Prevalence of Hepatitis B and C Viruses in Alfa-Thalassemia Major and Beta-Thalassemia Major and Intermedia in Duhok City, Iraq

Original Article

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ABSTRACT

Introduction and Objectives: Thalassemia is characterized by a hereditary reduction in hemoglobin production, either partial or total failure of globin chain synthesis.

End-stage liver disease has been linked to hepatitis B and hepatitis C. Present study aims to estimate the prevalence of Hepatitis B, and Hepatitis C viruses in Alpha major, Beta thalassemia major, and intermedia patients, and its relation with type of thalassemia, age group and blood transfusion in Duhok city.

Materials and Methods: A cross-sectional study was conducted on 200 multi-transfused thalassemia patients. Data required in the study had been collected from patients, their guardians (parents), and from the records, then 3-5 ml of each participant blood was withdrawn for Hepatitis C virus antibodies, and Hepatitis B surface antigen by ELISA, and PCR done for detection of these viruses.

Results: A total of 200 patients were included in the study. Out of which 93(46.5%) male, and 107(53.5%) female. The mean number of blood transfusions per year is 26.92 +17.1 SD. Among a total of 200 patients, 37(18.5%) were hepatitis C positive and their mean number of blood transfusions received was 36.21+13.108 SD. In total 1 (0.5%) patients were hepatitis B surface antigen positive. HCV infection in Beta thalassemia major was diagnosed earlier than other types of thalassemia with $p < 0.05$.

Conclusions: The study revealed that the prevalence of HCV infection is higher in multi-transfused thalassemia patents than in the general population. The risk of having HBV and HCV infection increases with the progression of age specially HCV. There was a highly significant association in the number of blood transfusions across different thalassemia diagnoses and HCV status.

Key Words: ELISA, HCV, HBS, PCR, thalassemia.

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INTRODUCTION

Thalassemia refers to a group of genetic disorders of globin chain production in which there is an imbalance between the α-globin and β-globin chain production[1].

Globally, thalassemia is the most common blood hereditary disease, and occurs in 4.4/10,000 live births. In the developing world, the majority of patients die before the age of 20 years. In Iraq, there is little data on the prevalence and epidemiology of thalassemia[2]. The exact genetic mutation causing the illness and its effect on the globin-chain components of the hemoglobin tetramer determine how the disorder is classified[2]. A genetic blood condition called alpha thalassemia is mainly caused by deletions in the α-globin gene and, less often, by point mutations[3]. Within the hemoglobin tetramer, these mutations result in either decreased or absent α-globin chains[3]. A thorough worldwide evaluation conducted in 2013 classified the Syrian Arab Republic as having a low prevalence of HBV and a high prevalence of HCV[4].

The cause of beta-thalassemias is point mutations or, less frequently, deletions in the beta globin gene on chromosome 11. This leads to reduced synthesis of the Hb beta chains (beta+) or no synthesis at all (beta0). A group of inherited blood illnesses known as beta thalassemias

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(β thalassemias) are typified by either decreased or nonexistent beta chain synthesis of hemoglobin[1]. The delta globin gene, the embryonic epsilon gene, the fetal A-gamma and G-gamma genes, and a pseudogene (ψB1) are all located in the same area on the short arm of chromosome 11 as the beta globin (HBB) gene[5].

As a major global public health concern, hepatitis B is caused by the hepatotropic virus HBV. According to[6], it can cause acute and chronic infections that can be fatal and may eventually lead to cirrhosis and HCC. The main cause of long-term liver diseases that frequently result in cirrhosis and hepatocellular carcinoma (HCC) is HCV[7,8]. General transfusion guidelines recommend initiating transfusions at a Hb threshold of 60–100 g/L, depending on the presence and severity of comorbidity[9].

Even though these patients have a higher risk of blood-borne infections, regular blood transfusions are essential for prolonging their lives[10]. Hepatitis B immunoglobulin (HBIG) must be administered and vaccinated against the virus as soon as possible after exposure, but no later than 24 hours later[11]. Regretfully, there isn't a vaccine against HCV at this time[12].

MATERIALS AND METHODS

Study area

A cross-sectional study was conducted on 200 multi-transfused thalassemia patients attends for 1 month from February to March 2024 in Jeen Center for Hematology and Children Cancer, Duhok City, Iraq.

Sample collection and processing

Data required in the study had been collected from patients, their guardians (parents), and from the records after obtaining the approval of the ethics committee from the General Duhok Directorate of Health (13122023-11-16). Their demographic characteristics including age, gender, number of blood transfusions required per year, and their vaccination status regarding HBV were collectedand entered on a preformed questionnaire. About 3-5ml of venous blood was drawn and collected in a serum tube under aseptic measures. A Collected blood sample was sent to the Jeen Center for Hematology and Children Cancer, Duhok City, Iraq, laboratory for evaluation of HBsAg and anti-HCV Antibodies by third generation ELISA, confirmation by PCR was done in patients in whom HBV or HCV was positive by ELISA.

Statistical analysis

All the data was entered and analyzed in Statistical Package for Social Sciences version 16.0. to explore the associations between infections and age, gender and number of blood transfusions, the ANOVA table and Bar chart test were conducted.

RESULTS

Two-hundred thalassemia patients' records from the Jeen Center for Hematology and Children Cancer, Duhok City, Iraq were studied. The patients diagnosed during 1996 – 2024.

Gender and group distribution

One hundred seven patients 107 (53.5%) were females and 93 (46.5%) male, a slightly higher representation of females among the thalassemia patients in the clinic, 64 (32.0%) patients were < 15 Years, this breakdown illustrates that significant majority of the patients (68%) are aged 15 years or older, Table.1 provide valuable insights into the gender and age distribution of thalassemia patients at the clinic, indicating a relatively even distribution between genders, but a notable predominance of patients aged 15 years and above as shown in (Table 1, Figure 1)

Table 1: Gender and age group distribution.

Variables		Frequency	Percent
Gender	Female	107	53.5
	Male	93	46.5
Age Group	< 15 Years	64	32.0
	>= 15 Years	136	68.0
Total		200	100.0

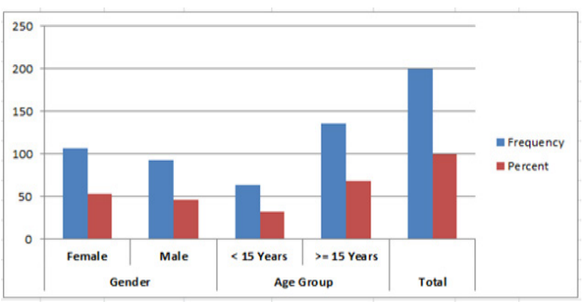

Fig. 1: Sex and age distribution

Frequency and percentages of thalassemia types and HBV, HCV

An overall of diagnoses and the presence of (HBV) and (HCV) among thalassemia patients, and types of thalassemia at the Jeen Specialized Center in Duhok governorate. Alpha thalassemia patients were 32 (16.0%), 77 (38.5%) Beta thalassemia intermedia, and 91(45.5%) Beta thalassemia major. Also revealed (HBV) composed only one patient 1 (0.5%) was diagnosed with HBs antigen positive, and 199 (99.5%) were negative indicating a low prevalence of this virus among thalassemia patients at the center. While (HCV) antibodies, 37 (18.5%) were positive, and 163 (81.5%) were negative. The presence of HCV is more common as shown in (Table 2, Figure 2)

Table 2: Thalassemia diagnosis, HBV, and HCV prevalence.

Variable		Frequency	Percent %
Diagnosis	Alpha thalassemia major	32	16.0
	Beta thalassemia intermedia	77	38.5
	Beta thalassemia major	91	45.5
	Total	200	100
HBV	Positive	1	0.5
	Negative	199	99.5
	Total	200	100
HCV	Positive	37	18.5
	Negative	163	81.5
	Total	200	100.0

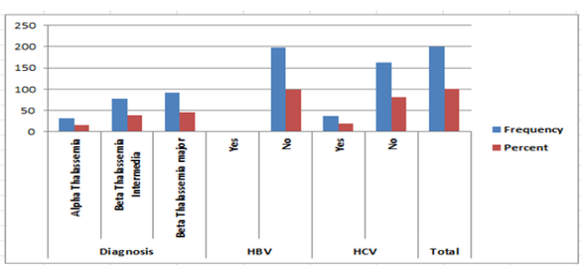

Fig. 2: Type of thalassemia and HCV, HBS distribution

Descriptive study of the present age, year of diagnosis and number of blood transfusions

The average age of the patients is approximately (19.79 ±10.734) years, with a considerable spread of ages around this mean. On average, patients have received approximately 26.92 blood transfusions, with notable variability in the number of transfusions received among the patients. A standard deviation of 6.491 years, suggests some variability in the timing of diagnosis among the patients, with years since diagnosis of (13.31± SD 6.491) years from the time of diagnosing the disease (Table 3).

Table 3: Descriptive statistics of present age, blood transfusion, and years since diagnosis.

Variable	Mean and Std. Deviation
Present age (years)	19.79 ±10.734
Years since Diagnosis (years)	13.31±6.491
No. Blood transfusion	26.92± 17.100

Association between thalassemia diagnosis and gender

The distribution of thalassemia diagnoses is based on gender and provides a chi- square test for independence to examine whether there is a significant association between diagnosis and gender.

The proportion of females diagnosed with Alpha Thalassemia (43.8%) is slightly lower than males (56.3%), but this difference is not statistically significant ($P > 0.05$).

A higher proportion of females (59.7%) are diagnosed with Beta thalassemia intermedia compared to males (40.3%), but again, this difference is not statistically significant ($P > 0.05$). Meanwhile, for Beta thalassemia major, females have a slightly higher proportion of diagnosis (51.6%) compared to males (48.4%), but like the other diagnoses, this difference is not statistically significant ($P > 0.05$) as shown in (Table 4, Figure 3).

Table 4: Comparing the distribution of thalassemia diagnoses based on gender

Diagnosis	Gender		Total, n (%)	Chi-square	Df	Sig. p < 0.05
	Female,	Male,				
	n (%)	n (%)				
Alpha Thalassemia	14 (43.8)	18 (56.3)	32 (16)	3	2	0.279
Beta Thalassemia Intermedia	46 (59.7)	31 (40.3)	77 (38.5)			
Beta Thalassemia major	47 (51.6)	44 (48.4)	91 (45.5)			
Total	107 (53.5)	93 (46.5)	200 (100.0)			

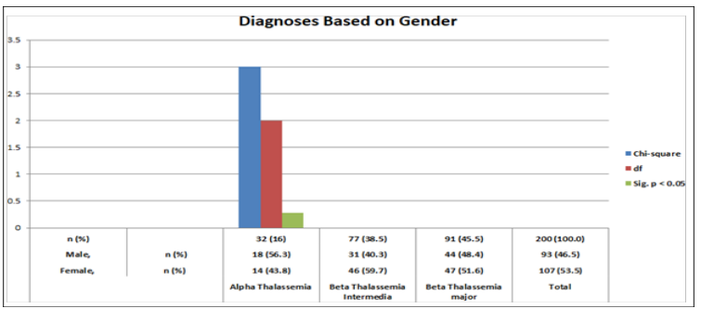

Fig. 3: Illustrates the distribution of thalassemia diagnoses across gender groups

Association between thalassemia diagnosis and age groups

The proportion of patients diagnosed with alpha thalassemia differs slightly between age groups. Among those aged < 15 years, 25.0% are diagnosed with alpha thalassemia, while among those aged >= 15 years, 75.0% have this diagnosis. However, the chi-square test indicates the difference is not statistically significant ($P > 0.05$).

The majority of patients diagnosed with beta thalassemia intermedia are aged >= 15 years (63.6%) compared to those aged < 15 years (36.4%). Beta thalassemia major is similar to beta thalassemia intermedia, a higher proportion of patients diagnosed with beta thalassemia major are aged >= 15 years (69.2%) compared to those aged < 15 years (30.8%) as shown in (Table 5, Figure 4).

Table 5: Comparing the distribution of thalassemia diagnoses across different age groups

Diagnosis	Age Group		Total, n(%)	Chi- square	Df	Sig. $p < 0.05$
	< 15 Years, n (%)	> = 15 Years, n (%)				
Alpha Thalassemia	8 (25.0)	24 (75.0)	32 (16)			
Beta Thalassemia Intermediate	28 (36.4)	49 (63.6)	77 (38.5)	1.458	2	0.482
Beta Thalassemia major	28 (30.8)	63 (69.2)	91 (45.5)			
Total	64 (32.0)	136 (68.0)	200(100.0)			

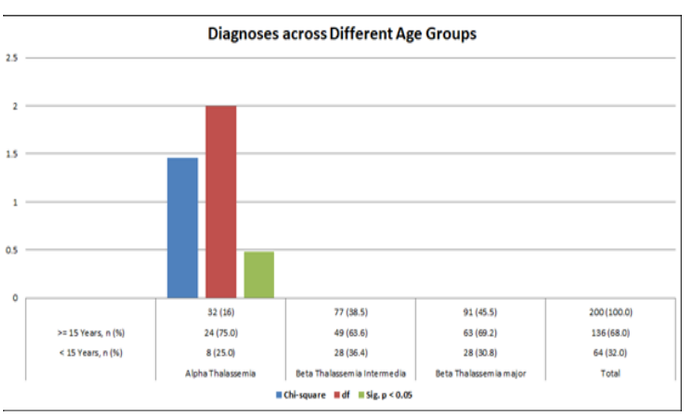

Fig. 4: Illustrates the distribution of thalassemia diagnoses across age group

Association of present age of thalassemia patients with HCV status

The average current age (Years) of thalassemia patients is categorized by their HCV status, along with the results of an ANOVA test examining the differences in age across different thalassemia diagnoses and HCV status. Patients diagnosed with Alpha thalassemia and HCV positive have an average age of (35.50 ±7.778) years. Those diagnosed with beta thalassemia intermedia and HCV positive have an average age of (33.50 ±3.536) years. Patients diagnosed with Beta thalassemia major and HCV positive had an average age of (23.09±6.429) years. Patients diagnosed with alpha thalassemia and HCV negative have an average age of (23.87±14.012) years. Those diagnosed with beta thalassemia intermedia and HCV negative have an average age of (19.24 ±11.452) years. Patients diagnosed with beta thalassemia major, and HCV negative have an average age of (15.48± 7.616) years as shown in (Table 6).

Table 6: The average current age (Years) of thalassemia patients categorized by their HCV Status

Average (SD) present age (Years)					ANOVA Test		
HCV		N	Mean	SD	F	df	Sig.
HCV (+ve)	Alpha thalassemia	2	35.50	7.778	8.422	1	.004
	Beta thalassemia Intermedia	2	33.50	3.536			
	Beta thalassemia major	33	23.09	6.429			
	Total	37	24.32	7.196			
HCV(-ve)	Alpha Thalassemia	30	23.87	14.012			
	Beta Thalassemia Intermedia	75	19.24	11.452			
	Beta Thalassemia Major	58	15.48	7.616			
	Total	163	18.75	11.147			
Total	Alpha thalassemia	32	24.59	13.921			
	Beta thalassemia Intermedia	77	19.61	11.536			
	Beta Thalassemia Major	91	18.24	8.060			
	Total	200	19.79	10.734			

Across all patients, regardless of HCV status, Alpha thalassemia: average age of (24.59±13.921) years, Beta Thalassemia Intermedia: The average age of (19.61±11.536) years, Beta thalassemia major: average age of (18.24 ±8.060) years.

The ANOVA test results indicate a significant difference in age across different thalassemia diagnoses and HCV status (F = 8.422, df = 1, p = 0.004). This suggests that both thalassemia diagnosis and HCV status contribute to the variation in the average age of thalassemia patients.

Association of blood transfusion and HCV status

The average number of blood transfusions received by thalassemia patients, categorized by their HCV status. It also includes the results of an ANOVA test examining the differences in the average number of blood transfusions across different thalassemia diagnoses and HCV status.

Patients diagnosed with beta thalassemia intermedia and HCV positive have an average of blood transfusions (3.00 ± 9.922), Beta thalassemia major (37.65±11.284). The overall average for HCV- positive patients is (36.21± 13.108) blood transfusions.

Patients diagnosed with Alpha thalassemia major, and HCV negative have an average of (1.25±0.50) blood transfusions, beta thalassemia intermedia (11.71±10.553) blood transfusions, and Beta thalassemia major(35.95±12.673) blood transfusions. The overall average for HCV-negative patients is (24.21±17.240) blood transfusions as shown in (Table 7).

Table 7: The average number of blood transfusions received by thalassemia patients, categorizedby their HCV status

Average Number of Blood Transfusions					ANOVA Table		
HCV		N	Mean	Std. Deviation	F	df	Sig.
HCV(+ve)	Beta thalassemia intermedia	1	3.00	9.922	9.922	1	.002
	Beta thalassemia major	23	37.65	11.284			
	Total	24	36.21	13.108			
HCV (-ve)	Alpha thalassemia	4	1.25	.500			
	Beta thalassemia intermedia	34	11.71	10.553			
	Beta thalassemia major	44	35.95	12.673			
	Total	82	24.21	17.240			
Total	Alpha Thalassemia	4	1.25	.500			
	Beta Thalassemia Intermedia	35	11.46	10.500			
	Beta Thalassemia major	67	36.54	12.155			
	Total	106	26.92	17.100			

Across all patients, regardless of HCV status, alpha thalassemia major had an average of (1.25±0.50), Beta thalassemia intermedia (11.46±10.500), and Beta thalassemia major had an average of (36.54±12.155) blood transfusions. The overall average for all patients is (26.92±17.100) blood transfusions.

The ANOVA test results indicate a significant difference in the average number of blood transfusions across different thalassemia diagnoses and HCV status ($F = 9.922$, $df = 1$, $p = 0.002$). This suggests that both thalassemia diagnosis and HCV status contribute to the variation in the average number of blood transfusions received by thalassemia patients.

DISCUSSION

This study was undertaken to assess the prevalence of HBV and HCV infections among thalassemia patients at the Jeen Center for Hematology and Children Cancer, Duhok City, Iraq.

Given that these patients require lifelong multi-transfusion therapy, they face a heightened risk of acquiring blood transfusion-transmitted infections such as HBV and HCV. These infections can be transmitted through various routes beyond blood transfusions, potentially leading to severe complications such as hepatocellular carcinoma or mortality. Beta thalassemia major patients were specifically targeted in this study due to their need for early-life blood transfusions, primarily within the first year of life. Therefore, any HBV or HCV infections detected in these patients would likely be attributed to transfusion of contaminated blood. The age distribution of thalassemia patients included in the study ranged from <15 to >15 years old, with approximately 32% of them being under 15 years old. This may be attributed to the higher mortality rate among thalassemia patients compared to the general population. Descriptive statistics for various factors among thalassemia patients revealed an average patient age of around 19.79 years, with considerable variability in ages. Both male and female patients exhibited similar average ages, although females displayed slightly higher age variability with unexplained cause. On average, patients received approximately 26.92 blood transfusions, with notable variability in the number of transfusions received. Patients were typically diagnosed around the year 2011, with some variability in the timing of diagnosis. The study also found that only one patient (0.5%) was diagnosed with HBV, and in Iran (0%) in which they didn't find any HBV positive cases[13], and India (0%)[14], indicating a low prevalence of this virus among thalassemia patients at the center, and the presence of vaccination program against this virus. In contrast, 37 patients (18.5%) tested positive for HCV which is near to study in Pakistan (18.3%)[2], and Egypt (20.7%)[15], making it more common among thalassemia patients, and may be due to absence of vaccine of this type of virus.

On the other hand, we didn't take thalassemia minor because our study was in a center where they usually received patients who need a blood transfusion, and as we know that this type of thalassemia is diagnosed accidentally or in our locality in the Marriage program they can be presented, also because they didn't require blood transfusion usually, so we canceled this type of thalassemia.

CONCLUSION

Prevalence of HCV infection is more in multi-transfused thalassemia patent than in the general population. Measures taken in the blood preparation and checking are still not enough to prevent infections transmitted by blood transfusion. The risk of having HBV and HCV infection in multi-transfused patients increases with the progression of age especially HCV. There was a highly significant association between the average number of blood transfusions across different thalassemia diagnoses and HCV status. There's a notable incidence of HCV among individuals with thalassemia, with blood transfusions representing a significant risk, therefore is recommended all thalassemia patients should be screened for HCV infection periodically.

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ABBREVIATIONS

ANOVA: Analysis of variance,

ELISA: Enzyme Linked Immune Assay,

HB: Hemoglobin,

HBB: Hemoglobin subunit Beta,

HBIG: Hepatitis B Immunoglobulin,

HBsAg: Hepatitis B surface antigen,

HBV: Hepatitis B virus,

HCC: Hepatocellular Carcinoma,

HCV: Hepatitis C virus,

PCR: Polymerase chain reaction,

RBCs: Red Blood Cells,

TDT: Transfusion Depended Thalassemia,

TM: Thalassemia Major,

TTIs: Transfusion Transmitted Infections.

CONFLICT OF INTERESTS

There are no conflicts of interest.

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انتشار فيروسات التهاب الكبد (بى) والتهاب الكبد (سى) في مرضى الثلاسيميا ألفا وبيتا ثلاسيميا الكبرى والمتوسط في مدينة دهوك، العراق

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الخلفية والاهداف: يعد مرض الثلاسيميا من أكثر الأمراض الوراثية انتشارا، ويتميز بانخفاض وراثي في إنتاج الهيموجلوبين، إما فشل جزئي أو كلي في تصنيع سلسلة الجلوبين.
تم ربط مرض الكبد في المرحلة النهائية بالتهاب الكبد بي والتهاب الكبد سي وتحدث الأمراض المنقولة عن طريق نقل الدم عن طريق كلا الفيروسين. الهدف هو تقدير مدى انتشار فيروس التهاب الكبد بي وفيروس التهاب الكبد سي في مرضى الثلاسيميا ألفا وبيتا الكبرى والوسطى، وعلاقته بنوع الثلاسيميا والفئة العمرية ونقل الدم في مدينة دهوك.
طرق العمل: دراسة مقطعية على ٢٠٠ من مرضى الثلاسيميا الذين تم نقلهم في مركز جين العام المتخصص للثلاسيميا في مدينة دهوك، إقليم كردستان، العراق. جمع البيانات المطلوبة في الدراسة من المرضى وأولياءهم و السجلات، ثم تم سحب ٣-٥ مل من دم كل مشارك للكشف عن الأجسام المضادة لفيروس التهاب الكبد الوبائي سي، ومستضد التهاب الكبد الوبائي السطحي بواسطة ELISA، PCR للكشف عن هذه الفيروسات.
النتائج: ٩٣ (٤٦,٥٪) من المرضى كانوا ذكورا و١٠٧ (٥٣,٥٪) إناث. وكان متوسط عدد عمليات نقل الدم سنويا ٢٦,٩٢+١٧,١ إس دي. من بين إجمالي ٢٠٠ مريض، كان ٣٧ (١٨,٥٪) مصابين بالتهاب الكبد الوبائي سي وكان متوسط عدد عمليات نقل الدم التي تم تلقيها ٣٦,٢١ + ١٣,١٠٨ SD. في المجموع، تبين أن ١ (٠,٥٪) من المرضى لديهم مستضد سطحي لالتهاب الكبد بي إيجابي. تم تشخيص الإصابة بفيروس التهاب الكبد سي في بيتا ثلاسيميا الكبرى في وقت أبكر من الأنواع الأخرى من الثلاسيميا مع $P < 0.05$.
الاستنتاجات: إن انتشار عدوى فيروس التهاب الكبد الوبائي (سي) يكون أكثر في مرضى الثلاسيميا المنقولين أكثر من عامة السكان. يزداد خطر الإصابة بعدوى فيروس التهاب الكبد الوبائي(بى) وفيروس التهاب الكبد الوبائي(سى) لدى المرضى الذين يخضعون لعمليات نقل دم متعددة مع تقدم العمر، وخاصة فيروس التهاب الكبد الوبائي (سي). كان هناك ارتباط كبير للغاية في متوسط عدد عمليات نقل الدم عبر تشخيصات الثلاسيميا المختلفة وحالة فيروس التهاب الكبد الوبائي (سي).