SEROPREVALENCE OF HEPATITIS B VIRUS AMONG MULTI-TRANSFUSED BETA THALASSEMIA PATIENTS ATTENDING AT THE YEMENI SOCIETY FOR THALASSEMIA - SANA'A, YEMEN

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Abstract

Regular blood transfusion is one of the most important and emergency treatment methods for thalassemia patients. This category of patients is considered sensitive to infection because their continued survival is linked to blood transfusion, and therefore the possibility of infection is great, especially in light of the war and the deterioration of health services in Yemen. Among this infection is the hepatitis B virus, which causes serious complications such as acute hepatitis, liver cirrhosis and hepatocellular carcinoma (HCC). The aim of the study was done to determine incidence of hepatitis B virus and risk factors among Multi-Transfused Beta Thalassemia Patients attending at The Yemeni Society for Thalassemia and Genetic Blood Disorder (YSTGBD) Sana’a –Yemen. This study was conducted during the period from January 2021 to December 2021, during which 200 Blood samples were collected from beta thalassemia patients attending at (YSTGBD). All samples were examined using the ECLIA technique, during which the basic information was collected through special questionnaires for this purpose. The rate of incidence by HBsAg in all study samples was determined to be 7(3.5%). The study showed that there was a statistically significant relationship between infection with the hepatitis B virus and the age group of more than 10 years age \( P = 0.042 \), in addition to that number of blood transfusions received by HBsAg positive was significantly higher than that of HBsAg negative thalassemia patients \( P = 0.022 \).

Keywords: \( \beta \)-thalassemia patients, Hepatitis B virus, Yemeni Society for Thalassemia (YST).

1. Introduction

Thalassemia is one of the most common genetic diseases worldwide [1]. \( \beta \)-thalassemia constitutes a major health problem in Yemen. There are about 632 Beta-thalassemia patients that estimated Yemeni society for Thalassemia And genetic Blood disorders in Sana'a, Yemen. The prevalence of \( \beta \) thalassemia trait is 4.4% with an estimated incidence of 11.3/10,000 of homozygous [2]. Recent data indicate that about 7% of the World’s population is a carrier of a hemoglobin disorder and that 3,000,000-5,000,000 children are born each year with the severe homozygous states of these diseases [3] Regular blood transfusion in patients with thalassemia has improved their overall survival and quality of life, but it can increase risk of transmission of blood born viral infections, especially HBV [4].

2. Subjects and Methods

2.1 Study population and Setting:

This cross-sectional study was carried out using patients with \( \beta \)-thalassemia Patients \( n= 200 \) attending at the Yemeni Society for Thalassemia in Sana’a. Ethical approval for the study was obtained from Department of Biological Science, Faculty of Sciences, Sana'a University. Information was collected by questionnaire that included demographical data, including: (age, gender, frequency of blood Transfusions, and risk factors including Hepatomegaly, splenectomy, splenomegaly and Iron overload).

2.2 Samples Collection:

5-ml of Blood samples were collected into sterile tube before the blood transfusion allowed to clot at room temperature for 25 minutes, and centrifuged.
2.3 Sampling processing:
The sera were transported inside cryo box to Molecular Biology Research Center (USTY) Sana'a, at the time of study the determinate of Hemoglobin (Hb) levels, Serum Ferritin Levels, Alanine Transaminase level (ALT) were performed by using Semi-Automatic Clinical Chemistry Analyzer spectrophotometer (Mod-Analyzer BAS-100 plus, Labomed, USA). All sera separated into Eppendorf tubes were labeled and stored in deep freeze at -20°C. Serological assay were performed by using the Electrochemiluminescence immunoassay (ECLIA) method on Roche cobs e 411, the ECLIA kits used to serological analysis of HBsAg infection markers amongst the participants in this study were performed by using Roche Cobas reagents [5].

2.4 Statistical analysis:
The collected data was coded and entered in a data base file. After complete entry, data were transferred to the IBM SPSS statistics 21 (Copyright IBM Corporation, 2012). The Chi-square test for categorical variables was used to find significant associations between patient’s characteristics and HBV positivity. Statistical tests were conducted at the P < 0.05 significance level.

3. Results:

3.1 Patients disposition:
in this study, a total of two hundred multi-transfused beta-thalassemia patients, 111(55.5%) males and 89(44.5%) Females were enrolled during a period of Jun 2021 to Dec. 2021 in Sana'a city in this study, 7(3.5%) were positive for Hepatitis B Virus. All thalassemic patients collected from 12 governorates. The average age of patient was divided in two groups, ≤ 10 years age group was 103(51.5%) and > than 10 years age group was 87(48.5%).

3.2 Distribution of Hepatitis B Virus according the socio demographic condition:
The current study results presented prevalence of the HBV-positive among thalassemia patients were found 6 (3%) among more than 10 years age group, followed 1(0.5%) among ≤ 10 years age group There was significant association between HBsAg positive and > than 10 years of blood transfusion age group, Chi-Square = 4.140, P = 0.042 and OR= 8.8, the high prevalence of HBV was recorded in the group <100 times of blood transfusion 6 (3%), while was 1(0.5%) in >100 times of blood transfusion group., the sero-prevalence of HBV markers (HBsAg) was 2 (1%) in male and 5 (2.5%) in female thalassemia patients Table 1.

Table 1: Distribution of HBsAg positivity according to their socio demographic condition:

| HBsAg        | Positive (n=7) | Negative (n=193) | Total | P. value X2 OR |
|--------------|----------------|------------------|-------|----------------|
| Age          |                |                  |       |                |
| ≤ 10 years   | 1 (0.5)        | 102 (51)         | 103   | 51.5           |
| > than 10    | 6 (3)          | 91 (45.5)        | 97    | 48.5           |
| years        | 7 (3.5)        | 193 (96.5)       | 200   | 100            |

Number of blood transfusion time

| Blood transfusion time | ≤ 10 years of blood transfusion | > than 10 years of blood transfusion | Total |
|------------------------|---------------------------------|-------------------------------------|-------|
|                        | 1 (0.5)                         | 7 (3.5)                             | 8      |

Gender

| Gender | Male | Female | Total |
|--------|------|--------|-------|
|        | 2    | 5      | 7     |

3.3 Distribution of HBsAg positivity according to their Governorates:

Regions wise distribution of the prevalence of HBV-positive among thalassemia patients were found, 1 (14.3%) were from Amran, 2 (28.6%) from Hajjah, 1(14.3%) from Al-Hudaydah, 1 (14.3%) from Rima, 1(14.3%) from Sana’a, 1(14.3%) from Taiz, and sero-negative of HBV were found in Al-Bayda, Ibb, Al-Mahwit, Al-Dhali, Sa’da, and Dhamar Table 2.

Table 2: Distribution of HBsAg positivity according to their Governorates

| Governorates | Positive (n=7) | Negative (n=193) | Total |
|--------------|----------------|------------------|-------|
| Amran        | 1 14.3         | 31 16.1          | 32 16 |
| Al-Bayda     | 0 0            | 4 2              | 4 2   |
| Al-Dali      | 0 0            | 1 0.5            | 1 0.5 |
| Hajjah       | 2 28.5         | 38 19.7          | 40 20 |
| Al-Hudaydah  | 1 14.3         | 10 5.2           | 11 5.5|
| Ibb          | 0 0            | 22 11.4          | 22 11 |
| Al-Mahwit    | 0 0            | 27 14.1          | 27 13.5|
| Rima         | 1 14.3         | 8 4.1            | 9 4.5 |
| Sa’da        | 0 0            | 1 0.5            | 1 0.5 |
| Sana’a       | 1 14.3         | 24 12.4          | 25 12.5|
| Taiz         | 1 14.3         | 9 4.7            | 10 5  |
| Dhamar       | 0 0            | 18 9.3           | 18 9  |

Total 7 100 193 100 200 100

\( X^2 = \text{Chi-Square}, \text{OR} = \text{Odd Ratio} \).
3.4 Distribution of HBsAg positivity according to Clinical examination:

The rates HBV viral infection in thalassemia patients according to clinical examination were 7 (3.5%) pallor, 2 (1%) splenectomy, 3 (1.5%) jaundice, 1 (0.5%) splenomegaly, 5(2.5%) Iron overload and 2(1%) enlarged Spleen, Table 3.

Table 3: Distribution of HBsAg positivity according to Clinical examination

| HBsAg          | Positive (n=7) | Negative (n=193) | Total | P. value |
|----------------|---------------|------------------|-------|----------|
| Pallor         |               |                  |       |          |
| Yes            | 7             | 169              | 176   | 0.089 X2 = 0.989 P= 0.320 (Non-Sig.) |
| No             | 0             | 24               | 24    |          |
| Total          | 7             | 193              | 200   | 0.0004 X2 = 8.08 (Sig.) |
| Jaundice       |               |                  |       |          |
| Yes            | 3             | 18               | 21    | 0.001 X2 = 27.7 P= 0.01 (Sig.) |
| No             | 4             | 175              | 179   |          |
| Total          | 7             | 193              | 200   | 0.150 X2 = 2.07 (Non-Sig) |
| Splenomegaly   |               |                  |       |          |
| Yes            | 1             | 0.5              | 1     | 0.5      |
| No             | 6             | 193              | 199   |          |
| Total          | 7             | 193              | 200   | 0.035 X2 = 15.10 P= 0.0035 (Sig.) |
| Spleenectomy   |               |                  |       |          |
| Yes            | 2             | 21               | 23    | 0.051 X2 = 4.10 P= 0.052 (Non-Sig) |
| No             | 5             | 172              | 177   |          |
| Total          | 7             | 193              | 200   |          |
| Iron overload  |               |                  |       |          |
| Yes            | 5             | 2.5              | 184   | 92       |
| No             | 2             | 14               | 16    |          |
| Total          | 7             | 193              | 200   |          |
| Enlarged Spleen|               |                  |       |          |
| Yes            | 5             | 2.5              | 48.5  | 51       |
| No             | 2             | 96               | 98    |          |
| Total          | 7             | 193              | 200   |          |

3.5 Distribution of HBsAg positivity according to their laboratory tests:

The distribution was not significantly association with Ferritin level, less than 1000 μg/L group was 0 (0%) in positive cases compared to 5 (2.5%) cases in negative cases. While Ferritin level more than 1000 μg/L group was 7 (3.5%) in positive cases compared to 188 (94%) cases in negative cases, (P = 0.666). There was highest significantly (P= 0.001) association with the Alanine transaminase (ALT) level in the Normal range: 0-40 U/L were 0 (0%) in positive cases compared to 121 (60.5%) cases in negative cases. While the high level group (> 45 U/L) were 7 (3.5%) in positive cases compared to 72 (36%) cases in negative cases.

| Ferritin level (μg/L) | Positive (n=7) | Negative (n=193) | Total | P value |
|-----------------------|---------------|------------------|-------|---------|
| (<1000 μg/L)          | 0             | 5                | 5     | 0.001 X2 = 0.018 P= 0.666 |
| (>1000 μg/L)          | 7             | 188              | 195   |         |
| Total                 | 7             | 193              | 200   | 0.001 X2 = 11.1 |

4. Discussion

The results obtained in this study indicated that HBsAg positive were 7(3.5%) of the samples studied (n= 200). Another study in Egypt reported 4.21% were infected with hepatitis B virus [6]. Another studies (29%) and (32.5%) were reported positive for HBsAg among thalassemia patients in Egypt [7, 8] and Several studies such as in Palestine 23.3% [9], Pakistan 7.4% [10]. Another study was conducted in which total 32 of Thalassemia Patients in West North of Iran observed All of them Patients were seronegative for HBs antigen [4].

Showed similar results for the prevalence of hepatitis virus among thalassemia patients in a global countries such as in Pakistan 3.13% [11], and 3.69% in West Bengal [12]. Another studies reported 6.4% in Taiwan [13] and 6.31% in Pakistan [14]. Low incidence of HBsAg (1.5%) was found among thalassemia patients in India [15]. In our study, the average age of patient was divided in two groups, ≤ 10 years age group was 103(51.5%) and > than 10 years age group was 87(48.5%). In the present study, it was found that there high significant different between the prevalence of HBV infection in relation to age group. The highest incidence was 6 (3%) observed among > than 10 years age group, followed 1(0.5%) among ≤ 10 years age group, X2 = Chi-Square Value =4.140, P. value = 0.042 and OR= 8.88.Seronegative for HBsAg in 0–5 years age group while 3 (7.7%) in 5–10 years age group were positive for HBsAg and 1 (4.0%) were positive in 10–15 years age group reported by Jaiswal et al [16]. Another studies reported 0/27 children aged 1–5 years and 1/59 children aged 6–10 years [17]. In the current study there was significant association between HBsAg positive and more than 100 times of blood transfusion, X2 = 5.71, P = 0.022 and OR= 6.8, this study a similar finding Egyptian study was reported by [6], this is due to non-compliance with international standards and protocols in blood
transfusion procedures. In the present study, there was no significant association between HBsAg positive and gender 1% in male 2.5% in female (X² = 2.13 P. value = 0.144). In the current study there was no significantly association different between HBsAg positive and some clinical examination such as pallor 7 (3.5%) P = 0.320 and Splenectomy was 2 (1%) P = 0.150. While a significantly associated with jaundice 3 (1.5%) P = 0.004), high significantly associated with Splenomegaly, 1 (0.5%) and P. value = 0.01. **Ramadan et al**, found that no significant association between HBsAg seropositive and Splenectomy [6]. In the current study there were a significant association between HBsAg seropositive and iron overload were detected 5(2.5%), while there was no significant with enlarged spleen were detected 5(2.5) P. value =0.052. There was elevated of serum Alanine transaminase level (ALT) was 7 (100%) in HBsAg positive cases P. value = 0.001, this investigation findings conducted by Ramadan et al [6].

5. Conclusions

According to present study the following facts can be concluded: low rate for HBsAg Positive sign among beta thalassemia patients that attending at the YST and these seropositive cases were significantly associated with older age, Multi blood transfusion, some clinical examination such as (jaundice, iron overload and Splenomegaly) and elevated of serum Alanine transaminase level (ALT). The risk factors associated with old age and frequency blood transfusion.

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