Prevalence of transfusion-transmitted infections in multiple blood transfusion-dependent thalassemic patients in Asia: A systemic review

Muhammad Riaz1, Mazhar Abbas2, Ghulam Rasool1, Ibrahim Salam Baig3, Zahed Mahmood4, Naveed Muni5, Imtiaz Mahmoud Tahir6, Syed Muhammad Ali Shah7 and Muhammad Akram7

Abstract

Background: Thalassemia is a hereditary hemolytic anemia marked by a defect in synthesizing one or more globin chains in hemoglobin. In Pakistan, approximately 10,000 patients with thalassemia are primarily dependent on blood transfusions. The β-thalassemia patients require blood transfusions and iron chelation therapy. Patients who need blood transfusions are at an increased risk of contracting transfusion-transmitted infections (TTIs) such as hepatitis B and C viruses (HBV and HCV, respectively), as well as the human immunodeficiency virus (HIV).

Objective: This systemic review aims to assess the prevalence of TTIs in transfusion-dependent β-thalassemia patients in Asia.

Methods: The data for the systematic review were gathered from PubMed, Google Scholar, the Directory of Open Access Journals (DOAJ), and ScienceDirect using the following keywords: "prevalence, HBV, HCV, HIV, thalassemia, and transfusion-transmitted infections (TTIs)," and so on. This review includes the research articles that address the prevalence of viral infections in thalassemic patients following blood transfusion.

Results: A preliminary search of various databases identified 231 potential studies. 157 duplicate studies were eliminated, and the eligibility of 59 full-length articles was determined. Only 43 studies met the inclusion criteria. Among the 43 studies analyzed, 11 reported a high prevalence of HCV alone in thalassemic patients, while 21 reported a high prevalence of HCV and HBV infection in thalassemic patients. Eight studies reported the prevalence of all three TTIs examined, namely, HCV, HBV, and HIV, in patients with transfusion-dependent thalassemia.

Conclusion: Preventable transfusion-transmitted infections occur frequently, and robust national policies and hemovigilance are required to detect and mitigate the infection risk.

1Department of Allied Health Sciences, University of Sargodha, Sargodha, Pakistan
2Department of Biochemistry, University of Veterinary and Animal Sciences Lahore (Jhang Campus), Jhang, Pakistan
3Department of Biological Sciences, University of Veterinary and Animal Sciences Lahore (Jhang Campus), Jhang, Pakistan
4Department of Biochemistry, Government College University Faisalabad, Faisalabad, Pakistan
5Department of Medical Laboratory Sciences, School of Health Sciences, University of Management and Technology, Lahore, Pakistan

Corresponding author:
Muhammad Riaz, Department of Allied Health Sciences, University of Sargodha, Sargodha 40100 Pakistan.
Email: riazmlt786@gmail.com

Creative Commons Non Commercial CC BY-NC: This article is distributed under the terms of the Creative Commons Attribution-NonCommercial 4.0 License (https://creativecommons.org/licenses/by-nc/4.0/) which permits non-commercial use, reproduction and distribution of the work without further permission provided the original work is attributed as specified on the SAGE and Open Access pages (https://us.sagepub.com/en-us/nam/open-access-at-sage).
Introduction

Thalassemia is one of the most common genetic abnormalities worldwide, affecting approximately 3% (1.5 million) of the population and resulting in serious health problems such as increased morbidity and premature death and financial and emotional strain on affected families. Thalassemia is a group of hereditary hemolytic anemias defined by the defective synthesis or absence of one or more globin chains of hemoglobin. Thalassemia is classified into two types: alpha (α) and beta (β). Beta thalassemia is caused by a mutation in the beta-globin gene, whereas alpha thalassemia is caused by a mutation in the alpha-globin gene. 

The breakdown of RBCs characterizes β-thalassemia because of defective synthesis of the β-globin chain leading to the complete absence of β or reduced synthesis of β in β-globin chains. Clinically, β-thalassemia is classified into three major subtypes depending on disease severity. These subtypes include transfusion-dependent β-thalassemia major (BTM), moderate β-thalassemia intermedia (BTI), and asymptomatic β-thalassemia trait (BTT). Thalassemias is more prevalent in parts of Africa, the Middle East, Southeast Asia, the Mediterranean Basin, the Indian subcontinent, and Pacific Islands, which range from 1 to 20% as a carrier in these areas. Figures showed that around 270 million people globally are the carriers of variant hemoglobins, approximately 80 million people are β-thalassemia carriers. About 300,000 to 400,000 babies are born annually with a severe hemoglobin disorder, of which 23,000 suffer from thalassemia major, with almost 90% occurring in low-resource countries. Patients affected with thalassemia major require blood transfusions regularly for their survival.

The cornerstone therapy in improving the lifespan and the existence of thalassemic patients is the safe and adequate blood transfusions with regular iron chelation. Suboptimal screening of donors or testing and blood processing will increase the risk of additional complications such as transfusion-transmitted infections. Blood transfusion practices challenge the life of thalassemia patients and can cause new medical problems such as transfusion-transmitted infections like HCV, HBV, HIV, and syphilis infections.

In developing countries, the lives of β-thalassemic patients remain more prone to TTIs as they need frequent blood transfusions for their survival due to unreliable screening tests for blood donors with non-compliance with bio-safety standards. Repeated transfusions could cause further difficulties, like iron overload, since it is not monitored appropriately. However, relatively safe blood transfusion methods are adopted in the developed world due to highly sensitive screening techniques for blood donors such as nucleic acid technology to minimize the chances of infectious blood units being collected during the “window period.” Chiavetta et al. (2003) reported an estimated risk of TTIs in developed countries including Canada, USA, and other European countries are less than 2.5% for each one million contributions. Whereas developing countries still have not integrated the critical requirements for practicing the modern blood transfusion processes.

HBV is one of the most widespread viral infections and is considered the 10th most common cause of death. An expected one-third of the biosphere’s population has serological evidence of HBV infection. The most frequent symptoms of hepatitis B are nausea and jaundice; however, the clinical picture varies among individuals. One patient dies after 30–45 s due to HBV infection. According to The Center for Disease Control and Prevention (CDC) USA, HBV is 10 times more contagious than HCV and a hundred times than HIV. Studies reported that large numbers of individuals have asymptomatic, chronic, and occult hepatitis B infection (OBI). Thalassemia patients are at risk of infection due to frequent blood transfusion along with its constituents. To minimize the spread of HBV infection, most blood transfusion services use an ELISA test for HBsAg detection during donor screening. Despite this, cases of post-transfusion hepatitis have been reported, indicating that the ELISA technique is not entirely effective. HBsAg test is a rapid test routinely used for screening blood donors which is a simple qualitative test and the result is read visually within few minutes. This test is not reliable because of its lower sensitivity and specificity, and subjective evaluation of its result. Nucleic acid amplification technology (NAT) test is an advanced molecular test which detects specific target DNA/RNA segment of the virus amplified in vitro, enabling the detection of low levels of virus in blood sample by increasing the specific target RNA/DNA present in detectable level. NAT testing is the highly recommended and reliable method of donor screening in developed countries.

HCV is an RNA virus from the hepativirus genus and belongs to the family Flaviviridae. It is a hepatotropic virus. Nearly 180 million people are infected with chronic hepatitis with a prevalence of around 2%, and about 3–4 million individuals are being infected every year globally.

Keywords
prevalence, HIV, hepatitis B, hepatitis C, thalassemia

Date received: 15 September 2021; accepted: 5 April 2022
Six genotypes have been recognized amongst the hepatitis C virus identified worldwide. Prevalence of HCV in β-thalassemia patients have been reported in published studies conducted on multi-transfused thalassemia patients. The higher risk of HCV infection in β-thalassemia patients is mainly linked with the mean age of the patients, mean duration, and the extent of blood transfusion. HCV virus is the most prevalent infection in thalassemic patients worldwide, and in Pakistan, 2.2–14% prevalence of HCV infection is described.

Human immunodeficiency virus (HIV) infection and acquired immune deficiency syndrome (AIDS) are caused by HIV, which belongs to a subdivision of retrovirus called lentivirus. HIV is transmitted from infected people through exchanging various body fluids such as blood, semen, breast milk, vaginal secretions, and from mother to child during pregnancy and delivery. Risk factors for the transmission of HIV include unprotected sex, needle stick injuries, unsafe blood transfusion, and tissue transplantation. The transmission risk for the spread of HIV infection through the transfusion of screened blood is minimal. Also, there is a marked reduction in the likelihood of HIV transmission via blood transfusion due to the usefulness of new screening tests. By 2009, UNAIDS Pakistan and National AIDS Control Programme assessed that nearly 130,000 HIV cases in Pakistan had been identified, with an overall prevalence of less than 0.05% in the general population. The frequency of TTIs in transfusion-dependent β-thalassemia patients will be reduced by following the upgraded parameters of blood banks by using globally approved kits to screen donor blood before transfusion to the patients. Furthermore, strict serological blood screening for TTIs before transfusion must be applied in blood banks.

This systemic review aims to assess the prevalence of TTIs in transfusion-dependent β-thalassemia patients from Asia.

Material and methods

**Literature search, inclusion criteria, and data extraction**

The data for the present systematic review was collected from PubMed, Google Scholar, Science direct, PakMediNet, Digital Libraries, and Directory of Open Access Journals (DOAJ) search engine by using the keywords: “Prevalence, HBV, HCV, HIV, Thalassemia, Blood transfusion, Transfusion-transmitted infections (TTIs), Frequent transfusion, and Repeated blood transfusion complications.” All the research articles addressing the prevalence of viral infections amongst thalassemic patients following blood transfusion published during the last 20 years were included. All the studies either reporting the prevalence of all the three types of TTIs, that is, HBV, HCV, and HIV or either one or two of these infections in thalassemic patients were included in the study. Studies reporting other co-infections in thalassemic patients or no co-infection were excluded from the study. In contrast, those studies reporting coinfection of HBV, HCV, and HIV without thalassemia were also excluded from the current systematic review. Those studies were excluded if they were published other than the English language, such as review articles, editorials, letters, and

![Figure 1. Prisma 2009 flow chart summary of selection process.](image-url)
Table 1. Seroprevalence of TTIs amongst multi-transfused thalassemia patients.

| Authors                        | Year  | Country     | Province, city     | Method                           | Sample size | Seroprevalence (%) of anti HCV, HBsAg, and anti-HIV, respectively | Ref. |
|--------------------------------|-------|-------------|--------------------|----------------------------------|-------------|-------------------------------------------------------------------|------|
| Ahmed et al.                   | 2021  | Pakistan    | Baluchistan, Quetta | ELISA, PCR                       | 400         | 18.3, 2.8, *                                                      | 33   |
| Bhyun et al.                   | 2021  | Bangladesh  | Dhaka, Savar       | ELISA                            | 148         | 13.51, 3.37, 0                                                   | 34   |
| Kandi et al.                   | 2021  | India       | Telangana          | ELISA, chemiluminescence         | 100         | 28, *                                                            | 35   |
| Ghafoor et al.                 | 2021  | Pakistan    | Rahim Yar Khan     | ICT                              | 350         | 34.8, 5.1, 1.1                                                   | 36   |
| Mishra et al.                  | 2020  | India       | Khatodara, Surat   | ELISA, NAT                       | 196         | 51.1, 1.5, 3.1                                                   | 37   |
| Yasmeen and Hasnain            | 2019  | Pakistan    | Lahore, Multan,   | ICT                              | 350         | 29.4, 7.4, 0                                                    | 38   |
|                                |       |             | Karachi, and Peshawar |                                 |             |                                                                   |      |
| Bhattacharyya et al.           | 2018  | India       | Hooghly            | ELISA, PCR                       | 300         | 65, *, *                                                         | 39   |
| Dumaidi et al.                 | 2018  | Palestine   | Jenin Nablus       | ELISA, PCR                       | 139         | 10, 0.7, *                                                      | 2    |
| Najim and Hassan               | 2018  | Iraq        | Basra              | ELISA, PCR                       | 2778        | 42.5, *, *                                                      | 40   |
| Ahmadi Vasmehjani et al.       | 2018  | Iran        | Tehran             | ELISA, RIBA, Western blot       | 143         | 4.2, 0, 0                                                        | 41   |
| Vasmehjani et al.              | 2018  | Iran        | Lorestan           | ELISA, RIBA, Western blot       | 143         | 4.2, 0, 0                                                        | 41   |
| Atwa and Wahed                 | 2017  | Egypt       | Fayoum             | ELISA, RT-PCR                    | 121         | 20.7, 5, 0                                                      | 42   |
| Khan et al.                    | 2017  | Pakistan    | Lahore             | ELISA                            | 470         | 45.96, 7.87, 0                                                  | 43   |
| Mittal et al.                  | 2017  | India       | Haryana            | ELISA                            | 211         | 35.5, 2.36, 0                                                   | 7    |
| Mukherjee et al.               | 2017  | India       | Kolkata            | ICT, ELISA                       | 207         | 24.6, 3.38, *                                                   | 44   |
| Gugnani et al.                 | 2017  | India       | Amritsar           | ELISA                            | 126         | 13.4, 0.79, 0                                                   | 45   |
| Wahidyat et al.                | 2017  | Indonesia   | Jakarta            | ECLA                             | 621         | 17.8, 0.8, *                                                    | 46   |
| Yousefi et al.                 | 2017  | Iran        | Zabol              | ELISA, RT-PCR                    | 152         | 8.5, *, *                                                       | 47   |
| Ahmed Kiani et al.             | 2016  | Pakistan    | Islamabad         | CLIA                             | 1253        | 21.7, 3, 0.5                                                    | 32   |
|                                |       |             | Rawalpindi, Karachi |                                 |             |                                                                   |      |
| Mahmoud et al.                 | 2016  | Egypt       | Sohag and Minia    | ELFA                             | 97          | 37.11, 4.12, 0                                                  | 48   |
| Patel et al.                   | 2016  | India       | Jamnagar           | #                                | 177         | 3.95, 2.25, 2.25                                                | 49   |
| Sadia Sultan et al.            | 2016  | Pakistan    | Karachi            | CLIA                             | 100         | 27, 3, 0                                                        | 50   |
| Shah et al.                    | 2016  | India       | Gujarat            | ELISA                            | 55          | 20, 0.0, 3.63                                                   | 51   |
| Goyal et al.                   | 2015  | India       | Rajkot             | ELISA                            | 237         | *, 1.26, 3.37                                                   | 52   |
| Haque and Bin AbLatiff         | 2015  | Malaysia    | Ipoh               | #                                | 100         | 18, 4, *                                                        | 53   |
| Saeed et al.                   | 2015  | Pakistan    | Islamabad         | ICT, ELISA, PCR                  | 262         | 55.73, 3.08, *                                                  | 18   |
|                                |       |             | Rawalpindi         |                                  |             |                                                                   |      |
| Sidhu et al.                   | 2015  | India       | Jammu and Kashmir  | ELISA                            | 138         | 13.04, 0, 0.72                                                 | 54   |
| Hussein                        | 2014  | Egypt       | Cairo              | ELISA, RT-PCR                    | 200         | 24, 3, 0                                                        | 55   |
| Khaled                         | 2014  | Iraq        | Nineveh            | ELISA, PCR                       | 480         | 10.4, 0.4, 0                                                   | 56   |
| Iqbal et al.                   | 2013  | Pakistan    | Rawalpindi         | ELISA                            | 95          | 42.1, *, *                                                      | 57   |
| Karim et al.                   | 2013  | Bangladesh  | Dhaka              | ELISA, Western blots             | 100         | 31, 3, 0                                                        | 58   |
| Khattak et al.                 | 2013  | Pakistan    | Swat               | ELISA                            | 170         | 21.76, 5.88, *                                                  | 59   |
| Said et al.                    | 2013  | Egypt       | #                  | ELISA, RT-PCR                    | 137         | 34.4, *, *                                                      | 60   |
| Ansari et al.                  | 2012  | Pakistan    | Karachi            | ELISA                            | 160         | 13.1, 1.25, 0                                                  | 61   |
| Jain et al.                    | 2012  | India       | Ahmedabad          | ELISA, Western blots             | 115         | 25, 1.04, 1.04                                                 | 62   |
| Azarkeivan et al.              | 2011  | Iran        | Tehran             | #                                | 395         | 27.5, *, *                                                     | 63   |

(continued)
commentaries. Initial search through different databases retrieved 231 studies. After duplicate removal, 157 studies were reviewed, out of which 59 full-text articles with relevant studies were assessed for eligibility while 98 studies were excluded with reasons, as shown in the Prisma flow chart (Figure 1). Only 43 studies on TTIs in thalassemic patients fulfilled the inclusion criteria for this systematic review and were analyzed.

**Statistical analysis**

Data were extracted from all the searched articles and then entered into Microsoft Office Excel 2013. The extracted data was then analyzed for the frequency percentage of the transfusion-transmitted infections (TTIs) in thalassemic patients.

**Results**

A preliminary search of various databases identified 231 potential studies for the current systematic review. 157 of these were duplicates and were therefore excluded from the study. After excluding 98 studies for various reasons, there were 59 full-length articles assessed for eligibility. Only 43 studies involving 12,446 thalassemic patients co-infected with TTIs, such as HCV, HBV, or HIV, met the inclusion criteria and were analyzed.

Table 1 contains data on the percent prevalence of HBV, HCV, and HIV infection in thalassemic patients from various populations. According to the findings above, HCV was the most frequently encountered TTI in multi-transfused thalassemia patients. Among the 43 studies included in this systematic review, 11 reported only HCV prevalence in thalassemic patients, while 21 reported both HCV and HBV prevalence in thalassemic patients. Only one study reported the prevalence of HIV alone in transfusion-dependent thalassemia patients. The lower HBV infection rate among patients with thalassemia major is due to an effective HBV vaccination program that has been in place since 1992 (Table 2).

### Table 1. (continued)

| Authors            | Year | Country | Province, city | Method                          | Sample size | Seroprevalence (%) of anti HCV, HBsAg, and anti-HIV, respectively | Ref. |
|--------------------|------|---------|----------------|--------------------------------|-------------|------------------------------------------------------------------|------|
| Bhavsar et al.     | 2011 | India   | Gujarat        | ELISA, Bispot test and rapid visual band test | 100         | 18, 6, 9                                                         | 64   |
| Bhavsar et al.     | 2011 | India   | Gujarat        | ELISA, Bispot test and rapid visual band test | 100         | 18, 6, 9                                                         | 64   |
| Kalantari et al.   | 2011 | Iran    | Isfahan        | ICT, ELISA and PCR             | 545         | 89.2, 10.8, 0                                                   | 65   |
| Raham et al.       | 2011 | Iraq    | Diyala         | ELISA, RIBA                    | 110         | 26.4, *, *                                                      | 66   |
| Riaz et al.        | 2011 | Pakistan| Karachi        | ELISA                         | 79          | 43, 51, 0                                                      | 67   |
| Vidja et al.       | 2011 | India   | Jamnagar       | ELISA                         | 200         | 2, 2, 3                                                        | 26   |
| Al and Panhotra    | 2002 | Saudi Arabia | Al Hassa     | EIA, RIA                      | 86          | 12.7, *, *                                                     | 68   |

Key: *: Not determined, #: Not mentioned. PCR: Polymerase chain reaction, RIBA: Recombinant ImmunoBlot Assay, CLIA: Chemiluminescence Immunoassay, ELISA: Enzyme-linked immunosorbent assay, ELFA: Enzyme-linked fluorescence assay, RIA: Recombinant immunoblot assay, EIA: Enzyme immunoassay, ECLIA: Electrochemiluminescence immunoassays.

### Table 2. Summary of the studies reported the prevalence of TTIs in thalassemic patients.

| Transfusion-transmitted infections (TTIs) | No of studies reported the prevalence of TTIs (n) |
|------------------------------------------|-----------------------------------------------|
| HCV + HBV + HIV                          | 08                                             |
| HCV + HBV                                | 21                                             |
| HCV + HIV                                | 02                                             |
| HBV + HIV                                | 01                                             |
| HCV only                                 | 11                                             |
| HBV only                                 | 0                                              |
| HIV only                                 | 0                                              |
| Total no of studies                      | 43                                             |
Discussion

Both HBV and HCV have infected 530 million out of 6 billion worldwide. Pakistan is a developing country with a total population of 190 million, sharing a massive burden of infectious diseases. There is a consistent increase in viral infections in Pakistan. The reported prevalence of HBV and HCV is 4.6% and 4.9%, respectively, in local population of Pakistan; Pakistan has started an extended HBV vaccination program for children. However, consistent multiple transfusion to β-thalassemia primary patients has increased their life expectancy, which has also amplified the threat to acquire post-transfusion hepatitis.

This systemic review aimed to summarise the available literature on TTIs prevalence in thalassemic patients in Asia. The current literature survey showed that HCV was the prominent TTI among multi-transfused thalassemia patients. A lower prevalence of HBV infection than HCV among thalassemic patients is likely due to the effective HBV vaccination. Vaccination against HBV is highly effective (80–100%) in decreasing the HBV infection rate in people who received complete HBV vaccine course. Although HBV prevalence is lower than the prevalence of HCV because of improved immunization status, however, it is still on higher side in comparison to HBV global prevalence in thalassemic patients ranging from 0.3–5.7%. Our data on TTIs infections in thalassemic patients showed a higher prevalence of HCV coinfection than HBV and HIV coinfection in transfusion-dependent thalassemia patients in Asia. The HIV prevalence in blood donors is <0.18%. An alarming trend noticed in this review analysis is that variation in the prevalence of HIV in transfusion dependant thalassemic patients in Pakistan and India has been found to be higher than the overall prevalence of HIV in donor population. There is an urgent need to initiate regular testing for HIV for better risk assessment and actual disease burden in thalassemic population.

In low-resource countries, β-thalassemic patients have limited access to safe and regular blood transfusion, possibly due to inadequate testing of blood donations for STIs and the lack of altruistic voluntary blood donors.

Despite screening of blood donors by quick Immunochromatographic Method, HCV infection is still a crucial source of viral hepatitis infection among multi-transfused thalassemia children. There might be drawbacks in donor screening using this method. NAT testing is undertaken using PCR. It can be conducted in pooled testing and, therefore, cost-effective if carried out in large-scale testing and blood banking facilities. Conferring to the study results, TTIs were markedly high. HCV was identified as the leading viral infection among TTIs in transfusion dependant thalassemic patients and HIV infection prevalence was also noted in few cases which might be another distressing situation in the country. This study highlights the research reports from developing countries, as high-resource countries have recently implemented HCV testing since 1991 and NAT testing. It is cost-effective to test on scale through automated and centralized systems. In low-resource countries, particularly in the Indian subcontinent, the development of government policies, haemovigiliance, and education of blood banking provider is critical to reduce the risk of TTIs in blood transfused thalassemic patients.

This review study has certain limitations as limited countries’ data are analyzed. The extracted data is from the research reports published in the last 20 years, mainly from low-resource Asian countries.

Conclusion

The study concluded that the prevalence of TTIs such as HCV, HVB, and HIV is very high in transfusion-dependent thalassemia patients, emphasizing the high prevalence of HCV alone or in combination with HBV. These TTIs could be caused by a fragmented blood transfusion system, insufficient safety measures, or insufficient resources in transfusion-dependent thalassemia patients. To summarize, we propose that enacting robust policies at the regional and national levels regarding safe blood transfusion practices and donor screening under universally quality-assured procedures may help mitigate future TTI risk.

Declaration of conflicting interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) received no financial support for the research, authorship, and/or publication of this article.

Ethical approval

This study reviews published studies and do not require ethical approval.

ORCID iDs

Muhammad Riaz https://orcid.org/0000-0002-5524-7735
Syed Muhammad Ali Shah https://orcid.org/0000-0002-5451-9137

References

1. Surani CC, Shah RV and Sinha M (2018) A study of prevalence of hepatitis-B and hepatitis-C infection in thalassemic patients in a tertiary care hospital, Jamnagar, Gujarat, India. Int J Curr Microbiol App Sci 7: 3142–3146.
2. Dumaidi K, Al-Jawabreh A, Samarah F and Rabayaa M (2018) Prevalence of sero-molecular markers of hepatitis C and B viruses among patients with β-thalassemia major in Northern West Bank, Palestine. Canadian Journal of Infectious Diseases and Medical Microbiology 2018.

3. Ladis V, Karagiorgia-Laganima M, Tsatra I and Choularias G (2013) Thirty-year experience in preventing haemoglobinopathies in Greece: achievements and potentials for optimisation. European journal of haematology 90: 313–322.

4. Kountouris P, Lederer CW, Fanis P, Feleki X, Old J and Kleanthous M (2014) IthaGenes: an interactive database for haemoglobin variations and epidemiology. PloS one 9: e103020.

5. Williams TN and Weatherall DJ (2012) World distribution, population genetics, and health burden of the hemoglobinopathies. Cold Spring Harbor perspectives in medicine 2: a011692.

6. De Sanctis V, Kattamis C, Canatan D, et al. (2017) β-thalassemia distribution in the old world: an ancient disease seen from a historical standpoint. Mediterranean journal of hematology and infectious diseases 9.

7. Mittal K, Abrol P and Yadav J (2017) Prevalence of transfusion transmitted infections amongst multiple blood transfused patients of β-thalassemia major in a tertiary care hospital. Int J Res Med Sci 5: 181–185.

8. Al-Ani SK, Al-Ouqaili MT and Awad MM. Molecular and genotypic study of SENV-D virus coinfection in β-thalassemic patients infected with the hepatitis C virus in Iraq.

9. Viprakasit V, Lee-Lee C, Chong QT, Lin K-H and Voiplong V (2009) Infection therapy in the management of thalassemia: the Asian perspectives. International journal of hematology 90: 435–445.

10. Engelfriet C, Reesink H, McCullough J, et al. (2002) Perioperative triggers for red cell transfusions. Vox Sanguinis 82: 215–226.

11. Chiavetta JA, Escobar M, Newman AM, et al. (2003) Incidence and estimated rates of residual risk for HIV, hepatitis C, hepatitis B and human T-cell lymphotropic viruses in blood donors in Canada, 1990–2000. Cmaj 169: 767–773.

12. Al-Sheyyab M, Batieha A and El-Khateeb M (2014) IthaGenes: an interactive database for haemoglobin variations and epidemiology. PloS one 9: e103020.

13. Raimondo G, Navarra G, Mondello S, et al. (2008) Occult hepatitis B virus in liver tissue of individuals without hepatic disease. Journal of hepatology 48: 743–746.

14. Arababadi MK, Hassanshahi G and Yousefi H (2009) HBV-DNA in hemodialysis patients infected by HCV. Saudi Journal of Kidney Diseases and Transplantation 20: 398.

15. Waheed Y, Saeed U, Anjum S, Afzal MS and Ashraf M (2012) Development of global consensus sequence and analysis of highly conserved domains of the HCV NSSB prote in. Hepatitis monthly 12.

16. Yoshikawa A, Gotanda Y, Minegishi K, et al. (2007) Lengths of hepatitis B viremia and antigenemia in blood donors: preliminary evidence of occult (hepatitis B surface antigen–negative) infection in the acute stage. Transfusion 47: 1162–1171.

17. Yuen M-F, Lee C-K, Wong DK-H, et al. (2010) Prevalence of occult hepatitis B infection in a highly endemic area for chronic hepatitis B: a study of a large blood donor popula tion. Gut 59: 1389–1393.

18. Saeed U, Waheed Y, Ashraf M, Waheed U, Anjum S and Afzal MS (2015) Estimation of hepatitis B virus, hepatitis C virus, and different clinical parameters in the thalassemic population of capital twin cities of Pakistan. Virolology: research and treatment 6: VRT.S1744.

19. Al-Kanaan BM, Al-Ouqaili MT and Al-Rawi KF (2020) Comparative study of the molecular, biochemical, and other parameters in Iraqi hepatitis B patients. Drug Invention Today 14.

20. Plymoth A, Viviani S and Haintaut P (2009) Control of hepatocellular carcinoma through hepatitis B vaccination in areas of high endemicity: perspectives for global liver cancer prevention. Cancer Letters 286: 15–21.

21. Arababadi MK, Hassanshahi G, Yousefi H, Zarandi ER, Moradi M and Mahmoodi M (2008) No detected hepatitis B virus-DNA in thalassemic patients infected by hepatitis C virus in Kerman province of Iran. Pak J Biol Sci 11: 1738–1741.

22. González R, Torres P, Castro E, et al. (2010) Efficacy of hepatitis B virus (HBV) DNA screening and characterization of acute and occult HBV infections among blood donors from Madrid, Spain. Transfusion 50: 221–230.

23. Organization WH (2010) Screening Donated Blood for Transfusion-Transmissible Infections: Recommendations. World Health Organization.

24. Simmonds P, Alberti A, Alter HJ, et al. (1994) A proposed system for the nomenclature of hepatitis C viral genotypes. Hepatology 19: 1321–1324.

25. Mahmood Z, Shakoor A and Riaz M (2013) Investigation of selective biochemical markers from chronic hepatitis C patients in relation to environmental pollutants. World Applied Sciences Journal 24: 1084–1090.

26. Vidja PJ, Vachhiani J, Sheila S and Santwani P (2011) Blood transfusion transmitted infections in multiple blood transfused patients of beta thalassaemia. Indian Journal of Hematology and Blood Transfusion 27: 65–69.

27. Hossain B, Khan W, Tawfiq M and Rahman F (2018) Prevalence of hepatitis C virus infection in multi-transfused Thalassaemia patients in Bangladesh. Journal of Enam Medical College 8: 16–19.

28. Sy T and Jamal MM (2006) Epidemiology of hepatitis C virus (HCV) infection. Int J Med Sci 3: 41.

29. Krämer A, Kreteschmar M and Krickeberg K (2010) Modern Infectious Disease Epidemiology: Concepts, Methods, Mathematical Models, and Public Health. Springer.

30. Ruxruntham K, Brown T and Phanuphak P (2004) Hiv/aids in Asia. Lancet 364: 69–82.
31. Schreiber GB, Busch MP, Kleinman SH and Korelitz JJ (1996) The risk of transfusion-transmitted viral infections. *New England journal of medicine* 334: 1685–1690.

32. Ahmed Kiani R, Anwar M, Waheed U, Asad MJ, Abbasi S and Abbas Zaheer H (2016) Epidemiology of transfusion transmitted infection among patients with β-thalassemia major in Pakistan. *Journal of blood transfusion* 2016.

33. Ahmed S, Ayub M, Naem M, et al. (2021) Thalassemia Patients from Baluchistan in Pakistan Are Infected with Multiple Hepatitis B or C Virus Strains. *The American journal of tropical medicine and hygiene* 104: 1569.

34. Bhuyan GS, Noor AUZ, Sultana R, et al. (2021) Frequency of Hepatitis B, C and HIV infections among transfusion-dependent Beta Thalassemia patients in Dhaka. *Infectious Disease Reports* 13: 89–95.

35. Kandi V, Vinjamuri SR and Tanikella BP (2021) Hepatitis C Viral Infection Among Beta-Thalassemia Patients: A Study From the Centre for Excellence in Thalassemia and Other Blood Disorders. *Cureus* 13.

36. Ghafoor MB, Memon FA, Saleem M and Shabbir R (2021) Prevalence of hepatitis B and hepatitis C virus infection in frequently transfused thalassemic children. *Journal of Liaquat University of Medical & Health Sciences* 20: 31–36.

37. Mishra K, Shah A, Patel K, Ghosh K and Bharadva S (2020) Seroprevalence of HBV, HCV and HIV-1 and correlation with molecular markers among multi-transfused thalassemia patients in Western India. *Mediterranean Journal of Hematology and Infectious Diseases* 12.

38. Yasmeen H and 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: 316–322.

39. Bhattacharyya KK, Biswas A, Gupta D and Sadhukhan PC (2018) Experience of hepatitis C virus seroprevalence and its genomic diversity among transfusion-dependent thalassemia patients in a transfusion center. *Asian Journal of Transfusion Science* 12: 112.

40. Najim OA and Hassan MK (2018) Prevalence of hepatitis C virus seropositivity among multi-transfused patients with hereditary anemias in Basra, Iraq. *Iraqi Journal of Hematology* 7: 39.

41. Ahmadi Vasmehjani A, Yaghushi B, Hashemi SM, et al. (2018) The Prevalence of Hepatitis B, Hepatitis C, and Human Immunodeficiency Virus Infections among β-thalassemia Major: A Multicenter Survey in Lorestan, West of Iran. *Iranian Journal of Pediatric Hematology and Oncology* 8: 111–117.

42. Atwa ZT and Wahed WYA (2017) Transfusion transmitted infections in frequently transfused thalassemic children living in Fayoum Governorate, Egypt: Current prevalence and risk factors. *Journal of infection and public health* 10: 870–874.

43. Khan MR, Anwar S, Faizan M and Nosheen S (2017) The burden of transfusion related infections on thalassemia major children. *Pak J Med Health Sci* 11: 882–886.

44. Mukherjee K, Bhattacharjee D and 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: 4558–4562.

45. Gugnani N, Pandit I, Gupta M, Gugnani S, Soni S and Goyal V (2017) Comparative evaluation of esthetic changes in nonpitted fluorosis stains when treated with resin infiltration, in-office bleaching, and combination therapies. *Journal of Esthetic and Restorative Dentistry* 29: 317–324.

46. Wahidiyat PA, Liauw F, Adnani NB and Putriashih SA (2017) Prevalence of hepatitis B and its correlation with serum ferritin and aminotransferase levels among thalassemia major patients in Indonesia. *Paediatr Indones* 57: 177.

47. Yousefi M, Dehesti MM, Ebad M and Dehghan A (2017) The prevalence of hepatitis C virus infection in patients with thalassemia in Zabol city of Iran. *International Journal of Infection* 4.

48. Mahmoud RA, El-Mazary A-AM and Khodeary A (2016) Seroprevalence of hepatitis C, hepatitis B, cytomegalovirus, and human immunodeficiency viruses in multitransfused thalassemic children in upper Egypt. *Advances in hematology* 2016.

49. Patel N, Unadkat S, Mehta J and Yada S (2016) A study on transfusion transmitted infections (TTIs), transfusion-related complications, and quality of life among the beta-thalassemia major patients in Jamnagar district. *Int J Med Sci Public Health* 5: 1447–1451.

50. Sadia Sultan MBBS F, Siddiqui M and Zaidi SMH (2016) Current trends of seroprevalence of transfusion transmitted infections in Pakistani [Beta]-thalassaemic patients. *The Malaysian journal of pathology* 38: 251.

51. Shah NH, Shrivastav A, Bhavsar U and Agnihotri AS (2016) Study of prevalence of seropositivity in multi-transfused thalassemia patients-a hospital based study. *International Journal of Medical Science and Public Health* 5: 2134–2138.

52. Goyal JP, Hapani PT and Gaggiya H (2015) Prevalence of human immunodeficiency virus and hepatitis B among multi-transfused thalassemia children. *Journal of Applied Hematology* 6: 70.

53. Haque AE and bin Ab Latiff HZ (2015) Prevalence of Hepatitis B and Hepatitis C Infections among Multi-transfused Thalassaemic Patients. *Pharmacology, Toxicology and Biomedical Reports* 1.

54. Sidhu M, Meenia R, Yasmeen I, Sawhney V and Dutt N (2015) Prevalence of transfusion-transmitted infections in multiple blood transfused thalassemia patients: A report from a tertiary care center in North India. *Ann Trop Med Publ Health* 8: 202.

55. Hussein E (2014) Evaluation of infectious disease markers in multitransfused Egyptian children with thalassemia. *Annals of Clinical & Laboratory Science* 44: 62–66.
56. Khaled MD (2014) Prevalence of Hepatitis B, Hepatitis C and human immunodeficiency virus infection among Thalassemia patients in Nineveh governorate. IRAQ. Age 1: 60.

57. Iqbal A, Farrukh H, Aslam S and Iqbal T (2013) Frequency of Hepatitis C in α-Thalassemia major patients. Rawal Medical Journal 38.

58. Karim AR, Islam A, Jamal CY, et al. (2013) Seroprevalence of Hepatitis B, Hepatitis C and Human Immunodeficiency Virus Among Multitransfused Thalassaemic Children in Dhaka, Bangladesh. Bangladesh Journal of Child Health 37: 146–153.

59. Khattak IUD, Shah M, Ahmed I, Rehman A and Sajid M (2013) frequency of hepatitis B and hepatitis C in multitransfused beta thalassaemia major patients in district Swat. Journal of Saidu Medical College, Swat 3: 299–302.

60. Said F, El Beshlawy A, Hamdy M, et al. (2013) Intrafamilial transmission of hepatitis C infection in Egyptian multitransfused thalassaemia patients. Journal of tropical pediatrics 59: 309–313.

61. Ansari SH, Shamsi TS, Khan MT, et al. (2012) Seropositivity of Hepatitis C, Hepatitis B and HIV in chronically transfused ββ-thalassaemia major patients. J Coll Physicians Surg Pak 22: 610–611.

62. Jain R, Perkins J, Johnson ST, et al. (2012) A prospective study for prevalence and/or development of transfusion-transmitted infections in multiply transfused thalassemia major patients. Asian Journal of Transfusion Science 6: 151.

63. Azarkeivan A, Nasiritoosi M, Kafiabad SA, Maghsudlu M, Hajibeigi B and Hadizadeh M (2011) Evaluation of new cases of HCV infection in thalassaemia patients for source of infection. Asian Journal of Transfusion Science 5: 132.

64. Bhavsar H, Patel K, Vegad M, et al. (2011) Prevalence of HIV, Hepatitis B and Hepatitis C infection in Thalassemia major patients in tertiary care hospital, Gujarat. Nat J Integr Res Med 2: 47–50.

65. Kalantari H, Mirzabaghi A, Akbari M and Shahshahan Z (2011) Prevalence of hepatitis C virus, hepatitis B virus, human immunodeficiency virus and related risk factors among hemophilia and thalassemia patients In Iran. Iran J Clin Infect Dis 6: 82–84.

66. Raham TF, Wahed SSA and Alhaddad HN (2011) Prevalence of Hepatitis C among patients with βthalassemia in Diyala-Iraq. Journal of Technique 24.

67. Riaz H, Riaz T, Ullah F, et al. (2011) Assessment of the seroprevalence of viral hepatitis B, viral hepatitis C and HIV in multitransfused thalassaemia major patients in Karachi, Pakistan. Trop Doct 41: 23–25.

68. Al Bahrani A and Panhotra B (2002) Prevalence of hepatitis C virus antibody in polytransfused β-thalassemia major patients. Annals of Saudi medicine 22: 270–272.

69. Saeed U, Waheed Y, Manzoor S and Ashraf M (2013) Identification of novel silent HIV propagation routes in Pakistan. World Journal of Virology 2: 136.

70. Control CfD and Prevention (2015). Epidemiology and Prevention of Vaccine-Preventable Diseases. In: Hamborsky J, Kroger A and Wolfe S (eds). Washington DC Public Health Foundation, p. 2020.

71. Di Marco V, Capra M, Angelucci E, et al. (2010) Management of chronic viral hepatitis in patients with thalassemia: recommendations from an international panel. Blood, The Journal of the American Society of Hematology 116: 2875–2883.

72. Attaullah S, Khan S and Khan J (2012) Trend of transfusion transmitted infections frequency in blood donors: provide a road map for its prevention and control. Journal of translational medicine 10: 1–5.