Red Blood Cells Alloimmunization and Transfusion Strategy in Transfusion Dependent B-Thalassemia Patients

Dr. Dharmesh Chandra Sharma[1]*, Dr. Sachin Singhal[2], Dr. Poonam Woike[3], Dr. A.S.Tomar[3], Dr. Nitesh Rawat[3], Dr. Anita Arya[3], Dr. Rajesh Gaur[3]

[1]*ABTO, Blood Bank, Department Of Pathology, G. R. Medical College, Gwalior*
[2]*Dr.Lal Pathlabs, Gwalior*
[3]*Department of Pathology, G. R. Medical College, Gwalior*

Abstract:

Background: The thalassemia syndromes are a heterogeneous group of inherited disorders caused by genetic lesions leading to decreased synthesis of one or more of the globin subunits. The β-thalassemia major is the most severe form and the affected children are dependent on regular blood transfusions for survival. One of the major complications in chronically transfused patients is development of irregular antibodies and in this situation; further transfusion of compatible red cell is difficult.

Methodology: A total number of 120 patients, those are regularly taking Packed RBC/other RBC components for transfusion from our blood bank are registered for the study. Detection and identification of irregular antibodies was done as per the facilities available. The clinical findings of patients were recorded and the serological results were analyzed prospectively. All data was collected, compiled and compared statistically by frequency distribution and percentage proportion.

Results: In the present study, the mean age of patients was 10.4 ±7.12 SD years, age group wise distribution of patients; 0-5 , 5-10, 11-15, 16-20, 21-25, 26-30, 31- 35 and 36-40 years was 28,52,23,09,06,02,01 and 02 patients respectively. ABO group distributions among Patients were; A: 25, B: 47, O: 40 and AB: 8 where Rh positive and negative patients were 106 and 14respectively. The male: female ratio was 1: 2.3. Mean Hemoglobin was 7.96 ± 1.60 SD g%. Mean interval between 2 transfusions was 22 ± 6.42 SD days. Average number of units (200 ml Packed RBC / Unit) /patient/ month was 1.25. Mean serum ferritin level was 2001 ± 300.7 SD µg/l. Out of 120 patients, 04 patients developed irregular antibodies (p=0.00002) i.e. one patient had anti-e, another had anti- e & anti-K and two had pan-agglutinating antibodies.

Conclusion: Prevalence of alloimmunization in the present study was 3.3%. Extended Blood group i.e. complete Rh and Kell along with ABO matched transfusion from the beginning is helpful in preventing Alloimmunization to much extent. Choice of RBC components is helpful in reduction of serum ferritin level and prolonging interval between two transfusions.

Keywords: Alloimmunization, Irregular antibodies, Thallassemia, Transfusion

I. Introduction

Thalassemias' are widespread throughout the Mediterranean region, Africa, the Middle East, the Indian subcontinent and South East Asia. They occur due to decreased/absent synthesis of β globulin chains [1]. β-thalassemia major is an autosomal recessive genetic disorder, which is clinically; the most severe form and the affected child is dependent on regular blood transfusion for survival [2]. The current management of β-thalassemia major patient is based on regular transfusion of packed red cells and effective chelating therapy [3–6].The aim of the transfusion therapy is to correct anemia and to maintain circulating level of hemoglobin (Hb) sufficient to suppress endogenous erythropoiesis [7].

The adverse effects in these β-Thalassemic Major patients is due to chronic transfusions include iron overload, increased rate of transfusion transmitted infections and alloimmunization to red cell antigens [8]. Though antibody formation can take place against any antigen present on the surface of red cells such as Rh and Kell blood group system [9]. The development of alloantibodies can significantly complicate transfusion therapy [10, 11], due to difficulty in getting compatible blood, delayed haemolytic transfusion reaction and life-threatening hyper-haemolysis syndrome [11, 12, 13]. In our previous study we found that after β-Thallassemia, Hb E/β-Thallassemia is the second most common cause of transfusion-dependent thalassemia in the Gwalior-Chambal region of central India [14]. Present study is aimed to evaluate the prevalence of alloimmunization in the chronically transfused β-Thalassemic major patients and its management at our tertiary care hospital of Gwalior division, Madhya Pradesh, India.

II. Aims And Objectives

1. To assess the rate of sensitization in multi-transfused β- Thallassemia major patients at a tertiary care hospital.
2. Evaluate the ABO, Complete Rh, and Kell identical transfusion in prevention of alloimmunization in β-Thallassemia patients.
3. Helpfulness of different RBC components transfusion in thalassemic patients.
4. Present scenario of β- Thallassemia patients in our region.

III. Methodology

This retrospective, cross-sectional study was conducted at blood bank, J. A. Hospital Gwalior. A total number of 120 patients were selected for the study; those were taking regular transfusion of different RBC components from our blood bank. The RBC components transfused were Packed Red Blood Cells, Saline wash RBCs, Neocytes concentrates and umbilical cord blood [15, 16]. Transfusion strategy for compatibility matching was not only the Rhesus D and ABO but complete Rhesus and Kell blood group was also considered in thalassemic patients at our center by coloumn agglutination method (Matrix Gel Card, Tulip Diagnostic). Direct Coomb's Test (DCT) and Indirect Coomb's Test (ICT) was done on patient blood sample to detect the irregular antibodies (Matrix Gel card, Tulip Diagnostic) and identification of irregular antibodies was done complementarily by tulip diagnostic (P) Ltd. For laboratory investigations we have outsourced Dr. Lal Pathlabs, Gwalior. The clinical findings of patients were recorded and the serological results were analyzed prospectively. We have also maintained the data of the patients which includes age, sex, age of diagnosis, interval between transfusions, number of transfusion, Hb%, serum ferritin level, status of red cell alloimmunization. Most of our patients were on anti-chelating therapy which was supplied free of cost by the administration. All data was collected, compiled and compared statistically by frequency distribution and percentage proportion. Chi-square ($\chi2$) test was applied to know the statistically significant difference (*p* value) of the data. Epicalc version 2000 software was used for the same.

IV. Result

In the present study, the mean age of patients was 10.4 ±7.12 SD years; age group wise distribution of patients was 0-5 , 6-10, 11-15, 16-20, 21-25, 26-30, 31- 35 and 36-40 years was 28,52,23,09,06,02,01 and 02 patients respectively (*p*=0.00001) and statistically significant ((Figure No. 1).
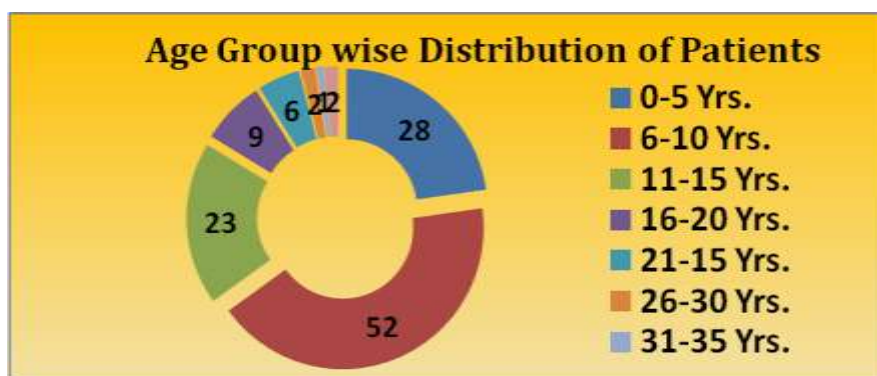

Figure No. 1 Age group wise distribution of the Patients in the present study

ABO group distributions among Patients were; A: 25, B: 47, O: 40 and AB: 8 (*p*=0.00002) where Rh positive and negative were 106 and 14 respectively (*p*=0.00002) and statistically significant. (Figure No. 2).
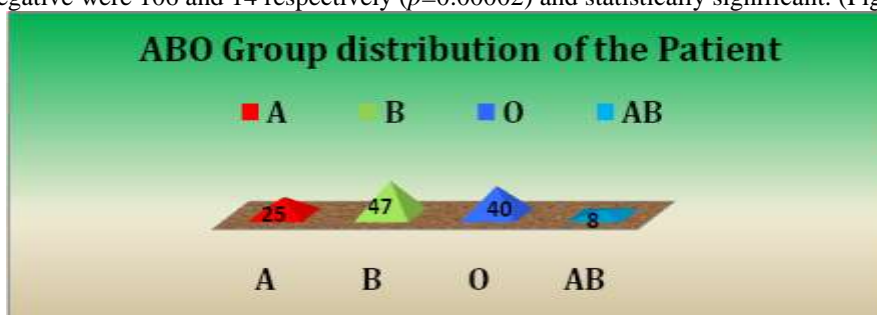

Figure No. 2. ABO Blood group distribution in the present study.

The male**:** female ratio was 1: 2.3. Mean Hemoglobin was 7.96 ± 1.60 SD g%. Mean interval between 2 transfusions was 22 ± 6.42 SD days. Average number of units (200 ml RBC per Unit) / patient/ month was 1.25. The mean number of transfusions given per patient was 127 ± 76.5 SD units till date. Components provided for transfusion were packed red cells, saline wash RBC, Neocytes concentrate, Leukoreduced RBCs and cord blood. Mean serum ferritin level was 2001 ± 300.7 SD µg/l.

Out of 120 patients, 04 patients developed irregular antibodies (*p*=0.00002) and significant strategically (Figure No.3) i.e. one patient had anti-e, another had anti- e & anti-K and two had pan-agglutinating antibodies (Figure No.4)
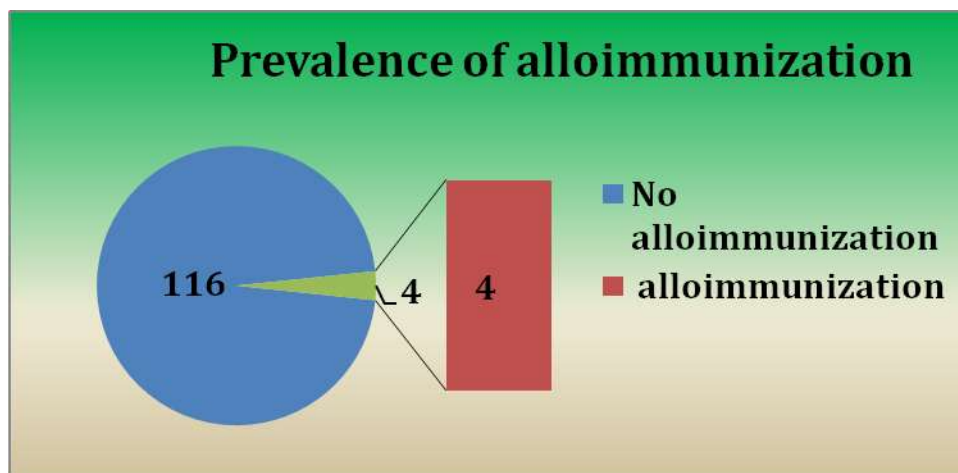

Figure No. 3. Prevalence of alloimmunization in the present study
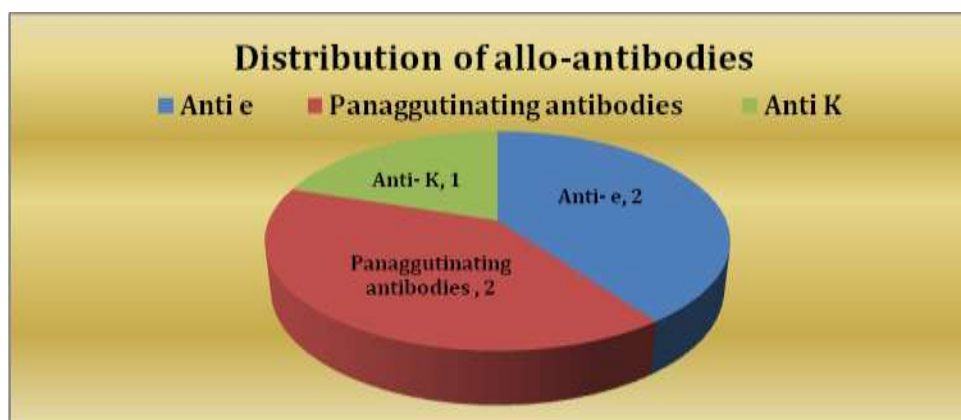

Figure No. 4. Distribution of alloantibodies in the present study

Out of 120 patients, 03 patients had splenectomy and one had successful bone marrow transplant one year back and is alive.

V.   Discussion

Thalassemia was first reported in the literature in 1925, when Cooley and Lee [17] described a form of severe anemia, occurring in children and associated with bone changes and splenomegaly. Although bone marrow transplantation is the only cure, regular blood transfusion is available treatment for these patients [18]. Early and regular blood transfusion therapy in patients with thalassemia decreases the complications of severe anemia and prolongs survival. In the long term, however, the beneficial effects of transfusions are limited by complications such as chronic viral infections, hemosiderosis and alloimmunization against RBC [19].

The factors for alloimmunization are complex and involve at least three main contributing elements: (1) the RBC antigenic difference between the blood donor and the recipient; (2) the recipient's immune status; (3) the immuno- modulatory effect of the allogenic blood transfusions on the recipient's immune system.[20] The frequency of alloimmunization in thalassemia patients in the present study was 3.3% (04/ 120 cases) similar incidence was reported by Mohammad H S *et al.* from north-east Iran 2.87% [21], from Chandigarh, India by Dhawan H K *et al*. 5.64% (18/ 319) [22],), from Rawalpindi, Pakistan by Bhatti FA *et al.* 4.97% (08/ 161)

[23], 5% (71/ 1435) from Italy by Sirchia G *et al*. [24] and 3.7% (44/ 1200) from Greece by Spanos T *et al*. [25] while high incidence 30% (57/190) was reported from Kuwait by Ameen R *et al.* [26].

The previous studies have reported quite variable rate of alloimmunization ranging from 3.1% to 37% in patients of different ethnic origin. A low rate of alloimmunization may be expected when there is homogeneity of RBC antigens between the blood donors and recipients [20]. At our center, most of our patients and blood donor population is from Gwalior and adjoining area of states of Madhya Pradesh. This homogeneity between the patient and blood donors population may be the reason of low rate of alloimmunization in our study.

We did not find any association of gender (male/female) with rate of alloimmunization. In literature, the studies of Ameen R *et al*. [26] El Danasoury *et al*. [27] and Hendrickson *et al*. [28] showed that gender was not a significant factor in the development of alloimmunization. However, Reisner *et al*. [29] reported a significant association between alloimmunization and gender, as they found alloimmunization to be associated more with female patients while on the other hand Saied *et al*.[30] found more association in male patients.

The relationship between the number of units transfused and alloimmunization is unknown in thalassemia. [31] However, some of the studies reported that alloimmunization is more likely in patients who receive more units of blood [32, 33] while Schonewille *et al*. [34] and Saied *et al*.[30] found no significant association between alloantibodies and autoantibodies formation and the number of transfused packed RBCs. In our study we did not find any such relationship.

Dhawan H K *et al*. [22] reported that out of total 23 alloantibodies detected in 319 transfused thalassemic patients, 87.17% belonged to Rh and Kell blood group system, so complete Rh and Kell blood group matched transfusion from the starting will prevent the alloimmunization up to greater extant. As in two patients we reported pan-agglutinating antibodies, in this situation transfusion of compatible RBC is difficult, so least incompatible RBCs were transfused. We reported a very high serum ferritin level in our study; the Mean serum ferritin level was 2001 ± 300.7 SD μg/l, similarly reported by Mishra A k *et al*. 2767.52 μg/l [35]. Better control of Serum ferritin level and prolonged interval between two transfusions was observed by the author in his previous studies [15, 16] by transfusing saline was RBCs, leukoreduced RBCs, Neocytes concentrate and cord blood transfusion in thalassemic patients.

VI. Conclusion

Prevalence of Alloimmunization in the present study was 3.3%. Extended Blood group i.e. complete Rh and Kell along with ABO matched RBC transfusion from the beginning is helpful in preventing Alloimmunization to much extent. Choice of RBC components i.e. saline was RBCs, leukoreduced RBCs, Neocytes concentrate and cord blood transfusion is helpful in reducing serum ferritin level and prolonging transfusion interval between two transfusions.

Ethical Approval

All author(s) hereby declare that all procedure have been examined and approved by the appropriate ethics committee of Gajra Raja Medical College, Gwalior, India and research have therefore been performed in accordance with the ethical standards laid down in the 1964 declaration of Helsinki.

Patient's consent

Informed written consent has been taken from the patient's/ guardians for the study.

Competing Interests

Authors have declared that there are no competing interests.

References

[1]. John NL. The thalassemias and related disorders: Quantitative disorders of hemoglobin synthesis; Wintrobe's Clinical Hematology, tenth edition, Vol. I, chapter 1999 53:1405–1448.
[2]. Collins AF, Goncalves Dias C, Haddad S et al Comparison of a transfusion preparation of newly formed red cells and standard washed red cell transfusion in patients with homozygous beta Thalassemia. Transfusion (1994) 34:517–520
[3]. Thuret I Therapeutic management of patients with thalassemia major. Bull Soc Pathol Exot (2000) 94:95–97

[4].   Weiner M, Kartpatkin M, Hart D et al Cooley's anemia: High transfusion regimen and chelation therapy. Results and prospective. J Paediatr (1978) 92:653–658
[5].   Proter JB Practical management of iron overload. Curr Opin Hematol (1997) 4:436–441
[6].   Graziano JH, Markenson A, Miller DR et al Chelation therapy in β-thalassemia major intravenous and subcutaneous desferoxime. J Paediatr (1978) 92:646–651
[7].   Cazzola M, Stefeno PD, Panchio L et al Relationship between transfusion regimen and suppression of erythropoesis in β-thalassemia major. British J Hematology (1995) 89:473–478
[8].   Suteenee Jansuwan, Orathai Tangvarasittichai, & Surapon Tangvarasittichai Alloimmunization to Red Cells and the Association of Alloantibodies Formation with Splenectomy Among Transfusion-Dependent β-Thalassemia Major/HbE Patients. Indian J. Clin Biochem. 2015 Apr; 30(2): 198–203. doi: 10.1007/s12291-014-0424-6
[9].   Ansari S, Voosogh P, Moshtaghian S Assessment of frequency of alloimmunization and erythrocyte autoimmunization in transfusion dependent thalassemia patients. Acta Medica Iranica. 2008; 46:137–140.
[10].   Schonewille H, Haak HL, van ZijI AM. Alloimmunization after blood transfusion in patients with hematologic and oncologic diseases. Transfusion. 1999; 39:763–771. doi: 10.1046/j.1537-2995.1999.39070763.x. [PubMed] [Cross Ref]
[11].   Eder AF, Chambers LA Noninfectious complications of blood transfusion. Arch Pathol Lab Med.2007;131:708–718. [PubMed]
[12].   Chonewille H, van de Watering LM, Brand A. Additional red blood cell alloantibodies after blood transfusion in a non hematologic alloimmunized patient cohort: is it time to take precautionary measures? Transfusion. 2006;46:630–635. doi: 10.1111/j.1537-2995.2006.00764.x. [PubMed] [Cross Ref]
[13].   Wang LY, Liang DC, Liu HC, Chang FC, Wang CL, Chan YS, et al. (2006) Alloimmunization among patients with transfusion-dependent thalassemia in Taiwan. Transfus Med. 2006;16:200–203. doi: 10.1111/j.1365-3148.2006.00656.x. [PubMed] [Cross Ref]
[14].   Kumar R, Sharma D C, Kishor P. "Hb E/β-Thalassemia: The Second Most Common Cause of   Transfusion-Dependent Thalassemia in the Gwalior-Chambal Region of Central India." Hemoglobin Oct 2012, Vol. 36, No. 5: 485–490. PMID: 22738610 http://lib.bioinfo.pl/paper:22738610
[15].   Dharmesh Chandra Sharma, Sunita Rai, Navneet Agarwal, Satya Sao, Ajay Gaur, Rahul   Sapra."Transfusion of neocytes concentrate/pooled neocytes in β-thalassemic   patients" Indian Journal of Hematology and Blood Transfusion 24(4):December 2008; 173–177. link.springer.com/article/10.1007/s12288-008-0042-7
[16].   Dharmesh Chandra Sharma, Nidhi Agrawal , Jyoti Bindal, Poonam Woike, Rajesh Gaur 120 Units of Placental Umbilical Cord Blood Transfusions in 77 Patients with Different Clinical Conditions. Abstract ; Transmedicon 2016 Bhopal O044 p 181.
[17].   Cooley TB, Lee P. A series of cases of splenomegaly in children with anemia and peculiar bone changes. Trans Am Pediatr Soc 1925;37:29-30
[18].   Weatherall DJ. Disorders of globin synthesis: thalassemia. In: Lichtman MA, Beutler E, Kipps TJ, Seligson U, Kaushansky K, Prchal JT, editors Williams hematology. 7th ed. New York: McGraw-Hill; 2006. pp. 633–57.
[19].   Borgna-Pignatti C, Galanello R. Thalassemias and related disorders: Quantitative disorders of hemoglobin synthesis. In: Greer JP, Rodgers GM, Paraskevas F, Foerster J, Lukens JN, Glader B, editors. Wintrob's Clinical Hematology. 11th ed. Philadelphia: Lippincott Williams and Wilkins; 2004. pp. 1332–5.
[20].   Singer ST, Wu V, Mignacca R, Kuypers FA, Morel P, Vichinsky EP. Alloimmunization and erythrocyte autoimmunization in transfusion-dependent thalassemia patients of predominantly Asian descent. Blood.2000;96:3369–73. [PubMed]
[21].   Mohammad Hadi Sadeghian, Mohammad Reza Keramati, Zahra adiei, Mehrangiz Ravarian, Hossein Ayatollahi,Houshang Rafatpanah, and Mohammad Khajeh Daluei. Alloimmunization among transfusion-dependent thalassemia patients Asian J Transfus Sci. 2009 Jul; 3(2): 95–98. doi: 10.4103/0973-6247.53884 PMCID: PMC2920480
[22].   Hari Krishan Dhawan, Vijay Kumawat, Neelam Marwaha, Ratti Ram   sharma, Suchet Sachdev, Deepak Bansal, Ram Kumar Marwaha, and Satyam Arora. Alloimmunization and autoimmunization in transfusion dependent thalassemia major patients: Study on 319 patients. Asian J Transfus Sci. 2014 Jul-Dec; 8(2): 84–88. doi: 10.4103/0973-6247.137438 PMCID: PMC4140069
[23].   Bhatti FA, Salamat N, Nadeem A, Shabbir N. Red cell immunization in beta thalassaemia major. J Coll Physicians Surg Pak. 2004;14:657–60. [PubMed]
[24].   Sirchia G, Zanella A, Parravicini A, Morelati F, Rebulla P, Masera G, et al. Red cell alloantibodies in thalassemia major.Results of an Italian cooperative study. Transfusion. 1985;25:110–2. [PubMed]
[25].   Spanos T, Karageorga M, Ladis V, Peristeri J, Hatziliami A, Kattamis C. Red cell alloantibodies in patients with thalassemia. Vox Sang. 1990;58:50–5. [PubMed]
[26].   Ameen R, Al-Shemmari S, Al-Humood S, Chowdhury RI, Al-Eyaadi O, Al-Bashir A. RBC alloimmunization and autoimmunization among transfusion-dependent Arab thalassemia patients. Transfusion. 2003;43:1604–10. [PubMed]
[27].   El Danasoury AS, Eissa DG, Abdo RM, Elalfy MS. Red blood cell alloimmunization in transfusion-dependent Egyptian patients with thalassemia in a limited donor exposure program. Transfusion.2012;52:43–7. [PubMed]
[28].   Hendrickson JE, Desmarets M, Deshpande SS, Chadwick TE, Hillyer CD, Roback JD, et al. Recipient inflammation affects the frequency and magnitude of immunization to transfused red blood cells. Transfusion.2006;46:1526–36. [PubMed]
[29].   Reisner EG, Kostyu DD, Phillips G, Walker C, Dawson DV. Alloantibody responses in multiply transfused sickle cell patients. Tissue Antigens. 1987;30:161–6. [PubMed]
[30].   Saied DA, Kaddah AM, Badr Eldin RM, Mohaseb SS. Alloimmunization and erythrocyte autoimmunization in transfusion-dependent Egyptian thalassemic patients. J Pediatr Hematol Oncol.2011;33:409–14. [PubMed]
[31].   Shamsian BS, Arzanian MT, Shamshiri AR, Alavi S, Khojasteh O. Frequency of red cell alloimmunization in patients with b-major thalassemia in an Iranian referral hospital. Iran J Pediatr.2008;18:149–53.
[32].   Fluit CR, Kunst VA, Drenthe-Schonk AM. Incidence of red cell antibodies after multiple blood transfusion. Transfusion. 1990;30:532–5. [PubMed]
[33].   Rosse WF, Gallagher D, Kinney TR, Castro O, Dosik H, Moohr J, et al. Transfusion and alloimmunization in sickle cell disease. The Cooperative Study of Sickle Cell Disease. Blood.1990;76:1431–7. [PubMed]
[34].   Schonewille H, van de Watering LM, Loomans DS, Brand A. Red blood cell alloantibodies after transfusion: Factors influencing incidence and specificity. Transfusion. 2006;46:250–6. [PubMed]
[35]. Amit Kumar Mishra and Archana Tiwari. Iron Overload in Beta Thalassaemia Major and Intermedia Patients. Maedica (Buchar). 2013 Sep; 8(4): 328–332. PMCID: PMC3968466