

## Prevalence and Risk Factors of Hepatitis B and Hepatitis C Virus Infections in Repeatedly Transfused Thalassemia Patients in a Tertiary Care Hospital

Dr. Kanta Halder<sup>1\*</sup>, Dr. Md. Selimuzzaman<sup>2</sup>, Dr. Riffat Mohiuddin<sup>3</sup>, Dr. Md. Naim Hossain Ratan<sup>4</sup>, Dr. Nazmul Hasan<sup>5</sup>, Dr. Shahrina Afroze Tisha<sup>6</sup>, Dr. Md. Kamrul Hasan<sup>7</sup>, Dr. Md. Saiful Islam<sup>8</sup>

<sup>1</sup>Resident Medical Officer, Department of Neonatology, Bangladesh Shishu Hospital & Institute, Dhaka, Bangladesh

<sup>2</sup>Professor & Head, Department of Pediatrics Hematology & Oncology, Bangladesh Shishu Hospital & Institute, Dhaka, Bangladesh

<sup>3</sup>Assistant Professor, Bashundhara Addin Medical College & Hospital, Dhaka, Bangladesh

<sup>4</sup>Assistant Professor, Department of Pediatrics, East-West Medical College and Hospital, Dhaka, Bangladesh

<sup>5</sup>Assistant Professor, Department of Pediatrics, Ashiyan Medical College and Hospital, Dhaka, Bangladesh

<sup>6</sup>Consultant, Pediatrics, Sajida Hospital, Dhaka, Bangladesh

<sup>7</sup>Registrar, Department of Pediatrics, Monno Medical College & Hospital, Manikgonj, Bangladesh

<sup>8</sup>RMO, Department of Neonatology, Bangladesh Shishu Hospital & Institute, Dhaka, Bangladesh

DOI: [10.36347/sjams.2023.v11i01.010](https://doi.org/10.36347/sjams.2023.v11i01.010)

| Received: 11.11.2022 | Accepted: 19.12.2022 | Published: 11.01.2023

\*Corresponding author: Dr. Kanta Halder

Resident Medical Officer, Department of Neonatology, Bangladesh Shishu Hospital & Institute, Dhaka, Bangladesh

### Abstract

### Original Research Article

**Background:** Thalassemia patients who are conventionally treated by regular transfusion regimen are at a risk of acquiring transfusion transmitted infections, including hepatitis B and hepatitis C. Getting blood transfusion in different places makes them vulnerable to these blood borne infections. It is important to assess and update the prevalence of these infections along with their contributing factors for ensuring optimum preventive measures and further strengthening of the screening program. **Objectives:** To estimate the prevalence of hepatitis B and hepatitis C virus infections in repeatedly transfused thalassemia patients and to determine the risk factors for acquiring these infections. **Methods:** This cross-sectional study was carried out in the Department of Pediatric Hematology and Oncology, Bangladesh Shishu Hospital & Institute, Dhaka, Bangladesh during the period July 2018 to December 2019. Total 73 thalassemia patients of 2 to 18 years were enrolled into the study following the inclusion and exclusion criteria. Demographic data and other related information were recorded in a standard data sheet. Hb%, SGPT, HBsAg, Anti-HBs titre, Anti-HCV were done in all patients. Collected data was checked and analyzed by computer based program SPSS version 26.0 for Windows. **Results:** Out of total 73 thalassemia patients, 44 were male and 29 were female. Mean age was  $8.3 \pm 3.45$  years where maximum number of patients belonged to 6-10 years. 2(2.7%) patients had positive HBsAg and 11(15.1%) had positive Anti-HCV antibody at the end of study. Prevalence of hepatitis B infection was associated with lack of immunization against it which was statistically significant ( $P < 0.001$ ). Hepatitis C virus infection in thalassemia patients was significantly associated with increasing duration of transfusion ( $P = 0.043$ ), frequency of transfusion ( $P < 0.001$ ) and elevated SGPT level ( $P < 0.001$ ). Comparing Anti-HBs titre, it is also found that there was decreased level of immunity against hepatitis B in older age group ( $P < 0.001$ ). **Conclusions:** Prevalence of hepatitis B is 2.7% and hepatitis C virus infection is 15.1% in repeatedly transfused thalassemia patients in a tertiary care hospital (Bangladesh Shishu Hospital & Institute) and older age group with increased duration and frequency of transfusion are associated with increasing hepatitis B and hepatitis C virus infections.

**Keywords:** Thalassemia, Regular transfusion regimen, transmitted infections, Hepatitis B, Hepatitis C, Blood borne infections.

Copyright © 2023 The Author(s): This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International License (CC BY-NC 4.0) which permits unrestricted use, distribution, and reproduction in any medium for non-commercial use provided the original author and source are credited.

## INTRODUCTION

Thalassemia is one of the most common genetic diseases in the world. It is a major health problem, causing morbidity, early mortality and a great deal of misery for a family both financially and

emotionally [1]. About 15 million people have clinically apparent thalassemic disorders worldwide [2]. About 23% of world's population resides in South Asia. In South Asia, most studies regarding thalassemia conducted in India. In India the overall prevalence of beta thalassemia carriers has been estimated to be

**Citation:** Kanta Halder, Md. Selimuzzaman, Riffat Mohiuddin, Md. Naim Hossain Ratan, Nazmul Hasan, Shahrina Afroze Tisha, Md. Kamrul Hasan, Md. Saiful Islam. Prevalence and Risk Factors of Hepatitis B and Hepatitis C Virus Infections in Repeatedly Transfused Thalassemia Patients in a Tertiary Care Hospital. Sch J App Med Sci, 2023 Jan 11(1): 46-54.

between 2.78 and 4%, whereas in Pakistan it is 5-7% [3]. Bangladesh lies in the world's thalassemia belt, but information about epidemiology, clinical course, mortality, complications and treatment outcomes of thalassemia is lacking. However, World Health Organization (WHO) estimates that approximately 3% of our populations are carriers of beta-thalassemia and 4% are the carriers of hemoglobin E (HbE) in Bangladesh [3]. Although red cell transfusions are the mainstay of treatment for patients with thalassemia, they are responsible for a series of complications and expose the patients to a variety of risks. These patients are already compromised with various disease related complications e.g. iron toxicity, hypersplenism, venous thrombosis and osteoporosis. Moreover, receiving repeated transfusions, they are confronted by new clinical challenges, particularly in the form of transfusion transmitted disease, especially hepatitis B virus and hepatitis C virus [4]. Prevention of transfusion transmitted viral infection is one of the important aspect of management of transfusion dependent thalassemia patients. Most effective way of controlling these infections are by immunization and effective donor screening [5]. Bangladesh has integrated the hepatitis B (HepB) vaccine in Expanded Program on Immunization, which was primarily initiated in 2003 and was then expanded in 2005 to all districts [6]. Nowadays, vaccination against HBV has efficiently been able to restrict the transmission of Hepatitis B virus infection. However, post-transfusion transmission of Hepatitis C Virus has still remained a major health concern in thalassemic patients [7]. Although rigorous donor screening, testing procedures and suitable donor selection programs have reduced transmission of HCV via transfusion of blood products, there are still many countries where standards of blood product management do not adequately protect chronically transfused patients like thalassemia patients from complications [8]. Various studies among multi-transfused thalassemic children in different countries demonstrated a wide range of prevalence of transfusion transmitted infections particularly in developing countries where maintaining the standard of pre-transfusion screening of blood and blood products looks challenging [9]. According to national guidelines for screening donated blood for transfusion transmissible infections in Bangladesh (2013), mandatory screening of donated blood for transfusion has begun in 2000 after implementation of the Safe Blood Transfusion Program (SBTP) at all hospital based blood transfusion centers [10]. To assess the donor screening quality of different blood transfusion centers of Bangladesh, WHO, DGHS (Director General Health Services) & IEDCR (Institute of Epidemiology, Disease Control and Research) has conducted a study in 2013 which includes 27 testing centers. Among them 9(33.33%) centers had incorrect results when compared with the results obtained at IEDCR. HBV (53.3%) is the commonest one among the incorrect ones followed by HCV (40%) and malaria (6.7%). Currently the hospitals and blood donation

centers use Rapid Device (Strip) method for screening of the donated blood. But presence of transfusion transmitted viral infections indicates that there are some pitfalls in currently practicing screening system [11]. There is chance of false positive/negative results in case of rapid strip test. A false positive result can unnecessarily exclude a potential blood donor. On the other hand, a false negative result poses a great challenge to the reliability of the blood screening system. Improperly stored or transported samples can also alter the test results. These limitations could be overcome when screening is done by ELISA, chemiluminoassay or PCR technique [12]. So, there is a constant need to explore the effect of currently used protocols of blood donor screening by determining the prevalence of transfusion transmitted infections in multitransfused patients.

## OBJECTIVES

### General Objective:

To estimate the prevalence and to identify the risk factors for hepatitis B and hepatitis C virus infections in repeatedly transfused thalassemia patients in Bangladesh Shishu Hospital& Institute.

### Specific Objectives

- To observe the prevalence of HBV seropositive cases in thalassemia patients.
- To observe the prevalence of HCV seropositive cases in thalassemia patients.
- To identify the demographic and clinical factors associated with seroprevalence.
- To see the immune response to hepatitis B vaccination in thalassemia patients.

## METHODOLOGY AND MATERIALS

This was cross sectional observational study by using simple random sampling method, conducted from the July 2018 to December 2019, on the children with thalassemia who received repeated blood transfusion in outpatient department of Thalassemia Center of Bangladesh Shishu Hospital& Institute, Dhaka, Bangladesh. A total 73 patients were included as study population in the age from 2 to 18 years.

### Study Procedure

Thalassemia patients attending Bangladesh Shishu Hospital& Institute were selected on the basis of inclusion criteria. The parents or legal guardians were informed of the purpose of the study and their informed written consent was taken. Detailed family history including history of consanguinity, family history of same type of illness and history of jaundice or HBV and HCV positivity in family was taken. In addition to that, history of immunization against HBV, socio-economic status, H/O regular blood transfusion, duration of receiving blood transfusion, total number of transfusion and frequency of transfusion per year, type of donor and

number of centers used for transfusion, need for iron chelation therapy or history of splenectomy was also taken. Physical examination was carried out in all patients to assess hepatomegaly, splenomegaly and growth status. Then with all aseptic precaution, 5 ml of venous blood was collected from ante-cubital vein of each participating child for biochemical and serological test. Blood for Hb% and SGPT was collected in a EDTA containing tube and sent to the laboratory of Bangladesh Shishu Hospital & Institute, while blood for hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (anti-HBs) and antibody to hepatitis C (anti-HCV) was collected in a tube without anticoagulant, serum was separated and sent to a private standard laboratory recommended by Bangladesh Shishu Hospital & Institute for analysis. Serological tests were assessed using a third generation enzyme-linked immunosorbent assay (ELISA) and estimation was carried out by Abbott architect i2000SR/VitrosECi System J&J/ Siemens Advia Centaur XP Random Access Multibatch Immunoassay analyzer. Patients who were seropositive for HBV or HCV, were referred to Hepatologist for further management and booster dose

of recombinant hepatitis B vaccine was advised for the patients having Anti-HBs titer <10 mIU/L.

### Data processing and data analysis

Statistical analysis was carried out using the SPSS version 26.0. The quantitative observations were indicated by frequencies and percentages, while qualitative data were presented by mean and standard deviations. Chi square test was used for qualitative variables and unpaired t-test was used for quantitative variables. P values <0.05 were considered as statistically significant.

### Ethical consideration

Informed written consent was taken from parents or legal guardian. Ethical clearance was taken from the ethical review committee of Bangladesh Shishu Hospital & Institute (No. BICH-ERC-15/02/2018).

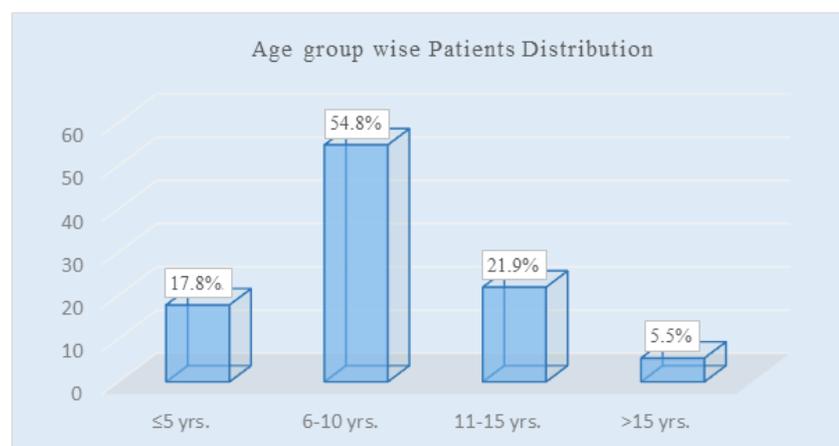
## RESULTS

**Table I: Demographic characteristics of the study patients (N=73)**

Variables	Frequency (n)	Percentage (%)	Mean $\pm$ SD	Range
Age (years)				
$\leq 5$ yrs.	13	17.8	8.3 $\pm$ 3.45	3.0-17.50
6-10 yrs.	40	54.8		
11-15 yrs.	16	21.9		
>15 yrs.	4	5.5		
Gender				
Male	44	60.3		
Female	29	39.7		
Male : Female ratio	1.5:1			
Consanguinity	10	13.7		
Family H/O thalassemia	15	20.5		

Table I showed the majority of patients belonged to age 6-10 years. The mean age was 8.3 $\pm$ 3.45 years ranging from 3 years to 17 years 6 months.

Almost two-third (60.3%) patients were male with male: female ratio 1.5:1. 13.7% patients had H/O consanguinity and 20.5% had family H/O thalassemia.



**Figure 1: Bar chart showed age group wise distribution (N=73)**

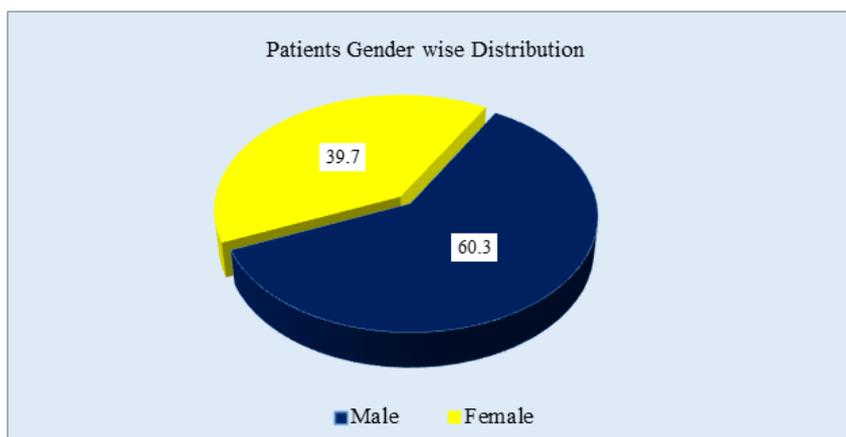


Figure 2: Pie chart showed patients gender distribution (N=73)

Table II: Clinical characteristics of the study patients (N=73)

Variables	Frequency (n)	Percentage (%)	Mean $\pm$ SD	Range
Type of thalassemia				
$\beta$ -thalassemia major	37	50.7		
Hb-E- $\beta$ thalassemia	36	49.3		
Age of diagnosis(months)			17.03 $\pm$ 6.75	8 - 48
Total duration of transfusion (years)			6.86 $\pm$ 3.56	1.67-16.25
Frequency of transfusion (years)			9.23 $\pm$ 3.03	4.0-24.0
Pre-transfusion Hemoglobin (gm/dl)			6.53 $\pm$ 0.64	5.30-8.50
Immunization status (against HBV)				
Immunized	70	95.9		
Not immunized	3	4.1		
Type of donor				
Voluntary	46	63.0		
Professional	27	37.0		
Centers used for blood transfusion				
Single center	18	24.7		
Multiple centers	55	75.3		
History of splenectomy	2	2.7		
Need for iron chelation	36	49.3		

Table II showed the clinical characteristics of study population. It was observed that 37(50.7%) patients had  $\beta$ -thalassemia major and 36(49.3%) had HbE- $\beta$ thalassemia. The mean age of diagnosis was 17.03 $\pm$ 6.75 months, mean total duration of transfusion was 6.86 $\pm$ 3.56 years and mean frequency of transfusion was 9.23 $\pm$ 3.03 per year. Total 70 (95.9%) patients were

immunized against hepatitis B. 46(63.0%) patients received blood from voluntary donor and 27(37.0%) patients from professional donor. 18(24.7) patients received transfusion in only 1 authorized center, while 55(75.3%) used multiple centers for transfusion. 2(2.7%) patients had H/O splenectomy and 36(49.3%) needed iron chelation therapy.

Table III: Prevalence of hepatitis B and hepatitis C virus infection in thalassemia patients (N=73)

	Frequency (n)	Percentage (%)
HBsAg		
Positive	2	2.7
Negative	71	97.3
Anti-HCV		
Positive	11	15.1
Negative	62	84.9

Table III showed the prevalence of hepatitis B and hepatitis C virus infection in the study population. It was observed that 2(2.7%) patients had positive

HBsAg, whereas 11(15.1%) patients had positive Anti-HCV antibody.

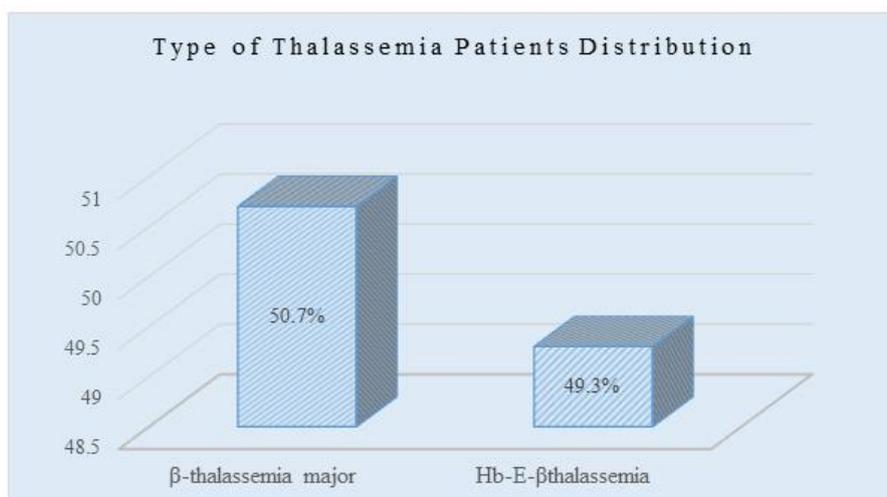


Figure 3: Bar chart showed type of thalassemia of Patients (N=73)

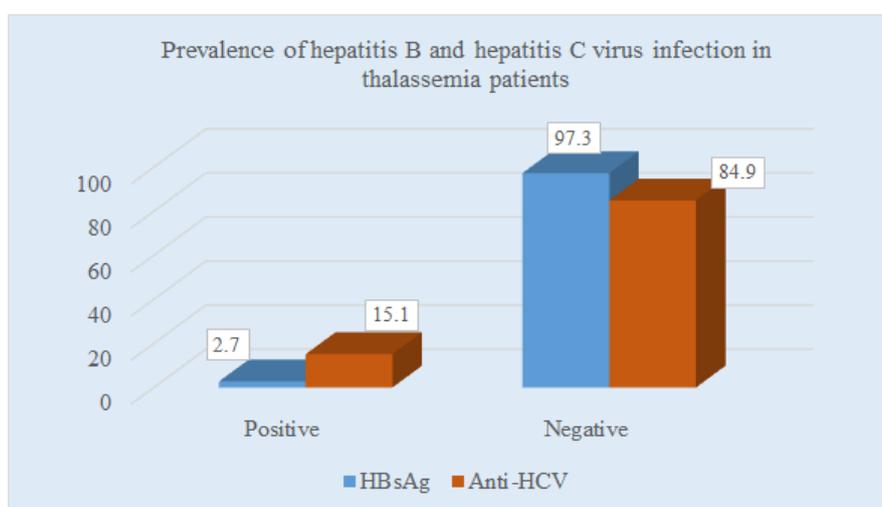


Figure 4: Bar chart showed prevalence of hepatitis B and hepatitis C virus infection (N=73)

Table IV: Risk factors for HBV infection (N=73)

Variables	HBV		P-value
	Positive (n=2)	Negative (n=71)	
Age (years)	14.75 $\pm$ 3.89	8.05 $\pm$ 3.28	0.006*
Gender			
Male	2(100.0%)	42(59.2%)	
Female	0(0.0%)	29(40.8%)	
Type of thalassemia			
B-thalassemia major	1(50.0%)	36(50.7%)	0.984
HB-E- $\beta$ thalassemia	1(50.0%)	35(49.3%)	
Age of diagnosis (months)	13.50 $\pm$ 2.12	17.13 $\pm$ 6.81	0.457
Duration of transfusion (years)	13.63 $\pm$ 3.71	6.67 $\pm$ 3.38	0.006*
Frequency of transfusion/years	12.0 $\pm$ 0.0	9.15 $\pm$ 3.03	0.192
Type of donor			
Voluntary	2(100.0%)	44(62.0%)	0.272
Professional	0(0.0%)	27(38.0%)	
Immunization status (against HBV)			
Immunized	0(0.0%)	70(98.6%)	<0.001*
Not immunized	2(100.0%)	1(1.4%)	
SGPT (U/L)	80.50 $\pm$ 7.78	55.21 $\pm$ 19.01	0.066

Table IV showed that mean age of HBsAg positive patients was  $14.75 \pm 3.89$  years and HBsAg negative patients was  $8.05 \pm 3.28$  years. Mean age was statistically significant ( $P=0.006$ ). Mean total duration was  $13.63 \pm 3.71$  years in HBsAg positive and  $6.67 \pm 3.38$

years in HBsAg negative patients which was also statistically significant ( $P=0.006$ ). 2(100%) patients were not immunized against hepatitis B in HBsAg positive group and only 1(1.4%) in HBsAg negative group ( $P<0.001$ ).

**Table V: Risk factors for HCV infection (N=73)**

Variables	HCV		p-value
	Positive (n=11)	Negative (n=62)	
Age (years)	$9.97 \pm 3.28$	$7.92 \pm 3.41$	0.068
Gender			0.360
Male	8(72.7%)	36(58.1%)	
Female	3(27.3%)	26(41.9%)	
Type of thalassemia			0.351
$\beta$ -thalassemia major	7(63.6%)	30(48.4%)	
Hb-E- $\beta$ thalassemia	4(36.4%)	32(51.6%)	
Age of diagnosis (months)	$13.55 \pm 3.05$	$17.64 \pm 6.75$	0.063
Duration of transfusion (years)	$8.85 \pm 3.36$	$6.50 \pm 3.50$	0.043*
Frequency of transfusion/years	$12.55 \pm 4.39$	$8.65 \pm 2.31$	<0.001*
Type of donor			0.161
Voluntary	9(81.8%)	37(59.7%)	
Professional	2(18.2%)	25(40.3%)	
SGPT (U/L)	$81.45 \pm 14.15$	$51.37 \pm 16.27$	<0.001*

Table V showed mean total duration of transfusion was  $8.85 \pm 3.36$  years in Anti-HCV positive and  $6.50 \pm 3.50$  years in Anti-HCV negative patients which was statistically significant ( $P=0.043$ ). Mean frequency of transfusion was  $12.55 \pm 4.39$  per year in

Anti-HCV positive and  $8.65 \pm 2.31$  per year in Anti-HCV negative group ( $P<0.001$ ). Mean SGPT was also statistically significant ( $P<0.001$ ) which was  $81.45 \pm 14.15$  U/L in Anti-HCV positive and  $51.37 \pm 16.27$  U/L in Anti-HCV negative patients.

**Table VI: Comparison of protective response of Anti-HBs titer according to age (N=70)**

Age group (years)	Anti-HBs titer			p-value
	No protection (n=13)	Moderate protection (n=48)	Strong protection (n=9)	
$\leq 5$ yrs.	0(0.0%)	5(10.4%)	8(88.9%)	<0.001*
6-10 yrs.	4(30.8%)	35(72.9%)	1(11.1%)	
11-15 yrs.	8(61.5%)	7(14.6%)	0(0.0%)	
>15 yrs.	1(7.7%)	1(2.1%)	0(0.0%)	
Mean $\pm$ SD	$11.42 \pm 2.62$	$7.61 \pm 2.58$	$4.51 \pm 1.07$	

Table VI showed patients in the significant immunity group had mean age of  $4.51 \pm 1.07$  year, in weak immunity group it was  $7.61 \pm 2.58$  year, and in no immunity group patients' mean age was  $11.42 \pm 2.62$  which was statistically significant ( $P<0.001$ ).

## DISCUSSION

In this study majority (54.8%) of patients belonged to age 6-10 years. The mean age was  $8.3 \pm 3.45$  years ranging from 3 years to 17 years 6 months. The number of patients had decreased in the older age group. With increasing age, mortality and morbidity also increases in these patients, which might cause the decreased number of attending patients. Almost two-third (60.3%) patients were male with male:female ratio 1.5:1. Although both male and females are equally affected in thalassemia, the present study found that

male patients are more in number than female. It might be occurred due to gender bias among the parents of these children who seek medical care for their male children. Mittal *et al.*, (2017) from India also had similar findings in their study [13]. In the present study, it is observed that 10(13.7%) patients had H/O consanguinity and 15(20.5%) had family H/O thalassemia. Among this 15 patients having positive family history, 13 patients have parents with either  $\beta$ -thalassemia trait or HbE trait, and 2 patients have sibling with  $\beta$ -thalassemia major. HbE- $\beta$  Thalassemia is the commonest form of thalassemia in southeast Asia. But we had 37(50.3%) patients with  $\beta$ -thalassemia major and 36(49.7%) with HbE- $\beta$  thalassemia as the study population was selected from the transfusion dependant patients of a selected thalassemia center. In this study, the mean age of diagnosis was  $17.03 \pm 6.75$  months ranging from 8 months to 48 months. Mean

total duration of transfusion was  $6.86 \pm 3.56$  years and mean frequency of transfusion was  $9.23 \pm 3.03$  per year. Mean hemoglobin level was  $6.53 \pm 0.64$  gm/dl. 2(2.7%) patients had H/O splenectomy and 36(49.3%) needed iron chelation therapy. In a study by Mahmoud *et al.*, (2016) revealed that among total 97 thalassemic children, 63.92% patients were male and 36.08% were female. The mean age at the time of study was  $8.89 \pm 5.07$  years (range 6–18 years), mean age at diagnosis was  $11.40 \pm 13.18$  month with mean duration of the disease was  $7.94 \pm 4.90$  years. Frequency of blood transfusion was  $1.09 \pm 0.42$  with pre-transfusion hemoglobin level was  $6.77 \pm 1.87$  g/dl. 41(42.27%) patients had H/O splenectomy which was much higher than the present study. 46(63.0%) patients received blood from voluntary donor and 27(37.0) patients from professional donor. 18 (24.7) patients received transfusion in only 1 authorized center, while 55(75.3%) used multiple centers for transfusion. In the present study, 95.9% patients were found to be immunized against hepatitis B. In 2003 hepatitis B vaccination was integrated in the EPI schedule and expanded to all districts by 2005. In another study done in out-patient department of Pediatric Hematology & Oncology department of BSMMU and thalassemia center of Bangladesh Institute of Child Health, Karim *et al.*, (2013) found that 77% thalassemic children were vaccinated against Hepatitis B virus [5]. The increased rate of hepatitis B vaccination found in the present study indicates that we have a successful EPI program in our country. In this study, 2(2.7%) patients had positive HBsAg. A study done in 2003 by Mollah *et al.*, (2003) revealed that, 13.8% was HBsAg positive among multitransfused thalassemia patients. Mollah *et al.*, (2003) found higher rate of HBsAg positivity probably due to the fact that their study was taken place before introduction of hepatitis B vaccine in the national EPI schedule in 2003 [9]. Karim *et al.*, (2013) found prevalence of hepatitis B was 3%, and Chattopadhyay *et al.*, (2014) from West Bengal found 3.69% [5, 14]. However, the small number of hepatitis B positive cases in the present study can also be attributed to the availability of the vaccine against HBV and increasing rate of public awareness about vaccination. A total of 11(15.1%) patients were found to be positive for Anti-HCV antibody in this study. 12.5% patients were reported positive for anti-HCV by Mollah *et al.*, (2003) [9]. There, among the HBsAg or anti-HCV positive patients (n=40), 5(12.5%) had both HBV and HCV infection. But in this study, we found no patient with both HBV and HCV co-infection. Karim *et al.*, (2013) found high prevalence of hepatitis C (31%) among thalassemia patients. [15] In a study done in West Bengal by Chattopadhyay *et al.*, (2014) revealed that 8.97% patient was anti-HCV positive where 3(0.79%) patients showed seropositivity for both HBsAg and anti-HCV [14]. Mukherjee *et al.*, (2017) found that anti-HCV antibody seropositivity was 24.6% [4]. In 2018, Hossain B *et al.*, found overall prevalence of hepatitis C was 14.7% where 47 patients were

positive for anti-HCV among 320 thalassemia cases [16]. Yasmeeen and Hasnain (2019) observed that prevalence of hepatitis C was 29.4% [17]. The higher prevalence of hepatitis C is likely due to lack of adequate high quality testing kits used in screening methods and collection of blood during window period. Another contributing factor for high prevalence of hepatitis C compared to hepatitis B might be the non-availability of vaccine against HCV. In assessing the risk factors of HBV infection, we found that, mean age of HBsAg positive patients was  $14.75 \pm 3.89$  years and HBsAg negative patients was  $8.05 \pm 3.28$  years which was statistically significant ( $P < 0.05$ ). Mean total duration of transfusion was  $13.63 \pm 3.71$  years in HBsAg positive and  $6.67 \pm 3.38$  years in HBsAg negative patients which was also statistically significant ( $P < 0.05$ ). 2(100%) of HBsAg positive patients were not immunized against hepatitis B virus, whereas only 1(1.4%) in HBsAg negative group had no H/O immunization against hepatitis B ( $P < 0.001$ ). It denotes that, non-immunized individuals are at a significant risk of acquiring hepatitis B virus infection. In the present study, mean total duration of transfusion was  $8.85 \pm 3.36$  years in Anti-HCV positive and  $6.50 \pm 3.50$  years in Anti-HCV negative patients which was statistically significant ( $P < 0.05$ ). Mean frequency of transfusion was  $12.55 \pm 4.39$  per year in Anti-HCV positive and  $8.65 \pm 2.31$  per year in Anti-HCV negative group ( $P < 0.001$ ). Hepatitis C virus mainly transmitted through transfusion of blood and blood products, and there is no vaccine or post-exposure prophylaxis. So, chance of acquiring hepatitis C is more than hepatitis B through repeated transfusion. Mahmoud *et al.*, (2016) found that risk factors for hepatitis C virus include age of the patients ( $P = 0.04$ ), duration of the disease ( $P = 0.02$ ), serum SGPT ( $P = 0.001$ ) which is similar to this study [18]. Hussain and Jaber (2018) found that, the prevalence of hepatitis C was significantly higher among older age group ( $P = 0.001$ ) with longer duration ( $P = 0.003$ ) and higher frequency of transfusion ( $P = 0.001$ ) which was also similar to the current study [19]. In the present study, mean SGPT was found statistically significant ( $P < 0.001$ ) among the Anti-HCV positive and Anti-HCV negative group which was  $81.45 \pm 14.15$  U/L and  $51.37 \pm 16.27$  U/L respectively. Here, significant increase in SGPT level in HCV positive group might not indicate a potential risk factor, rather it might indicate the hepatic inflammation due to viral hepatitis. Karim *et al.*, (2013) found significant difference in source of blood ( $P = 0.035$ ) and screening of blood before transfusion ( $P = 0.028$ ) among the seropositive and seronegative cases [13]. In the present study, all the patients receive blood after screening and no significant difference was found regarding donor criteria. It might be due to implementation of mandatory screening of donor blood before transfusion in the Safe Blood Transfusion Program. On the other hand, screening of HBsAg is the commonly used method to estimate the prevalence of hepatitis B virus, which cannot be detected during the window period.

For this, detection of antibody to hepatitis B core antigen (anti-HBc) may serve as a useful serological test during window period. In the current study, 70(95.89%) thalassemia patients received primary immunization against hepatitis B as per national EPI schedule. Among them, 9(12.9%) patients had strong protection, 48(68.6%) had moderate protection and 13(18.6%) had no protection against hepatitis B. Patients with strong protection group had mean age of  $4.51 \pm 1.07$  year, while moderate protection group had mean age of  $7.61 \pm 2.58$  year and no protection group had mean age of  $11.42 \pm 2.62$  year. Comparison of Anti-HBs titer with the mean age of that group showed that level of immunity against hepatitis B had decreased with increasing patients age and it was statistically significant ( $P < 0.001$ ). In a study done in Bangladesh, Hossain MM *et al.*, (2018) found that among 500 healthy participants 46.0% had titer below protection level, 36.4% had moderate protection and only 17.6% had strong protection [19]. In a study by Alssamei *et al.*, (2017), 72.2% children showed anti-HBs protective level of  $\geq 10$  IU/L, while 27.8% showed nonprotective anti-HBs titer levels of  $< 10$  IU/L and 88.9% protective rate in children less than one year and 55.4% in the older age group which is similar to this study [21]. These findings were also supported by a systemic review and meta-analysis of 12 studies reported by Mahmood *et al.*, (2018) [22]. However, in the present study, patients in the no protection group had higher mean age. The fact should be addressed that despite 13(18.6%) patients having no seroprotection against hepatitis B after primary immunization, none of these patients have been found positive for HBsAg. It signifies that vaccination against hepatitis B induces both cellular and immunological memory which stimulates the antibody production after any exposure, and age might not be a risk factor for acquiring hepatitis B virus in vaccinated children, rather lack of vaccination might cause hepatitis B infection.

## LIMITATIONS

- Anti-HBc was not done in this study which could identify more hepatitis B virus infection during window period.
- Non-transfusion related risk factors like surgical operation, dental procedure, ear, nose piercing were not addressed in this study as it was not planned initially.

## RECOMMENDATIONS

Steps should be taken to strengthen the screening procedure by implementing high quality methods like ELISA or introduction of more specific serological test (e.g. Anti-HBc) in screening program to address the donor window period to prevent these infections. Other than repeated transfusions, contributing factors like intravenous injections, dental procedures, surgical operations and lifestyle related

factors (e.g. drug abuse, ear, nose piercing, sharing razors) should be searched. Increased awareness about compulsory primary immunization should be done to prevent hepatitis B virus infection.

## CONCLUSION

Prevalence of hepatitis B is 2.7% and hepatitis C virus infection is 15.1% in repeatedly transfused thalassemia patients in a tertiary care hospital (Bangladesh Shishu Hospital & Institute). Older age group with increased duration and frequency of transfusion are associated with increasing hepatitis B and hepatitis C virus infections.

## REFERENCES

1. Mansour, A. K., Aly, R. M., Abdelrazek, S. Y., Elghannam, D. M., Abdelaziz, S. M., Shahine, D. A., ... & Darwish, A. M. (2012). Prevalence of HBV and HCV infection among multi-transfused Egyptian thalassemic patients. *Hematology/oncology and stem cell therapy*, 5(1), 54-59.
2. Mukherjee, K., Bhattacharjee, D., & Chakraborti, G. (2017). Prevalence of hepatitis B and hepatitis C virus infection in repeatedly transfused thalassemics in a tertiary care hospital in eastern India. *Int J Res Med Sci*, 5(10), 4558-4562.
3. Hossain, M. S., Raheem, E., Sultana, T. A., Ferdous, S., Nahar, N., Islam, S., ... & Morshed, M. (2017). Thalassemias in South Asia: clinical lessons learnt from Bangladesh. *Orphanet journal of rare diseases*, 12(1), 1-9. DOI 10.1186/s13023-017-0643-z.
4. Mukherjee, K., Bhattacharjee, D., & Chakraborti, G. (2017). Prevalence of hepatitis B and hepatitis C virus infection in repeatedly transfused thalassemics in a tertiary care hospital in eastern India. *Int J Res Med Sci*, 5(10), 4558-4562.
5. Karim, A. R., Islam, A., Jamal, C. Y., Matin, A., Hossain, M. M., & Shafiullah, M. (2013). 'Seroprevalence of hepatitis B, hepatitis C and human immunodeficiency virus among multitransfused thalassemic children in Dhaka, Bangladesh. *Bangladesh Journal of Child Health*, 37(3), 146-153.
6. Sheikh, N., Sultana, M., Ali, N., Akram, R., Mahumud, R. A., Asaduzzaman, M., & Sarker, A. R. (2018). Coverage, timelines, and determinants of incomplete immunization in Bangladesh. *Tropical medicine and infectious disease*, 3(3), 72. DOI: 10.3390/tropicalmed3030072.
7. Haque, A. E., & Latif, H. Z. (2015). Prevalence of hepatitis B and hepatitis C infections in multi-transfused thalassemic patients, *PTB Reports*, 1(2). DOI: 10.5530/PTB.1.2.6.
8. Alavian, S. M., Adibi, P., & Zali, M. R. (2005). Hepatitis C virus in Iran: Epidemiology of an

- emerging infection. *Archives of Iranian Medicine*, 8, 84-90.
9. Mollah, A. H., Nahar, N., Siddique, M. A., Anwar, K. S., Hassan, T., & Azam, M. G. (2003). Common transfusion-transmitted infectious agents among thalassaemic children in Bangladesh. *Journal of Health, Population and Nutrition*, 67-71.
  10. National Guidelines on Transfusion Transmissible Infections: Testing at Blood Transfusion Centers, 2013, External Quality Assessment Report, WHO, Bangladesh.
  11. Hossain, B., Khan, W. A., Tawfique, M., & Rahman, F. (2018). Prevalence of hepatitis C virus infection in multi-transfused Thalassaemia patients in Bangladesh. *Journal of Enam Medical College*, 8(1), 16-19.
  12. Mahmud, S., Hussain, M., Ahmed, S. S., Rahman, M., Tasneem, F., & Afroz, M. (2017). Burden of HCV in Bangladesh: warrants the screening of blood donors. *Acad J Ped Neonatol*, 4(5), 555706.
  13. Mittal, K., Abrol, P., & Yadav, J. (2017). Prevalence of transfusion transmitted infections amongst multiple blood transfused patients of  $\beta$ -thalassemia major in a tertiary care hospital. *Int J Res Med Sci*, 5(1), 181-185.
  14. Chattopadhyay, S., Mukherjee, R., Nandi, A., Chakraborty, P. S., Rit, K., & Chaudhuri, S. J. (2014). Prevalence of hepatitis B&C in thalassemia patients in a tertiary care hospital in West Bengal. *IOSR-JDMS*, 13(7), 68-70.
  15. Karim, A. R., Islam, A., Jamal, C. Y., Matin, A., Hossain, M. M., Shafiullah, M. (2013). Seroprevalence of hepatitis B, hepatitis C and human immunodeficiency virus among multitransfused thalassaemic children in Dhaka, Bangladesh. *Bangladesh Journal of Child Health*, 37(3), 146-153.
  16. Hossain, B., Khan, W. A., Tawfique, M., & Rahman, F. (2018). Prevalence of hepatitis C virus infection in multi-transfused Thalassaemia patients in Bangladesh. *Journal of Enam Medical College*, 8(1), 16-19.
  17. Yasmeen, H., & Hasnain, S. (2019). Epidemiology and risk factors of transfusion transmitted infections in thalassemia major: a multicenter study in Pakistan. *Hematology, transfusion and cell therapy*, 41, 316-323. <http://doi.org/10.1016/j.htct.2019.03.008>.
  18. Mahmoud, R. A., El-Mazary, A. A. M., & Khodeary, A. (2016). Seroprevalence of hepatitis C, hepatitis B, cytomegalovirus, and human immunodeficiency viruses in multitransfused thalassaemic children in upper Egypt. *Advances in hematology*, 2016. <http://dx.doi.org/10.11552/2016/9032627>.
  19. Ali, Z. H. (2018). Prevalence and risk factors for hepatitis C virus in Beta thalassaemic patients attending blood diseases center in Ibn-AL-Baladi Hospital, Baghdad. *Al-Kindy College Medical Journal*, 14(1), 42-49.
  20. Hossain, M. M., Alam, A. N., Siddiqua, M., Siddika, A., & Nessa, A. (2018). Immune response among the children to hepatitis B vaccination: A community-based study in Bangladesh. *Bangladesh Medical Research Council Bulletin*, 44(2), 103-108.
  21. Alssamei, F. A., Al-Sonboli, N. A., Alkumaim, F. A., Alsayaad, N. S., Al-Ahdal, M. S., Higazi, T. B., & Elagib, A. A. (2017). Assessment of immunization to hepatitis B vaccine among children under five years in rural areas of Taiz, Yemen. *Hepatitis research and treatment*, 2017. <https://doi.org/10.1155/2017/2131627>.
  22. Mahmood, S., Shah, K. U., & Khan, T. M. (2018). Immune persistence after infant hepatitis-B vaccination: a systematic review and meta-analysis. *Scientific reports*, 8(1), 1-8. DOI: 10.1038/s41598-018-30512-8.