STUDY OF CLINICAL PROFILE OF TRANSFUSED THALASSEMIC CHILDREN WITH SPECIAL REFERENCE TO HEPATITIS B PROFILE AND LIVER FUNCTION.

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Abstract:
Present study was conducted to assess the thalassemia patients and to find out the sero-prevalence of Hepatitis B among the thalassemic patients. The study was conducted in two tertiary care hospitals of Quetta city of Balochistan Province of Pakistan. A total of 289 confirmed thalassemic patients were screened for Hepatitis B virus. Data was collected on a pre-organized Performa that included patient’s demographics and general information. Patient’s clinical profile was recorded and blood samples were collected for laboratory analysis. The liver function test (ALT, ALP and Bilirubin) was performed. The results were then subjected to statistical analysis using SPSS. The study showed a prevalence of 3.1% of HBsAg patients in our population. The study also indicated elevated levels of ATL and ALP indicating liver infections among population. The study recommends using more accurate methods of screening for hepatitis B and C to reduce the high risk of transfusion hepatitis as the lives of these sick patients rely on blood transfusions.

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INTRODUCTION:
Thalassemia major is an inherited disorder significantly common in people of Mediterranean, South-east Asian and African [1]. Including Pakistan where thalassemia minor is estimated about 5–7% and thalassemia major is estimated approximately 100.00 patients are suffering from thalassemia and this number is elevating about 5–9000, annually (kiani et al.). It is characterized by decreased production of beta chain of hemoglobin [2], microcytic anemia due to defective hemoglobin synthesis. B-thalassemia is an autosomal recessive disorder occurs because of reduced or absent synthesis of alpha or β-globin chains [3], encoded by a structural gene found on the short arm of chromosome [4].

Approximately, 1.5% has been investigated to be carriers for β–thalassemia being born each year (kiani et al). β-thalassemia major, also known as Cooley’s anemia, is characterized by grossly defective synthesis of hemoglobin A, increased hemolysis of the defective RBCs, and excess of globin chains produced by the unaffected gene in which the excess of alpha chain will form unstable accumulates that distress the stability of erythrocyte membrane causing hemolysis of the red cell precursors in the bone marrow and spleen resulting in severe anemia [5]. It develops during the first year of life, the hemoglobin level falls to less than 7g/dl [6].

The clinical course of thalassemia major patients is short unless transfusion is given. Regular blood transfusion in patients of thalassemia has improved their overall survival, but transfusion of blood carries great risk of blood-borne virus infections, significantly viral hepatitis. Nowadays, vaccination against hepatitis B has efficiently been able to constrain the transmission of hepatitis B virus (HBV) infection. The prevalence of HBV infection among thalassemic patients has been reported to be up to 60% in Italy, although the compulsory screening of donated bloods has decreased the incidence of post-transfusion HBV [7].

The purpose of the present study was to investigate the prevalence of infection in thalassemia patients and to determine the sero-prevalence of Hepatitis B among patients attending tertiary care hospitals of Quetta. No study till date has reported such data so it will help assess the risk factors related to transfusion dependent thalassemia.

MATERIAL AND METHODS:
In this cross-sectional study a total of 289 with a confirmed thalassemia patients were included in this study in which 132 were male and 157 were female with the different age group of 6 months to 24 years old, receiving regular transfusion therapy at the Thalassemia Care Center at Bolan Medical Complex (BMC). Ethical approval for the study was obtained from the Medical Officer of BMC and written informed consents were obtained from all guardians/parents of the children prior to data and sample collection. The patients were subjected to a detailed history including number of transfused units, a thorough clinical examination and biochemical investigations. Special emphasis was given on personal history (age, gender, and location), family history (parent consanguinity, and family history of similar conditions), clinical data (age of diagnosis and number of blood transfusions) and laboratory data (liver enzymes) were recorded.

For laboratory assessment, 7ml of blood was obtained using sterile venipuncture into a tube. Sera were separated, all quoted and stored at −20°C until used. For each patient, liver enzymes (alkaline Phosphatase, alanine transaminase and Bilirubin). In children receiving blood transfusions, samples were drawn before packed-RBC transfusion. Serum was collected in tube for serological testing and centrifuged 3000 rpm for at least 20 minutes. The serum was kept at −20°C until the time of the assay. For the serological tests HBsAg were measured by the enzyme linked immunosorbent assay ELISA (Diamat 710) following the manufacturer’s instructions. Briefly, the instrument uses a disposable pipette tip called the solid-phase receptacle, which is coated with antigens and also acts as a pipetting device. All the ready-to-use reagents are contained in a sealed strip. The specimen (serum or plasma) is added to the reagent strip, and all the following steps of the test are done automatically, without any further manipulation.

RESULTS:
In the current study total of 289thalassemiapatients were included out of which only 9 (3.11%) were positive for HBV and while remaining 280 (96.8%) were normal/negative.132 male patients exhibited 126 (95.4%) HBsAg negative and 6 (4.6%) HBsAg positive. In 157 female patients 154 (98%) was HBsAg negative and 3 (1.9%) were found HBsAg positive (Fig 1) (Table 1).

Liver function tests in (Figure 2) (Table 2) showed the increased levels of following enzymes. ALT in which 30.1% was in negative and 51% were found positive for HBsAg. Alkaline phosphate level was 134% in HBsAg negative patients and 255% in HBsAg positive patients. Bilirubin level was found 8% in negative and 1% in positive HBsAg patients.
Fig 1. Gender wise prevalence of HBsAg in thalassemic patients

Table 1. Total percentage of HBsAg in male and female thalassemic patients

<table>
<thead>
<tr>
<th>Gender</th>
<th>Male patients</th>
<th>Female patients</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>HBsAg negative (%)</td>
<td>132 (95.4%)</td>
<td>157 (98.1%)</td>
<td>289 (96.9%)</td>
</tr>
<tr>
<td>HBsAg positive (%)</td>
<td>126 (4.6%)</td>
<td>154 (1.9%)</td>
<td>280 (3.1%)</td>
</tr>
</tbody>
</table>

Fig 2. Liver function test results in patients

Table 2. Liver function test results in thalassemic patients which show ALT are increase in all HBs Ag positive patients.

<table>
<thead>
<tr>
<th>Liver function test</th>
<th>Negative % levels</th>
<th>Positive % levels</th>
</tr>
</thead>
<tbody>
<tr>
<td>ALT</td>
<td>30.1%</td>
<td>51%</td>
</tr>
<tr>
<td>ALP</td>
<td>134%</td>
<td>255%</td>
</tr>
<tr>
<td>Bilirubin</td>
<td>.8%</td>
<td>1%</td>
</tr>
</tbody>
</table>
DISCUSSION:
Regular blood transfusion remains the life line for thalassemic patients due to which they are more prone to various infections. This study showed 3.1% prevalence of HBsAg in thalassemic, as compared with older studies conducted worldwide the prevalence of HBV serology was higher than in our study in the following studies. Mansour et al, [9] reported 44% cases of thalassemia patients who were HBV positive. Elbahrawy et al, [10] investigated 32.5% hepatitis B in Eighty Egyptian thalassemia children. Bhavsar et al, [11] studied the prevalence of hepatitis B infection in thalassemia major patients in tertiary care hospital, Gujarat, and found 6% HBsAg. In a study conducted in Swat by Khattak et al, [12] reported 5.88% patients suffering from HBV. Khan et al, [13] studied 240 patients in which 12 (5%) exhibited positivity for HBV and 1 (0.41%) exhibited positivity for both HBV and HCV. In other studies, reported 6.4% cases of HBV by Rahman and Lodhi [14], 17% was reported by Kapoor et al, [15], 8.4% was reported by Shah et al, [16], 5.1% of HBV by Riyaz [17] and 7% by Calderon [18] in their studies. 

In studies by Gira et al, [19] reported 3.3% at the beginning of the study and 3.66% at the end of the studies. In another study by Chattopadray et al, [20] in Kolkata showed 14 (3.69%) of HBV positive among 379 thalassemic patients and 3(0.79%) cases of both HCV and HBV positive. These studies are in accord to the present study.

Other studies reported the lower serology than our study, from India 13 (2.8%) cases were reported positive for HBsAg by ELISA by Makroo et al, [21]. Ansari et al, [22] studied for sero-positivity of hepatitis C, hepatitis B, and HIV in chronically transfused β-thalassemia major patients in Pakistan and found that out of 160 patients, 21 (13.1%) cases were anti-HCV positive, and 2 (1.25%) were HBsAg positive. In other studies, HBsAg was investigated 2.4% cases of HBV by Jamal et al, [23], 2% by Chakrabarti [24], 2% by Vidga [25], 1.5% by Mirmomen et al, [26],0.5% in a research by Aza [27], 0.2% in a study by Remsar [28]. The low rate of HBsAg can be due to vaccination, which may reduce the prevalence of diseases in thalassemic patients.

CONCLUSION:
Pakistan is facing a great increase in thalassemic patients due to the inadequacy of any proper screening and policy in country. The transmission of HBV and other infections are huge problems where blood safety standards are not very high.

RECOMMENDATIONS:
Blood cannot be manufactured and there is no substitute for it. It is a constant demand for thalassemic worldwide. It is necessary to develop societies of young blood donors who donate regularly to save lives. Thalassemia major patients are not able to regulate normal hemoglobin level so they need regular blood transfusions. Without regular blood transfusions support about 85% of patients with β-thalassemia major would die. It is constant need of their lives to provide in time safe and match transfusion regularly to reduce complications.

Therefore, it is recommended to use more accurate methods of screening for hepatitis B to reduce/eliminate this high risk of transfusion hepatitis as the lives of these sick children are solely on blood transfusions.

REFERENCES:
status (human immunodeficiency virus, hepatitis B virus and hepatitis C virus) in multiply transfused thalassemia patients of North India. Indian Journal of Pathology and Microbiology, 56(4), 378.


