للسيطرة على انتشار هذا الفايروس في أوساط المجتمع اليمني.