PREVALENCE OF HBSAG AND ANTI-HBC AMONG BLOOD DONORS AT THE NATIONAL BLOOD TRANSFUSION AND RESEARCH CENTER, SANA'A, YEMEN

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ABSTRACT
Background: Substantial percentage of HBV cases were reported among our population. Blood transfusion represent major rout transmission of Hepatitis B virus. The recent situation in Yemen increase the demand of blood and its products due to anemia, accidents, malnutrition, etc. Objective: The aim of the study was to estimate the current prevalence of HBsAg and total anti-HBc among blood donors in Sana’a, Yemen. Methods: A descriptive cross-sectional study was carried out at the National Blood Transfusion and Research Centre in Sana’a Yemen, during the period from October 2016 to January 2017. A total of 3606 blood donors were included in this study. Serum was separated from each blood specimens, and then they were analyzed for the presence of HBsAg, total Anti-HBc antibodies using an Enzyme Immunoassay technique according to manufacturer instructions. Results: The result of this study shown that, 4.1% and 14.0% donors were found to be reactive for HBsAg and anti-HBc respectively. The overall prevalence of HBV (HBsAg and/or anti-HBc positive) was 16.2%. According to the residency of the blood donors, a high percentage of HBsAg were observed among donors from Raymah 10.8% and Almahwet 10.3% governments followed by Alhudidah 7.0%. However, the lowest of HBsAg among blood donors coming from Capital city of Sana’a, Sana’a rural area and Taiz government was 3.9%. 3.6% and 3.5% respectively. In other hand, anti-HBc was found higher among donors from Raymah followed by Almahwet. Alhudidah and Albaidah governarates at 35.1%, 25.9%, 20.9%, and 20.0% respectively. Conclusion: This study concluded that our subjects still under the moderate to high-risk communities.

KEYWORDS: Yemen; Hepatitis; HBsAg; anti-HBc; EIA; Blood donors.

INTRODUCTION
Hepatitis B virus (HBV) represents one of the most common public health problem universal. About approximately 1.5 million deaths worldwide in each year due to the HBV infection, the majority of which are attributable to primary hepatocellular carcinoma (HCC) as a subsequent to HBV infection.1 The estimated HBV chronically infected individuals were about 360 million.2 They are at higher risk of death from HBV-related complications and over a few millions new acute clinical cases arise.3

There are approximately 100 million hepatitis B carriers in the South-East Asia with approximately 500 000 deaths annually because of the chronic consequences of hepatitis B, particularly cirrhosis and liver cancer.4,5 HBV is a one of occupational risk to the health care personnel whom are considered to be at significant risk for getting or spreading the virus.6 This occupational risk varies according to many factors such as the work place in the health care location and the exposure times to the virus.7

HBV transmission is commonly occur by blood and/or its products that play a main role in the increase of HBV infection around the world.8,9 The HBsAg sero-prevalence among voluntary or replacement blood donors is still a most universal and public health concern. Therefore, it is necessary to investigate all the blood donors for presence of HBsAg in their serum. Serological examinations are one of the principal methods to define the prevalence of HBsAg.
According to the WHO the prevalence rate of < 2%, 2 – 8% is defined as low and intermediate, and that > 8% as high prevalence rate of HBV.\textsuperscript{[11]}

There are many diagnostic techniques used for detection of HBV antigens of antibodies that varies from simple chromatographic device, which used as a screening test, to high sensitive and specific enzyme immunoassays (EIA) and molecular methods that are used for confirmation and research situations.\textsuperscript{[8]}

Numerous factors as time, method of subject selection, differences in geographical location, and occupational differences were responsible on the variation of HBV infection prevalence in Yemen varied according. Its ranges from less than 2 % to 18%, as shown by many documents. Yemen consequently comes below the intermediate to high endemicity category.\textsuperscript{[12–17]}

HBV vaccination program in Yemen has been started since more than 10 years ago for children without program regulator on blood transfusion such as screening tests for blood donation that may cause the problem ongoing.\textsuperscript{[18]}

Therefore, we aimed of this study to estimate the current prevalence of HBsAg and total anti-HBc among blood donors in Sana’a, Yemen. That will provide rationalization for HBV infection control measures in Yemen.

In Yemen the most blood donors were not subjected to total anti-HBc screening test before blood transfusion in many centers. Total anti-HBc screening test may be used as one a additional marker of HBV infection. However, a high prevalence of total anti-HBc antibody have been verified in among those centers. Positivity without define anti-HBc IgG and anti-HBc IgM distinctly leading to the rejection of a substantial number of blood units.

MATERIAL AND METHODS

Study design
This is a descriptive cross-sectional study.

Date, place and population of study
The study was carried out on 3606 blood donors who attend the National Blood Transfusion and Research Centre in Sana’a Yemen, from 1 October 2016 to 15 January 2017 for blood donation. Analysis of blood donor’s samples was achieved at the laboratory virology unit. Donor informed consent was obtained and privacy was assured.

Data collection
A structured self-administered questionnaire was used to collect data.\textsuperscript{[14]} A trained public health specialist filled the questionnaire through face-to-face interview. The questionnaire sought information about personal characteristics such as age, gender, residency and occupation.

Methods
All the individuals coming for blood donations were grouped occupationally into the following four division, students, provisional, handicraft, and military. Professional workers are individual who have educational qualification such as teachers, engineers, health workers, employee etc. Handicrafts are persons who have less educational qualification or have free business, such as carpenters, plumbers, drivers, waiters…etc.

All blood donors were examined for blood pressure, pulse, hemoglobin (Hb) content, etc., and other general health check up were done. Apparently, healthy persons of age 16 to 65 years with body weight above 45 kg would qualify for donations and those who satisfied the meet the requirements criteria for the donation were incorporated in the study. The study included both the voluntary blood donors and the replacement blood donors.

Five milliliters of blood was collected to a sterile test tube by standard aseptic technique from each subjects by trained laboratory technicians. After separation of serum, specimens were analyzed for the HBsAg, total Anti-HBc using EIA technique (Monolisa\textsuperscript{TM} HBsAg ultra, no. 72348; Monolisa\textsuperscript{TM} anti-HBc plus, no. 72316; Monolisa\textsuperscript{TM}; BioRad Diagnostics, 92430 Marnes-la-Coquette, France) according to manufacture instructions. Overall prevalence of HBV infection was determined by the presence of HBsAg and/or anti-HBc.

Statistical analysis
The prevalence of HBsAg and anti-HBc was determined as percentage of seropositive individuals from the total donor population under investigation. Descriptive statistics related to socio-demographic variable and other variables were determined by computer program. A \( p \) value less than 0.05 was calculated to be statistically significant. Chi-square test was applied to evaluate the statistical difference using Statistical Package for Social Sciences (SPSS) software package version 20. (SPSS Inc. Chicago, Illinois, USAT).

RESULT
Out of 3606 blood donors registered in the study, 3578 (99.2%) were males and 28 (0.8%) were females and with mean of age 29.95 years (range 45 years). Replacement donors were 2616 (72.6%) compared to 987 (27.4%) of voluntary donors as shown in table 1.
The results of our study showed that, the overall detected positivity for HBsAg and/or Anti-HBc were 584 (16.2%). The total positivity of HBsAg among all blood donors was detected in 147 (4.1%), compared to 79 (2.2%) of blood donors who had HBsAg alone and the difference between voluntary and replacement group blood donors was found to be statistically significant ($P < 0.017$) (Table 1). In addition the result of present study revealed that, 68 (1.9%) of blood donors had positive for both HBsAg and anti-HBc. In addition, it was found that, 505/3606 (14.0%) of blood donors were reactive for total Anti-HBc, of these 437 (12.1%) were reactive for Anti-HBc alone.

Table 1: Distribution of anti-HBsAg and anti-HBc in study population.

<table>
<thead>
<tr>
<th>Donor category</th>
<th>N (%)</th>
<th>Total Anti-HBc N (%)</th>
<th>Anti-HBc alone N (%)</th>
<th>Anti-HBc and HBsAg N (%)</th>
<th>HBsAg alone N (%)</th>
<th>Total HBsAg N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Voluntary</td>
<td>987 (27.4)</td>
<td>123 (22.7)</td>
<td>104 (10.5)</td>
<td>19 (1.9)</td>
<td>31 (3.1)</td>
<td>50 (5.1)</td>
</tr>
<tr>
<td>Replacement</td>
<td>2619 (72.6)</td>
<td>382 (14.6)</td>
<td>333 (12.7)</td>
<td>49 (1.9)</td>
<td>48 (1.8)</td>
<td>97 (3.7)</td>
</tr>
<tr>
<td>Total</td>
<td>3606 (100.0)</td>
<td>505 (14.0)</td>
<td>437 (12.1)</td>
<td>68 (1.9)</td>
<td>79 (2.2)</td>
<td>147 (4.1)</td>
</tr>
<tr>
<td>$P$</td>
<td>0.101</td>
<td>0.074</td>
<td>0.915</td>
<td>0.017*</td>
<td>0.065</td>
<td></td>
</tr>
</tbody>
</table>

$P$: Probability; * Statistically significant; N: Number.

Table 2: Distribution of HBsAg and Anti-HBc among blood donors in relation to gender, residency, occupational and age groups.

<table>
<thead>
<tr>
<th>Gender</th>
<th>Total 3606</th>
<th>HBsAg Positive 147 (4.1%)</th>
<th>$X^2$</th>
<th>$P$</th>
<th>Anti-HBc Positive 505 (14.0%)</th>
<th>$X^2$</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>3578</td>
<td>99.2</td>
<td>147</td>
<td>4.1</td>
<td>1.2</td>
<td>0.273</td>
<td>1.1</td>
</tr>
<tr>
<td>Female</td>
<td>28</td>
<td>0.8</td>
<td>0</td>
<td>0.0</td>
<td>0.0</td>
<td>0.000</td>
<td>0.000*</td>
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<tr>
<td>Residency governorate</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sana'a capital city</td>
<td>2319</td>
<td>64.3</td>
<td>90</td>
<td>3.9</td>
<td>15.3</td>
<td>0.196</td>
<td>34.0</td>
</tr>
<tr>
<td>Sana'a rural area</td>
<td>582</td>
<td>16.1</td>
<td>21</td>
<td>3.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Taiz</td>
<td>144</td>
<td>4.0</td>
<td>5</td>
<td>3.5</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ibb</td>
<td>171</td>
<td>4.7</td>
<td>7</td>
<td>4.1</td>
<td></td>
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<tr>
<td>Dahmar</td>
<td>119</td>
<td>3.3</td>
<td>5</td>
<td>4.2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amran</td>
<td>82</td>
<td>2.3</td>
<td>4</td>
<td>4.9</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Raymah</td>
<td>37</td>
<td>1.0</td>
<td>4</td>
<td>10.8</td>
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<tr>
<td>Almahwet</td>
<td>58</td>
<td>1.6</td>
<td>6</td>
<td>10.3</td>
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<td></td>
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<tr>
<td>Alhudidah</td>
<td>43</td>
<td>1.2</td>
<td>3</td>
<td>7.0</td>
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<td></td>
<td></td>
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<tr>
<td>Albaidah</td>
<td>30</td>
<td>0.8</td>
<td>1</td>
<td>3.3</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Other</td>
<td>21</td>
<td>0.6</td>
<td>1</td>
<td>4.8</td>
<td></td>
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<tr>
<td>Students</td>
<td>674</td>
<td>18.7</td>
<td>25</td>
<td>3.7</td>
<td>2.3</td>
<td>0.345</td>
<td>1.2</td>
</tr>
<tr>
<td>Professionals</td>
<td>951</td>
<td>26.4</td>
<td>31</td>
<td>3.3</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Handicrafts</td>
<td>1342</td>
<td>37.2</td>
<td>63</td>
<td>4.7</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Militaries</td>
<td>639</td>
<td>17.7</td>
<td>38</td>
<td>4.4</td>
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<td>Age groups</td>
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<td>16-25</td>
<td>1237</td>
<td>34.3</td>
<td>52</td>
<td>4.2</td>
<td>5.1</td>
<td>0.273</td>
<td>37.1</td>
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<tr>
<td>26-35</td>
<td>1601</td>
<td>44.4</td>
<td>72</td>
<td>4.5</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>36-45</td>
<td>631</td>
<td>17.5</td>
<td>21</td>
<td>1.1</td>
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<td></td>
</tr>
<tr>
<td>46-55</td>
<td>121</td>
<td>3.4</td>
<td>1</td>
<td>0.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;55</td>
<td>16</td>
<td>0.4</td>
<td>1</td>
<td>6.2</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* Statistically significant, $X^2$: Chi-square, $P$: probability. ($X^2 \geq 3.84, P < 0.05$: significant). No: number, %, percentage

**HBsAg**

The distribution of donors according to their residency were 64.3%, 16.1%, 4.0%, 4.7%, 3.3%, 2.3%, 1.0%, 1.6%, 1.2% and 0.8% from Capital city (Sana'a), Sana'a, Taiz, Ibb, Dahmar, Amran, Raymah, Almahwet, Alhudidah and Albaïdah respectively, while 0.3% from others governments as presented in Table 2.

The result of this study show that, the prevalence rate of HBsAg among blood donors from Capital city of Sana'a, Sana'a, Taiz, Ibb, Dahmar, Almahwet, Alhudidah, Albaïdah and Sana'a capital city of Sana'a, Sana'a, Taiz, Ibb, Dahmar, Almahwet, Alhudidah and Albaïdah respectively, while 0.3% from others governments as presented in Table 2.
3 (7.0%), while less prevalence among donors from Albaithad (1.3%) government, the difference was found to be statistically insignificant (P < 0.196).

According to occupational categories, 674 (18.7%), 951 (26.4%), 1342 (37.2%) and 639 (17.7%) were students, professional, handicrafts, and military respectively. The frequency of HBsAg among them was 25(3.7%), 31 (3.3%), 63 (4.7%) and 28 (4.4%) respectively, while the difference in the result among them was found to be statistically insignificant.

On other hand, the most represented blood donors occur between 26 to 35 old ages with a rate of 1601 (44.4%) followed by the age group 16-25, 36-45, 46-55 and more than 55 years old with a percentages of 1237 (34.3%), 631 (17.5%), 121 (3.4%) and 16 (0.4%) respectively. While, the rate of HBsAg in these groups was 72 (4.5%), 52 (4.2%), 21 (1.1%), 1 (0.8%) and 1 (0.2%) respectively.

Anti-HBc
The current study revealed that the frequency of total anti-HBc among donors in Sana'a capital city was 295 (3578 (12.7%), whereas, 86582 (14.8%), 19 / 144 (13.2%), 22 / 171 (12.8%), 21 / 119 (17.5%), and 12 / 82 (14.6%) of blood donors from Sana'a rural area, Taiz, Ibb, Dahmar, and Amran governorates respectively had anti-HBc and the difference was found statistically insignificant. Anti-HBc was found higher among donors from Raymah followed by Almahwet. Alhuwiedh and Albaithad governorates with an account (35.1 %), 15 (25.9%), 9 (20.9 %), and 6 (20.0 %) respectively. The difference was found statistically significant P < 0.001.

As seen in the table 2, it was calculated that, the frequency of total anti-HBc among blood donors students was 59/674 (8.8%) compared to 136 / 951 (14.3%), and 92/39 (14.4%) in Professional and Military respectively and the difference was found statistically significance P < 0.001. Where the total anti-HBc was found higher among handicrafts donors with an account 218 / 1342 (16.2%).

Furthermore, it was found that the difference in the total anti-HBc among the age groups was statistically significant P < 0.001. However, high [31 / 121 (25.6%)] anti-HBc value was found among 46-55 age group, as compared to 26-35, 36-45 and > 55 years old age groups, while anti-HBc among them was [231 / 1601 (14.4%)], [112 / 6311 (17.7%)] and [4 / 16 (25.0%)] respectively.

DISCUSSION
Our results revealed that, the overall HBsAg among blood donor was 4.1%. This result is in agreement with that reported (4.1%) by Michel et al. in Congo [19] and quite similar (4.5%) to that reported by Al-Waleedi et al., in study conducted in Aden city. [14] The prevalence of HBsAg in recent study was more than that reported by Alodini et al [13] (2.1%), Jadeja et al. (1.32%) [20] and Atallah et al., (0.6%) [21] in study conducted in Sana'a City, India and Baghdad respectively. Rodenas et al. [22] have reported higher result 7.67% of HBsAg. The differences in the prevalence rate may be due to the geographic variation among regions or due to a difference in the detection method used.

In other aspect the result of recent study showed that, 3.9% blood donors resident the Sana'a capital city had HBsAg, where the prevalence of HBsAg among Sana'a capital city donors was less than that (5.1%, 7.1%, 9.0%, 15.0%) have been previously reported among Yemeni blood donors [14–17].

These differences in the prevalence rates of HBsAg in our study as compared to others might be explained by the geographical differences in the availability of services and programs or might reflect a true reduction in prevalence over time following begging of vaccination program. Furthermore, the result of this study revealed that, the prevalence rate of HBsAg among blood donors from Sana’a and Taiz governments was 3.6% and 3.5% respectively compared to 4.1%, 4.2%, 4.9%, among blood donors from Ibb, Dahmar and Amran governments respectively. High prevalence rate of HBsAg among donors from Raymah 10.8% and Almahwet 10.3% governments followed by Alhuddidh 7.0%, while less prevalence among donors from Albaithad 3.9%. The difference in the prevalence rates of HBsAg. Numerous studies exhibited variation in the prevalence of HBV among blood donors, in relation to geographical, social and occupational status of blood donors. [14,17,19,23,24]

In this study it was found that HBsAg among students (3.7%), professional (3.7%) and handicraft (4.7% ) was less than that reported by Alodini et al. [13] and Wasfi et al., [23] who found that, 17.9% and 23.4%, to 11.9% and 21.3%, and 16.4 % and 46.8% of HBsAg among students, professional workers, and handicrafts respectively. This could be due to the differences in sample size, geographical and local culture. [14,17,19,23,24]

The prevalence of HBsAg among military our study was 4.4% compared to 28.4% and 6.1% reported by Alodini [13] and Al-Waleedi et al. 2012 [14] in Sana'a and Aden city respectively, in addition higher rate (10.6%) of HBsAg among military has been reported Nwobegahay et al., [25]

The prevalence of HBsAg among 25-36 age group in present study (4.5%) is in agreement to that (4.3%), reported by Abdullah, [27] who also reported that, 5.8% and 7.7% of HBsAg was among the age group 37-46 and more than 46 years old respectively which is slightly higher than the result of the recent study. In addition, the frequency of HBsAg among age groups up to 35 and from 36 to 55 years old was around 4.5% and 1% respectively. In comparison to 6.2% among age group more than 55 years old. This agrees to that reported by Al-Waleedi et al. [14]
Anti-HBc
It was found that, 437/3,606 (12.1%) were reactive for Anti-HBc alone compared to only 68 (1.9%) of blood donors had reactive to both markers (HBsAg and anti-HBc). High rate was reported by Sallam et al.[17] with an account 18.5% and 33.5% of donors in Sana'a city had only anti-HBc and both HBsAg and anti-HBc respectively. Olayinka et al.[28] reported that, 66.7% of blood donors had only anti-HBc and 29.2% had both anti-HBc and anti-HBsAg. Less prevalence rate (1.6% and 4.4%) has been reported by Muselmani et al.[29] and Salawu et al.[30] The differences in the prevalence rate may be due to a difference in the types and sensitivity/specificity of the detection techniques used and the sample size variation.[29,30]

CONCLUSION
This study established that our subjects still under the moderate to high-risk communities. The prevalence of HBV fluctuates according to geographical differences, occupational and age. Addition of anti-HBc tests to screen the blood donors along with HBsAg will enhance blood safety by rejecting the HBV infected donations. But unfortunately, this will lead to refusal of a large number of blood units.

RECOMMENDATION
Addition of anti-HBc (IgM and IgG) tests to screen the blood donors along with HBsAg as well as anti-HBs, result in elimination of a large number of blood units but lead to increase the safety measures in blood bank centers. In addition, it is better to add nucleic acid techniques for screening of donated blood to identify the most potentially infectious blood units for improving blood safety in our country.

ETHICAL APPROVAL
Approval for this study was obtained from the National Blood Transfusion and Research Centre in Sana'a. The purpose and procedures of the study were explained to all individuals, and a written informed consent was obtained from all of them.

COMPETING INTERESTS
Authors have declared that no competing interests exist.

ACKNOWLEDGMENTS
We would like to thank all coworkers in general and in particular, all staff members in virology unit in the National Blood Transfusion and Research Centre in Sana’a Yemen. The authors are grateful to the members of the blood banks’ teams for technical help and to all blood donors participated in this work.

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