

The Prevalence of Hepatitis C Virus in Patients with Thalassemia Major and Hemodialysis in Yasuj, Iran

Abstract

Hepatitis C is a viral infection that causes liver cirrhosis and cancer in chronic instances. Blood and blood products are one of the most common routes for the illness to spread. Patients who get HCV through blood transfusions are patients who require frequent blood transfusions and blood products. As a result, the present study aims to investigate the incidence of the hepatitis C virus in major thalassemia patients and hemodialysis patients in Yasuj. After documenting personal information like as age, sex, education, marital status, and blood type, blood samples were obtained from 91 patients with thalassemia major and 40 hemodialysis patients sent to Shahid Beheshti Hospital in Yasouj. Following serum isolation, serum samples were analyzed using an ELISA kit to see if they were infected with the hepatitis C virus. Four individuals with thalassemia major were positive for HCV ELISA and were between the ages of 30 and 40. Hemodialysis patients did not have any positive instances. The majority of thalassemia major patients were self-employed. Three of the four hepatitis C patients (75%) were blood type O. Furthermore; these four individuals were all single. One HCV-positive person was illiterate, one had a high school diploma, and two had a bachelor's degree in terms of education. The findings show that there is no latent hepatitis C in hemodialysis patients in Yasouj and that there are four individuals with thalassemia major who have hepatitis C, which is consistent with previous research.

Keywords: *Hepatitis C, major Thalassemia, Hemodialysis patient, Blood products*

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Introduction

Hepatitis C is a viral infection that primarily affects the liver and is responsible for a significant number of chronic cases as well as complications such as liver cirrhosis and cancer. Blood and blood products are one of the most common routes for this disease to spread. However, blood transfusions are the most prevalent route for HCV infection to spread (Beltran M et al., 2005). HCV is the most frequent kind of hepatitis transmitted by blood transfusions, according to studies. Patients who get HCV through their blood must obtain blood and blood products regularly if they are infected with the virus. Patients with thalassemia and chronic renal failure who are on maintenance hemodialysis are among them (Hassan Shahi, 2008). In numerous studies in Iran and other areas of the world, these difficulties have resulted in a substantially greater frequency of HCV infection in thalassemia patients and dialysis patients (Zahedi, 2003).

People who frequently receive blood products, such as patients with hemophilia, hemodialysis, and thalassemia, as well as injecting drug users, transplant recipients, and people with multiple sexual partners, are all at high risk for hepatitis C. Babies born to mothers with hepatitis C are also at high risk (Fissell, 2003). Due to continual interaction with blood products, patients with thalassemia major who are on a regular transfusion program are at risk for hepatitis, particularly hepatitis C. According to research performed in various nations, 30% to 60% of thalassemia patients in

various regions of the world are infected with the hepatitis C virus. We are seeing a rise in the life expectancy of thalassemia patients today, thanks to increased attention and care for particular people, and one of the primary difficulties that these patients face in their later years is liver failure and viral hepatitis especially hepatitis C. (Mirmomen, 2006).

Due to repeated blood transfusions, kidney transplants, and the use of injectable medications, hemodialysis patients are at significant risk. Because liver illness might impact a patient's mortality rate, antiviral medication for dialysis patients requires specific consideration. Suppression of the immune system might hasten HCV-related liver damage, and interferon alfa can cause kidney transplant rejection (European, 2014).

Hepatitis C has been investigated further in Iran in specific sectors of society, with various examples. In a survey of 1721 persons between Kermanshah in 2005 and 2006, 0.87 percent of the population was infected with hepatitis C. (0.4 percent men and 0.3 percent women). The most prevalent genotype found in these people was genotyped I, with subtype Ia being the most common, which is comparable to Russia and Turkey (Sayad, 2008). In separate research in Yazd, 104 dentists were tested for hepatitis C, but none were found to be sick (Behnaz, 2008). Hemophilia sufferers have been the subject of several investigations. Hepatitis C was found in 6.15 percent of these individuals in Fars province and 76.7 percent in northern Iran. Hepatitis C was found in 24.2 percent of

patients with thalassemia in research performed in Tehran, whereas the frequency of hepatitis C in patients with thalassemia was reported to range from 15.7 percent to 63.8 percent in other studies conducted in Iran (Editorial 2007).

According to a 2020 study on the frequency and recurrence of hepatitis C in hemodialysis patients in the Egyptian province of Assiut, 34.8 percent of hemodialysis patients had HCV, and 13.2 percent had a cervical conversion. In addition, significant risk factors for blood transfusion include a positive history of blood transfusion, frequency of blood transfusion medical personnel relocation to blood equipment and products, and the number of temporary dialysis catheters (Kerollos Motwade N et al., 2020).

In a 2020 study of hepatitis C virus infection in patients undergoing hemodialysis at Shikhopura Hospital, researchers discovered that 60 (39.21 percent) of 153 hepatitis C-negative patients were positive when dialysis was started, while 93 patients (60.78 percent) were still HCV negative at the time of the ELISA study. The number of dialyzes was significant for p (Hussain Y et al., 2020).

According to a 2020 research of hepatitis C virus RNA estimations in -thalassemia patients in Ninawa Province, the prevalence of HCV in thalassemia patients in Ninawa Province was lower than in other Iraqi provinces or surrounding countries. In clinical thalassemia and dialysis centers, immunobiological and biochemical testing for liver enzymes should be performed regularly (Dawood, et al., 2020). According to a 2020 research titled "Eradication of Hepatitis C Virus Infection in Thalassemia Patients Using Different Therapies," HCV can be eradicated sooner than previously anticipated in this group of patients thanks to the introduction of DAA, which has a high success rate in therapy. According to prior estimates, there will be fewer than 66 thalassemia patients infected with HCV by 2020. The DAA, on the other hand, is expected to be less than ten (Moghimbeygi, M et al., 2020). In a study published in 2018, researchers discovered that the most prevalent genotypes of hepatitis C virus in individuals with -thalassemia major were a3 (0.50 percent) and a1 (37.5 percent). There was no correlation between genotype and age, sex, serum ferritin, liver test, or HCV-RNA titer (Ghezeldasht et al., 2018). Because no research on the prevalence of hepatitis C virus in persons with thalassemia major and hemodialysis patients has been done in Yasuj, this study is unique in this regard and can bring new information.

Materials and methods

By gaining consent and completing a questionnaire, samples were obtained from thalassemia and dialysis patients in Yasouj who were sent to Shahid Beheshti Hospital for blood transfusion. Blood samples were obtained from 91 Cubital or Cephalic veins of 91 major thalassemia patients, roughly 18

of whom were referred to Shahid Beheshti Hospital daily, and their serum was extracted in the laboratory by centrifugation. After separating the serum and numbering them, and recording the patients' details, they were maintained at 7 ° C until the experiment day, and after collecting the samples, they were analyzed for hepatitis C antibodies using the ELISA technique one week later. A total of 40 dialysis patients were present. Individuals admitted to Shahid Beheshti Hospital for hemodialysis on certain days were also sampled, as were all blood samples following serum isolation and numbering, as well as patient information notes, until the day of the experiment at a temperature of -7 degrees. One week after collecting the samples, ELISA was used to check for the existence of an anti-hepatitis C antibody.

Serums generated from blood samples of people are tested using the ELISA technique and a Danesh Bonyan Pishtaz Teb Company kit. The indirect enzyme immunoassay technique is used to create the test in this kit. HCV recombinant NS3, NS4, NS5, and CORE antigens are coated in plate wells. If there are specific antibodies against HCV (IgM-IgG) present following the addition of serum during the test, the antibodies are bound to these good antigens of the good floor through the Fab region. An ELISA reader capable of measuring light absorption at 450 nm was used to read the data.

1. An ELISA apparatus read the optical absorption of the controls and samples at 450 nm and, if feasible, against a 630 nm reference filter.

2. The cut-off was calculated using the formula below (Blanc optical absorption should be reduced from the optical absorption of all samples and controls).

Cut-off value = Mean light absorption of negative controls + 0.2

The S/Co index may also be used to evaluate if a response is positive or negative.

To obtain the index $S/Co = \frac{\text{Sample OD}}{\text{Cut-off value}}$ was used. All responses with S/Co of 1 or more than one were judged positive, while all answers with S/Co of less than one were considered negative, according to this formula.

- A negative result suggests the lack of antibodies against HCV antigens or the inability to detect antibodies against HCV antigens (in the early stages of infection).
- Positive responses were given again and again. Positive samples that recurred as negative were recorded as negative.

The following are the steps for calculating and reviewing the results:

First, the cut-off is calculated as follows:

- Cut-off value = mean light absorption of negative controls + 0.2
- Cut-off value=0.031+0.2=0.246

- Cut-off value= $0.017+0.2=0.217$

The findings of positive controls are as follows, depending on the calculations of each:

- Controlling the positives
- Index value= $0.411/0.217=1.89$
- Index value= $0.934/0.246=3.79$

We conclude that the tests were appropriately completed because the positive control values are more than 1.

The computations for the positive instances may now be shown in the table below.

- Index value= $0.228/0.217=1.1$

Table 1. The results obtained by ELISA method

Row	Patients	Number of samples	number of HCV positive	Relative frequency percentage (%)	number of HCV negative	Relative frequency percentage (%)
1	Thalassemia major	91	4	4.39	87	95.61
2	Hemodialysis	40	0	0	40	100

There were 47 females and 44 males among the 91 thalassemia samples and 17 females and 23 males among the 40 hemodialysis patients evaluated in this study. According to the findings, 47 (52%) of the 91 thalassemia major patients are women, and 44 (48%) are males. 17 (42%) of the 40 hemodialysis patients evaluated in this study were women, whereas 23 (58%) were males. Hepatitis C infection was detected in 4% of the female population (8.5 percent of the overall population of women with thalassemia) and 0% of the male population with thalassemia major. Men and women on hemodialysis were also found to be free of the infection.

Patients with thalassemia and those on hemodialysis were also assessed in terms of schooling. The findings revealed that out of 91 patients with thalassemia major, nine were illiterate (9.9%), 6 were primary literate (6.6%), 25 (27.5%) had cycle literacy, 40 (44%) had diploma literacy, and 11 (12%) had bachelor literacy. One (25%) of the four patients with hepatitis C was illiterate, one (25%) had a diploma, and two (50%) had a bachelor's degree. 15 (37.5%) of 40 hemodialysis patients were illiterate, 9 (22.5%) were cycle literate, 11 (27.5%) were diploma literate, 4 (10%) were bachelor literate, and 1 (2.5%) had postgraduate literacy; none (zero percent) of the 40 illiterate hemodialysis patients with primary education, middle, diploma, or postgraduate degree had hepatitis C.

Out of 91 thalassemia major patients, 4 (4.93 percent) were in the age group of 10-10 years, 21 (23.1 percent) in the age group of 10-20 years, 37 (40.65 percent) in the age group of 20-30 years, 26 patients (28.57 percent) in the age group of

Table 2. Occupation and marital status of patients

		Number of thalassemia major patients	Number of hemodialysis patients

- Index value= $2.831/0.246=11.5$
- Index value= $2.344/0.246=9.53$
- Index value= $0.326/0.246=1.33$

Findings

According to the findings of this study, four individuals with thalassemia major were found to have positive HCV ELISA results, accounting for 4.39 percent of the overall population of thalassemia patients. Additionally, the virus was not found in 87 (95.61%) of these participants, and all 40 hemodialysis patients tested were HCV-negative, accounting for zero percent of the study population (Table 1).

30-40 years, and three patients (3.29 percent) in the age group of 40-50 years, according to the results of thalassemia and hemodialysis patients, and the majority of patients with hepatitis C (100%) are between the ages of 30 and 40. Among 40 hemodialysis patients, two patients (5%) were in the age range of 10-20, 3 patients (7.5%) in the age range of 20-30, 1 patient (2.5%) in the age group of 30-30, 2 (5%) in the age group 40-50 years, 11 patients (27.5%) in the age group 50-60 years, 11 patients (27.5%) in the age group 60-70 years, eight patients (20%) in the age group 70-70 years and two people (5%) were in the age group of 80-90 years, none of which (zero percent) have been infected with hepatitis C.

Patients with thalassemia and those on hemodialysis were also tested for blood group. The results indicated that out of 91 thalassemia major patients, 46 patients (50.5%) had O⁺ blood group, five patients (5.5%) had O⁻ blood group, 22 (24.2%) had A⁺ blood group, and 13 (14.3%) had blood type B⁺, one patient (1.1%) has blood type B⁻, three patients (3.3%) have blood type AB⁺ and one patient (1.1%) has blood type AB⁻, four patients with hepatitis C, two patients (50%) have blood type O⁺, one patient (25%) has blood type O⁻ and one patient (25%) has blood type A⁺. Among 40 hemodialysis patients, 19 patients (47.5%) had blood type O⁺, three patients (7.5%) had blood type O⁻, six patients (15%) had blood type A⁺, one patient (2.5) had blood type A⁻, eight patients (20%) have blood type B⁺, one patient (2.5%) have blood type B⁻, two patient (5%) have blood type AB⁺, none of which (zero%) have hepatitis C.

Occupation	Employed	0	10
	Free job	70	9
	Unemployed	21	21
Marital status	Married	0	10
	Single	70	9

Thalassemia patients and hemodialysis patients were also evaluated in terms of occupation. The results showed that out of 91 thalassemia major patients, 21 patients (23%) were unemployed and 70 (77%) were self-employed, of which 4 (5.7%) with Hepatitis C have a freelance job. Among 40 hemodialysis patients, ten patients (25%) were unemployed, nine patients (22.5%) were self-employed, and 21 patients (52.5%) were employed, none of whom (0%) were infected with hepatitis C.

Thalassemia major patients with hepatitis C virus were also evaluated for marital status. The results showed that out of 91 thalassemia major patients, 89 patients (98%) were single and two patients (2%) were married, of whom four patients were all single with hepatitis C. Out of 40 hemodialysis patients, 37 patients (92.5%) were married and three patients (7.5%) were single, none of whom (0%) had hepatitis C.

Discussion

The thalassemia patients with HCV in this research were all between the ages of 30 and 40; thus, these results are not surprising. This is because elderly patients and patients who receive blood for a longer period are more likely to be infected with the hepatitis C virus for a longer period. Brock et al. (2000) found that HCV-infected thalassemia patients had greater blood transfusion frequency and frequency than the general population. In addition, Rad et al. (2004) found no correlation between a history of blood transfusion and hepatitis C virus antibody-positive in a small study.

In the current study, 47 (52%) of the 91 thalassemia major patients were women, and 44 (48%) were men. 17 (42%) of the 40 hemodialysis patients evaluated in this study were women, whereas 23 (58%) were males. Four women (8.51 percent) of the entire population of women with thalassemia were infected with hepatitis C, while 0 percent of males with thalassemia major were infected. Men and women on hemodialysis were also found to be free of the infection. The findings of this study revealed that women are more likely than males to be infected with this virus, as well as having a history of hospitalization and requiring blood transfusions after surgery (Hofmann JN et al., 2011).

The incidence of hepatitis C virus infection in thalassemia patients and dialysis patients in Kerman province was studied using the RT-PCR technique in 384 participants in a research done in 2009. Women made up 218 (57%) of the population, while males made up 166 (43%). HCV infection was found in 130 of 384 patients (33%), of whom 16 were female, and the

remainder were male (83 percent female and 17 percent male) (Hassan Shahi et al., 2009). The study, which took place in Pakistan in 2021, looked at the relationship between viral loads and patient sex. The average transmission power of women was marginally higher than that of males, according to the findings. The fact that women have greater quantities of the virus than males is a risk factor (Mahnoor et al., 2021). In a research published in 2010, titled Frequency of hepatitis C virus risk factors in high-risk groups (hemophilia, thalassemia, hemodialysis, and injecting drug users) in Mazandaran, men made up the majority of the participants (68.9% men and 31.4% women) (Rafiei et al., 2010). In a study published in 1397 called Prevalence of serum levels of hepatitis C and B in beta-thalassemia major patients of the Amirkola Thalassemia Center, it was discovered that 79.5 percent of patients with thalassemia major were infected with HCV hepatitis virus, with 11 men (44.4%) and 19 women (7.1%) being HBsAg positive (Mahmoudi et al., 2018). In addition, in a research done in Iraq in 2020, men (62.3 percent) had a much greater rate of HCV infection than women (37.7%), with a significant difference ($p < 0.05$) (Khudhair H et al., 2020).

In this research, out of 91 thalassemia major patients, 46 patients (50.5%) have an O⁺ blood group, and five patients (5.5%) have an O⁻ blood group, which indicates the high population of this blood group in the present study and in the community, which four patients affected to hepatitis C, two patients (50%) has blood type O⁺, one patient (25%) have blood type O⁻ and one patient (25%) have blood type A⁺. 19 patients (47.5%) of the 40 hemodialysis patients had blood type O⁺, and three patients (7.5%) had blood type O⁻, indicating that this blood group has a large open population in hemodialysis patients; however none of them (zero percent) had hepatitis C.

According to a report published in Pakistan in 2021, considerable research has been carried out to relate the ABO blood type system to the blood of persons suffering from viral infections and other infectious disorders. There are considerable variances in the distribution of the ABO blood type system in each nation. For identifying a relationship to viral infection in hepatitis C patients, blood group variables show a relationship to the Rh factor, indicating that viral infection is linked to the Rh factor in patients. HCV infection was less common in Rh-negative individuals. HCV-positive individuals with Rh-positive blood type make up 70% of the

total, whereas Rh-negative patients make up 30%. (Mahnoor et al., 2021). HCV patients were more commonly reported in blood group O in the Poor Hassan research in 2014, with the lowest prevalence in blood group AB, and this prevalence in this study reveals a significant link (Pourhassan A, 2014).

Another study conducted in Iraq discovered no link between blood type and the incidence of hepatitis C. Most patients with HCV-infected beta-thalassemia had blood types A and B, according to recent findings, whereas individuals with blood type O appeared to be less vulnerable to the condition (Omar AA et al., 2012). Hepatitis C was shown to be more frequent in patients with blood type A in another study conducted in Thailand (Luksamijarulkul P et al., 2002). Another research in Nigeria's Port-Harcourt found no link between blood type and the hepatitis C virus (Jeremiah ZA et al., 2008).

In the current study, 89 (98%) of the 91 thalassemia major patients were single, whereas 2 (2%) were married, with four of them having hepatitis C. 37 (92.5%) of the 40 hemodialysis patients were married, whereas 3 (7.5%) were single, and none of them (0%) had hepatitis C. There is a correlation between this marital status and the presence of hepatitis C in individuals. There was a significant relationship between hepatitis C genotype types and marital status in a study conducted by Dr. Rafiei et al. in Mazandaran under the title of examining the distribution of hepatitis C virus genotype in thalassemia patients referred to Abu Reihan Disease Center in Bandar Abbas since April (Rafiei et al., 2010).

Conclusion

In conclusion, four cases of hepatitis C virus were found in this study among thalassemia major patients, suggesting that the length and duration of the disease may be effective in hepatitis C. These patients' blood collection periods are likewise extended, implying that they may have been exposed to tainted blood as a result of this. Also, because children with thalassemia major should get blood monthly based on the amount of hemoglobin in their blood from childhood, the length of blood transfusion cannot be overlooked in this circumstance.

Blood transfusions should be screened for viral infections after being obtained from donors, and no blood should be given anywhere without being tested. Furthermore, before transfusing blood to patients, the appropriate departments in hospitals must conduct tests on the blood that has been received.

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Conflict of interest

None.

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Ethics statement

None

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9- The principle of national interests: commitment to respect national interests and considering the progress and development of the country in all stages of research.

In the name of Allah

Charter of research ethics

With the help of God Almighty and the belief that the world is the presence of God and always monitors human actions and in order to pass the high position of knowledge and research and considering the importance of the position of the university in the promotion of human culture and civilization, we, the students and the members of the academic faculty of the university units Azad Islami undertakes to consider the following principles in carrying out research activities and not to exceed them:

- 1- The principle of acquittal: commitment to seek acquittal from any unprofessional behavior and declare a position towards those who bring unscientific suspicions to the field of science and research.
- 2- The principle of respect for fairness and trust: the commitment to avoid any non-scientific bias and protection of property, equipment and resources.
- 3- The principle of promotion: the commitment to the spread of knowledge and the dissemination of research results and their transfer to scientific colleagues and students, except for cases that are prohibited by law.
- 4- The principle of respect: the commitment to respect sanctity and sanctity in conducting research and respect the aspect of criticism and refraining from any violation of sanctity.
- 5- The principle of respect for rights: commitment to fully respect the rights of researchers and researched (humans, animals and plants) and other rights holders.
- 6- The principle of confidentiality: the commitment to protect the secrets and confidential information of individuals, organizations and the country and all individuals and institutions related to research.
- 7- The principle of truth-seeking: trying to find the truth and being loyal to it and avoiding any concealment of the truth.
- 8- The principle of material and moral ownership: commitment to fully respect the material and moral rights of the university and all research partners.

