



Review on Thalassemia

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Abstract

Thalassemia was clinically described almost 100 years ago and treatment of this genetic disease has seen a great progress during this period. DNA-based diagnosis proved the molecular basis of the disease and clarified the variable clinical picture. It also laid path for modern methods of carrier identification and prevention through DNA-based prenatal diagnosis. All aspects of supportive care, like safer blood supply, regular transfusions, monitoring of iron overload, parenteral as well as oral chelation, and other therapies, helped to prolong life and improve the quality of life of the patients. Many advances are also seen in allogenic bone marrow transplantation, which is the only curative therapy. Recent research has mainly focused on studying thalassemia at the basic science level, which resulted in the identification of unknown mechanisms leading to anemia and enabling the development of novel therapies which helps to improve the treatment of, and possibly cure the disease. Various pathways involving activin receptors, JAK2 inhibitors, heat shock proteins and macrophage targeted therapy, etc are under study or are currently undergoing clinical trials for treating thalassemia. Novel genetic therapies are being investigated.

Keywords: *Alpha thalassemia; Hemolytic anemia; Transfusions; Hypercoagulability*

Introduction

The thalassemias are monogenetic hematologic disorders which are resulted by faulty synthesis of one or more of the hemoglobin chains. Alpha and beta thalassemias are caused by reduction or complete absence in the synthesis of alpha or beta globin chains respectively. Imbalances in globin chains leads to hemolysis and impair erythropoiesis. Silent carriers of alpha thalassemia and people with alpha or beta thalassemia trait are asymptomatic and doesn't need treatment. Alpha thalassemia intermedia, or hemoglobin H disease, causes hemolytic anemia. Alpha thalassemia major with hemoglobin Bart's causes fatal hydrops fetalis. Beta thalassemia major results in hemolytic anemia, poor growth, and skeletal abnormalities during infancy. Affected children need lifelong blood transfusions. Beta thalassemia intermedia is less severe when compared with beta thalassemia major and may require episodic blood transfusions. People with beta thalassemia major often die due to cardiac complications of iron overload by the age of 30. Hence it is the need of the hour to have proper understanding of mechanism underlining thalassemia and thorough knowledge of the recent advances in the thalassemia research. Open access journals provide a platform for the practitioner as well as researchers to share their research and findings and also to access

scientific work of their interest which are published in those journals. Open access journals not only disseminate knowledge but also help the researchers and authors to share their expertise and gain popularity in their respective fields.

Many physicians, haematologists, geneticists, researchers and NGOs unite to form societies and/or associations to bring awareness among the parents and caretakers of the patients suffering with thalassemia and to support them in various ways like collecting and providing the blood donors details, financial assistance and even funding thalassemia research etc. Many societies like Thalassemia International Federation aims to provide equal access to quality health care for every patient with thalassemia and other hemoglobin disorders across the world. Thalassaemia International Federation has developed an internationally recognised educational programme, with the objective to continuously educate health professionals, patients and their families, the community at large and policy-makers, based on four ways:

- By organising educational events including workshops, conferences and seminars at the regional, national and international level;
- The preparation, translation and distribution of publications;
- The preparation, organisation and contribution in courses, e.g., electronic educational platform and ENERCA, and;
- The preparation and organisation of educational courses for patients and parents, e.g., the Expert Patients' Programme and Capacity Building (<https://www.thalassaemia.org.cy/about-us/educational-programme>)

The societies support the understanding of thalassemia by creating awareness among the global communities and help in providing better service to the patients and work together to improve the quality of life of the patients.

Many international peer-reviewed scholarly journals such as Journal of Genetic Disorders and Genetic Reports, Blood Research & Hematologic Diseases publish valuable research papers on topics of genetic background of thalassemias and blood disorders, anemia's, thalassemia etc. respectively. Dr. Mahmoud Sirdah et al. had published a research article entitled "Genotype-phenotype characteristics of β thalassemia children in the Gaza Strip, Palestine" where they explored the molecular, biochemical and haematological aspects of β -thalassemia children aged between 5-12 years in the Gaza Strip, in about 325 patients who have been diagnosed with β -thalassemia major, they also investigated for any genotype/phenotype associations which could be used in improving management protocols, such as blood transfusions and iron chelation, for thalassemia patients.

Conferences like International Conference on Hematology & Blood Disorders allows students and researchers to present their research and ideas to the pool of delegates attending the conference and can receive suggestions of the experts, these types of interactions will motivate and help them excel in their field. Hongxia Yao presented a poster entitled 'The spectrum of α and β -thalassemia mutations of the Li people in Hainan province of China' in 3rd International Conference on Hematology & Blood Disorders.

Along with journals and conferences many researchers had penned their knowledge regarding thalassemia in the form of books which are equally helpful in throwing light regarding the disease. 'Thalassemia intermedia' a book written by Wijdene El Borgi and Naouel Ben Salah had detailed various aspects of thalassemia research like genetics, pathophysiology, diagnosis, complications and management of thalassemia [1-13].

Diagnosis of Thalassemia

It all started with electrophoresis of haemoglobin which provide the insights in pathophysiology of haemoglobinopathies. Later DNA and RNA analysis hastened understanding of the disease. Subsequently, restriction enzyme analysis of high molecular weight DNA with Southern blotting revealed crucial information . However, the complicated nature and was unsuitable for analyzing beta thalassemia, which is caused by point mutations encouraged the use of PCR which revolutionarised the research and Various permutations of PCR have revealed hundreds of point mutations leading to alpha- and beta-thalassemia, and gap-PCR can analyze deletions. The large quantity of information gathered about thalassemia mutations has been stored in a freely accessible online database like HbVar [14-38].

Therapy of Thalassemia

Various therapies are used in treatment as well as management if thalassemiias. They can be broadly grouped into conventional therapy and novel therapies.

Conventional Therapy

Transfusions and chelation: As defective erythropoiesis is one of the major pathogenic mechanisms of disease manifestations (bone deformities, hepatosplenomegaly due to extramedullary hematopoiesis) transfusion was the mainstay of therapy. However transfusion reactions due to alloantibodies, transfusion transmissible diseases were well documented in the older literature. The major reason being differences in ethnicity in donors and recipients of transfusions, alloimmunization etc. Resent advances in blood banking like better crossmatching, modern standards of safe blood banking with much reduced pathogen transmission etc benefitted the transfusion dependent patients.

Hemosiderosis is a common complication of long term transfusion therapy hence desferrioxamine was used to prevent iron overload since long time however it is not suitable to all patients this lead to investigations to find new chelators. Though different chelators were brought into use they have shown one or other minor complications [39-66].

Hypercoagulability: Hypercoagulability is one of the major factors contributing to death in thalassemia patients. Although many antithrombotic agents are available there are no clearcut protocols to deal with these complications [67-72].

Bone Marrow Transplantation: Allogenic bone marrow transplantation is a curative therapy which involves replacement of the patient's bone marrow with that of a suitable donor's. In spite of successful results this technique faces challenges from alloimmunization, availability of donors and huge expenses [73-80].

Novel Therapies

Fetal hemoglobin induction: Many agents have been used with varying success to pharmacologically raise the levels of HbF with tolerable adverse effects however results till date are mostly been disappointing. Recent studies on the regulatory pathways of HbF expression, including the influences of MYB and BCL11a, have suggested new therapeutic targets for raising HbF. These include interfering with MYB or BCL11a, which directly repress g-globin gene expression. However, currently there are no specific drugs which can target these pathways [81-86].

Activin receptor ligand traps: Recent findings of the role of GDF11 in erythropoiesis opened new areas for treating thalassemia. While testing a drug named sotatercept an activin odifier for osteoporosis it was found to raise haemoglobin levels by 12% which led to hypothesis that it can be used for anaemis. When sotatercept was tested in mouse model of thalassemia remarkable results were found with increased terminal differentiation of erythroblasts, long with correction of maturation arrest, along with RBC morphology improvement, with fewer aglobin precipitates on RBC membranes. Furthermore, splenomegaly decreased. Notably, hepcidin levels also raised and reduced iron and transferrin saturation was observed, which was independent of GDF15 expression. This is of importance because GDF15 has been suspected as the culprit in down regulating hepcidin in thalassemia, resulting in iron overload. Luspatercept a congener of sotatercept has been tested in Phase I study on normal human volunteers and in a Phase 2 study on thalassemia patients . Both the results shown a rise in haemoglobin levels with tolerable side effects [87-90].

JAK2 inhibitors: Rise in erythropoietin levels, as observed in thalassemia and other anaemia, results in increase in the number of erythroid precursors carrying phosphorylated Jak2. This is considered to contribute to ineffective erythropoiesis, with massive erythroid hyperplasia and extramedullary hematopoiesis. Hence it is plausible to administer JAK2 inhibitors to reduce splenomegaly and ineffective erythropoiesis in nontransfusion dependent thalassemia. However this may be counterintuitive, as these drugs can result in anemia and other cytopenias, This approach is presently being tested in clinical trials over selected patients [91-93].

Macrophage targeted therapy: The central macrophage of the erythroid island in the bone marrow interacts with surrounding erythroblasts and provide regulatory feedback during erythropoiesis along with nutrient supply and phagocytosing extruded erythroblast nuclei. In thalassemia these macrophages are found to exert a negative effect on hematopoiesis. Recent studies involving macrophage depletion showed significant improvement of clinical features in a murine model of thalassemia intermedia with improvement in splenomegaly, reduction in ineffective erythropoiesis, increased Haemoglobin with improved erythrocyte morphology (along with reduced accumulation of a-globin chains in the RBC membrane), and improved iron kinetics [94-97].

Heat shock protein 70: Thalassemic erythroid precursors undergo accelerated apoptosis while normal precursors don't. An essential erythroid transcription factor named GATA-1, is cleaved by transiently active caspases during late erythroid differentiation. In normal erythroid precursors this GATA-1 protein is protected from the premature capsase by a chaperon protein called heat shock protein 70. This protein is translocated to the nucleus and protects GATA-1 proteolysis. However, in b-thalassemia, heat shock protein 70 interacts with free a-globin chains present in the cytoplasm, which sequester it there and prevent its translocation to the nucleus to protect GATA-1. This leads to maturation arrest and apoptosis. Genetic manipulations like transfecting an uncleavable GATA-1 or a heat shock protein targeting the nucleus resulted in normal maturation of thalassemia erythroblasts [98,99].

Genetic therapies: Scientists have been trying to replace nonfunctional beta-globin genes with cloned beta globin genes but this has been hurdled by many factors like problems related to the vectors, to the complex regulation of globin gene expression, as well as the very high level expression which was required for correction of genetic defects [100-105].

Modulation of hepcidin: Though chelation is a successful therapy the ideal remedy for iron therapy would be its prevention. Along with transfusional iron overload, thalassemia patients show hyperabsorption of iron due to insufficient production of the peptide hepcidin. Many strategies have been attempted to compensate hepcidin insufficiency ranging from use of hepcidin mimetic agents (“minhepcidins”), transgenic overexpression of hepcidin, and repression of Tmprss6 etc [106].

Conclusion

Thalassemia can be well addressed through an integrative approach of neonatal screening and systematic treatment and maintenance therapy. A lot of information is shared in open access journals which are available to be accessed freely and used by various professionals like haematologists, physicians, geneticists and caretakers for treating thalassemias.

REFERENCE

1. Kaitoukov Y, Zdanovich E, Moser T, et al. Extramedullary Hematopoiesis in Beta Thalassemia Major: Multisystem Involvement. *OMICS J Radiol.* 2016; 5:234.
2. Jameel T, Suliman IM, Rehman D. The Compromised Quality of Life in β -Thalassemia Major Children in Non-Urban Setup in a Developing Country. *J Hematol Thrombo Dis.* 2016; 4:245.
3. Patir P, Soyer N, Saydam G. Beta-thalassemia Major and Non-Hodgkin Lymphoma. *J Blood Disord Transfus.* 2016; 7:352.
4. Vanikioti K, Preechawat P, Cheecharoen P, et al. Compressive Optic Neuropathy in Thalassemia: A Rare Ophthalmic Consequence of Extramedullary Hematopoiesis. *J Clin Exp Ophthalmol.* 2016;7:549.
5. Mrabet S, Bourkhis L, Abdelkarim SB, et al. IgA Nephropathy, β Thalassemia Minor, Psychosis and Deafness: A New Syndrome?. *J Nephrol Ther.* 2016; 6:237.
6. Inusa BP, Daniel Y, Lawson JO, et al. Sickle Cell Disease Screening in Northern Nigeria: The Co- Existence of B- Thalassemia Inheritance. *Pediat Therapeut.* 2015; 5:262.
7. Derakhshan SM, Khorrami A, Feizi AHP, et al. Spectrum of β -Globin Gene Mutations and β -Thalassemia Haplotype Analysis among the Iranian Azeri Turkish Population. *Epidemiology (sunnyvale).* 2015; 5:210.
8. Gokce M, Kup H, Tugcu D, et al. Insidious Renal Damage in Patients with Thalassemia Major: Is it More Serious than Appreciated?. *J Hematol Thrombo Dis.* 2014; 2:173.
9. El Beshlawy A, Fawaz L, Hamdy M, et al. Urinary Iodine in Egyptian Patients with Thalassemia Major: A Focus on its Potential Contributing Impact on Consequent Hypothyroidism. *J Mol Biomark Diagn.* 2015; 6:211.
10. Thakur S, Sharma R, Raw SN. Incidence of Thalassemia and Sickle Cell Disease in Chhattisgarh, Central India: Using Hardy-Weinberg Equations. *J Mol Genet Med.* 2015; 9:155.
11. Asadov ChD. Immunologic Abnormalities in β -Thalassemia. *J Blood Disord Transfus* 5:224.
12. Belhouli KM, Khadhim AM, Dewedar HE, et al. β -Thalassemia Intermedia with Immune Hemolysis during Pregnancy: A Report of Two Cases. *J Blood Disord Transfus.* 2013; 5:185.
13. Secer M, Ulutas M, Tosun O, et al. Vertebra Plana due to Thalassemia: A Case Report. *J Spine Neurosurg.* 2015; 4:5.
14. Dhawan D, Chaudhary S, Chandratre K, et al. Prenatal Screening for Co-Inheritance of Sickle Cell Anemia and β -Thalassemia Traits. *Clin Med Biochemistry Open Access.* 2015; 2:108.

15. Naveed S, Abbas SS, Qamar F, et al. Triumph Over Thalassemia- Universal Wakefulness. *J Bioequiv Availab*. 2016; 8:106-107.
16. Nienhuis AW, Anderson WF. Isolation and translation of hemoglobin messenger RNA from thalassemia, sickle cell anemia, and normal human reticulocytes. *J Clin Invest* 1971;50: 2458–2460.
17. Yassin M M, Sirdah M M, Al Haddad R M, et al. Genotype-phenotype characteristics of β thalassemia children in the Gaza Strip, Palestine. *J Genet Disor Genet Rep*. 2013; 2:2.
18. Kacian DL, Gambino R, Dow LW, et al. Decreased globin messenger RNA in thalassemia detected by molecular hybridization. *Proc Natl Acad Sci USA* 1973;70:1886–1890.
19. Arica V, Arica S, Özer C, Çevik M. Serum Lipid Values in Children with Beta Thalassemia Major. *Pediat Therapeut*. 2012; 2:130.
20. Saiki RK, Bugawan TL, Horn GT, et al. Analysis of enzymatically amplified beta-globin and HLA-DQ alpha DNA with allele-specific oligonucleotide probes. *Nature* 1986;324:163–166.
21. Hardison RC, Chui DH, Giardine B, et al. HbVar: A relational database of human haemoglobin variants and thalassemia mutations at the globin gene server. *Hum Mutat* 2002;19:225–233.
22. Abdullah UYH, Attraqchi AGFA, Ibrahim HM, et al. The Free Alpha-Hemoglobin: A Promising Biomarker for β -Thalassemia. *J Mol Biomark Diagn*.2014; 5:197.
23. Abdullah UYH, Al-Attraqchia AGF. Reasons of Unknown HPLC Peaks during Hemoglobin Analysis for Patients with Beta Thalassemias. *J Hematol Thrombo Dis*.2014; 2:160.
24. Giardine B, Borg J, Viennas E, et al. Updates of the HbVar database of human hemoglobin variants and thalassemia mutations. *Nucleic Acids Res* 2014;42:D1063–D1069. doi:10.1093/nar/gkt911
25. Wainscoat JS, Thein SL, Weatherall DJ. Thalassaemia intermedia. *Blood Rev* 1987;1:273–279.
26. Mishra G, Saxena R, Mishra A, et al. Recent Techniques for the Detection of β -Thalassemia: A Review. *J Biosens Bioelectron*.2012; 3:123.
27. Rund D, Rachmilewitz EA. Beta thalassemia. *N Engl J Med* 2005;353:1135–1146.
28. Weatherall DJ. The definition and epidemiology of non-transfusion-dependent thalassemia. *Blood Rev* 2012;26:S3–S6.
29. Cao A. Results of programmes for antenatal detection of thalassemia in reducing the incidence of the disorder. *Blood Rev* 1987;1:169–176.
30. Chiu RW, Lau TK, Leung TN, et al. Prenatal exclusion of beta thalassaemia major by examination of maternal plasma. *Lancet* 2002;360: 998–1000.
31. Papasavva T, van Ijcken WF, Kockx CE, et al. Nextgeneration sequencing of SNPs for non-invasive prenatal diagnosis: Challenges and feasibility as illustrated by an application to b-thalassaemia. *Eur J Hum Genet* 2013;21:1403–1410.
32. Berger VK, Baker VL. Preimplantation diagnosis for single gene disorders. *Semin Reprod Med* 2014;32:107–113.
33. Toufexis M, Lewin A, Storch EA, et al. A Possible Link Between Tic Disorders Associated with Beta Thalassemia Minor and Sickle Cell Disease. *J Clin Case Rep* 2013;3:244.
34. Cao A, Kan YW. Prevention of thalassemia. *Cold Spring Harb Perspect Med*. 2013;3: a011775.

35. Ray PF, Kaeda JS, Bingham J, et al. Preimplantation genetic diagnosis of beta-thalassaemia major. *Lancet* 1996;347:1696.
36. Traeger-Synodinos J. Preimplantation genetic diagnosis, an alternative to conventional prenatal diagnosis of the hemoglobinopathies. *Int J Lab Hematol* 2013;35:571–579.
37. Dickens BM. Preimplantation genetic diagnosis and 'savior siblings.' *Int J Gynaecol Obstet* 2005;88:91–96.
38. Hassan S, Ahmad R, Hamid FSA, et al. Compound Heterozygous -30 (T>C) and Cd 41/42 (-TTCT) Leading to β -thalassemia Major in a Malay Autistic Patient. *J Blood Disord Transfus.* 2015; 6:246.
39. Hassan S, Ahmad R, Hamid FSA, et al. Transfusion Dependent Homozygous α -Thalassemia in Patients Associated with Hypospadias in Three Survivors. *J Blood Disord Transfus.* 2015; 6:248.
40. Smith CH, Schulman I, Ando RE, Stern G. Studies in Mediterranean (Cooley's) anemia. II. The suppression of hematopoiesis by transfusions. *Blood* 1955;10:707–717.
41. Rasika S, Satyam A, Anil H, et al. Efficiency of Pre-Storage Leukoreduction of Red Cells: A Double Blind Quality Control Method. *J Blood Res Hematol Dis.* 2016; 1:2.
42. Chansiw N, Pangjit K, Phisalaphong C, et al. Toxicity Study of a Novel Oral Iron Chelator: 1-(N-Acetyl-6-Aminoethyl)-3 Hydroxy-2-Methylpyridin-4-One (CM1) in Transgenic β -Thalassemia Mice. *Vitam Miner.* 2013;2:116.
43. Vichinsky E, Neumayr L, Trimble S, et al. Transfusion complications in thalassemia patients: A report from the Centers for Disease Control and Prevention (CME). *Transfusion* 2014; 54:972–981.
44. Niihara Y, Macan H, Eckman JR, et al. L-Glutamine Therapy Reduces Hospitalization for Sickle Cell Anemia and Sickle β^0 -Thalassemia Patients at Six Months – A Phase II Randomized Trial. *Clin Pharmacol Biopharm.* 2014; 3:116.
45. Hagag AA, Abd Elfatah M. Therapeutic Value of Silymarin as Iron Chelator in Children with Beta Thalassemia with Iron Overload. *J Leuk (Los Angel).* 2014; 2:132.
46. Goss C, Giardina P, Degtyaryova D, et al. Red blood cell transfusions for thalassemia: Results of a survey assessing current practice and proposal of evidence-based guidelines. *Transfusion* 2014;54:1773–1781.
47. Rosnah B, Noor Halina MN, et al. The Level of Natural Anticoagulants in Transfusion Dependent Thalassemia Patients in Kelantan, Northeastern Malaysia. *J Hematol Thrombo Dis.* 2014;2:140.
48. Borgna-Pignatti C, Garani MC, Forni GL, et al. Hepatocellular carcinoma in thalassaemia: An update of the Italian Registry. *Br J Haematol* 2014;167:121–126.
49. Ibrahim HA, Fouda MI, Yahya RS, et al. Erythrocyte Phosphatidylserine Exposure in β -Thalassemia. *J Blood Lymph.* 2013; 4:115.
50. Romano C, Massimo L, Tortorolo G. Preliminary note on the therapeutic use of desferrioxamine in Cooley's Disease. *Minerva Pediatr* 1964; 16:711–713.
51. Smith RS. Chelating agents in the diagnosis and treatment of iron overload in thalassemia. *Ann NY Acad Sci* 1964;119:776–788.
52. Saboor M, Qudsia F, Qamar K, et al. Levels of Calcium, Corrected Calcium, Alkaline Phosphatase and Inorganic Phosphorus in Patients' Serum with β -Thalassemia Major on Subcutaneous Deferoxamine. *J Hematol Thromb Dis.* 2014; 2:130.

53. Brittenham GM, Griffith PM, Nienhuis AW, et al. Efficacy of deferoxamine in preventing complications of iron overload in patients with thalassemia major. *N Engl J Med* 1994;331:567-573.
54. Hagag AA, Elfragy MS, Elfatah MA, et al. Comparative Study of Deferiprone and Silymarin versus Deferiprone and Placebo as Iron Chelators in Children with Beta Thalassemia with Iron Overload. *J Leuk (Los Angel)*. 2014; 2:130.
55. Grady RW. The development of new drugs for use in iron chelation therapy. *Birth Defects Orig Artic Ser* 1976;12:161–175.
56. Aydinok Y, Kattamis A, Viprakasit V. Current approach to iron chelation in children. *Br J Haematol* 2014;745–755.
57. Sheth S. Iron chelation: An update. *Curr Opin Hematol* 2014;21:179–185.
58. Link G, Konijn AM, Breuer W, et al. Exploring the “iron shuttle” hypothesis in chelation therapy: Effects of combined deferoxamine and deferiprone treatment in hypertransfused rats with labeled iron stores and in iron-loaded rat heart cells in culture. *J Lab Clin Med* 001;138:130–138.
59. Vlachodimitropoulou Koumoutsea E, Garbowski M, Porter J. Synergistic intracellular iron chelation combinations: mechanisms and conditions for optimizing iron mobilization. *Br J Haematol* 2015;170:874–883.
60. Piga A, Longo F, Musallam KM, et al. Assessment and management of iron overload in bthalassaemia major patients during the 21st century: A real-life experience from the Italian WEBTHAL project. *Br J Haematol* 2013;161: 872–883.
61. Olivieri NF, Nathan DG, MacMillan JH, et al. Survival in medically treated patients with homozygous beta-thalassemia. *N Engl J Med* 1994;331:574–578.
62. Puliyl M, Sposto R, Berdoukas VA, et al. Ferritin trends do not predict changes in total body iron in patients with transfusional iron overload. *Am J Hematol* 2014; 89:391–394.
63. Wood JC. Estimating tissue iron burden: current status and future prospects. *Br J Haematol* 2015; 170:15–28.
64. Saha S, Halder S, Bhattacharya D, et al. Fractional Precipitation of Plasma Proteome by Ammonium Sulphate: Case Studies in Leukemia and Thalassemia. *J Proteomics Bioinform*. 2012; 5:163-171.
65. Sarigianni M, Liakos A, Vlachaki E, et al. Accuracy of magnetic resonance imaging in diagnosis of liver iron overload: A systematic review and meta-analysis. *Clin Gastroenterol Hepatol* 2015; 13:55–63.e5
66. Duman H, Canatan D, Alanoglu G, et al. The Antioxidant Effects of Capparis Ovata and Deferasirox in Patients with Thalassemia Major. *J Blood Disord Transfus*. 2013; 4:142.
67. Eldor Lellouche F, Goldfarb A, et al. In vivo platelet activation in beta-thalassemia major reflected by increased platelet-thromboxane urinary metabolites. *Blood* 1991;77:1749–1753.
68. Cappellini MD, Robbiolo L, Bottasso BM, et al. Venous thromboembolism and hypercoagulability in splenectomized patients with thalassaemia intermedia. *Br J Haematol* 2000;111:467–473.
69. Cappellini MD, Poggiali E, Taher AT, Musallam KM. Hypercoagulability in b-thalassemia: A status quo. *Expert Rev Hematol* 2012;5:505–511.
70. Villeneuve PM, Turvey SL, Gourishankar S. A Case of Yersinia Enterocolitica Sepsis in a Beta Thalassemia Patient on Deferasirox. *J Clin Case Rep*.2015; 5:479.
71. Basu A, Panja A. Pharmacogenomics of the Drugs used for the Treatment of Thalassemia. *J Cytol Histol*. 2015; 6:360.

72. Klaassen RJ, Alibhai S, Allen MK, et al. Introducing the Tran Qol: A New Disease-Specific Quality of Life Measure for Children and Adults with Thalassemia Major. *J Blood Disord Transfus.* 2013; 4:150.
73. Gaziev J, Sodani P, Lucarelli G. Hematopoietic stem cell transplantation in thalassemia. *Bone Marrow Transplant* 2008;42:S41
74. Mathews V, Savani BN. Conditioning regimens in allo-SCT for thalassemia major. *Bone Marrow Transplant* 2014;49:607–610.
75. Kharbanda S, Smith AR, Hutchinson SK, et al. Unrelated donor allogeneic hematopoietic stem cell transplantation for patients with hemoglobinopathies using a reduced-intensity conditioning regimen and third-party mesenchymal stromal cells. *Biol Blood Marrow Transplant* 2014;20:581–586.
76. Gaziev J, Marziali M, Isgr_o A, et al. Bone marrow transplantation for thalassemia from alternative related donors: Improved outcomes with a new approach. *Blood* 2013;122:2751–2756.
77. Parikh SH, Mendizabal A, Benjamin CL, et al. A novel reduced-intensity conditioning regimen for unrelated umbilical cord blood transplantation in children with nonmalignant diseases. *Biol Blood Marrow Transplant* 2014;20:326–336.
78. Faulkner LB, Uderzo C, Masera G. International cooperation for the cure and prevention of severe hemoglobinopathies. *J Pediatr Hematol Oncol* 2013;35:419–423.
79. Amin M, Gholamhossein T, Majid N, et al. Prevalence of Alloimmunization against RBC Antigens in Thalassemia Major Patients in South East Of Iran. *J Blood Disord Transfus.* 2013; 4:147.
80. Mehta PA, Faulkner LB. Hematopoietic cell transplantation for thalassemia: A global perspective BMT tandem meeting 2013. *Biol Blood Marrow Transplant* 2013;19:S70–S73.
81. Ismahanisa W, Hassali MA, Yunus RM, et al. Complementary and Alternative Medicines (CAM) and its Role in Thalassemia Treatment. *Altern Integr Med.*2013; 3:e111.
82. Sankaran VG, Weiss MJ. Anemia: Progress in molecular mechanisms and therapies. *Nat Med* 2015;21:221–230.
83. Rivella S. bthalassemias: paradigmatic diseases for scientific discoveries and development of innovative therapies. *Haematologica* 2015;100:418–430.
84. Ngo DA, Steinberg MH. Genomic approaches to identifying targets for treating b-hemoglobinopathies. *BMC Med Genomics* 2015;8:44–57.
85. Musallam KM, Taher AT, Cappellini MD, Sankaran VG. Clinical experience with fetal hemoglobin induction therapy in patients with b-thalassemia. *Blood* 2013;121:2199–2212.
86. Bauer DE, Kamran SC, Lessard S, et al. An erythroid enhancer of BCL11A subject to genetic variation determines fetal hemoglobin level. *Science* 2013;342:253–257.
87. Dussiot M, Maciel TT, Fricot A, et al. An activin receptor IIA ligand trap corrects ineffective erythropoiesis in b-thalassemia. *Nat Med* 2014;20:398–407.
88. Nemeth E. Hepcidin and b-thalassemia major. *Blood* 2013;122:3–4.
89. Attie KM, Allison MJ, McClure T, et al. A phase 1 study of ACE-536, a regulator of erythroid differentiation, in healthy volunteers. *Am J Hematol* 2014;89:766–770.
90. Piga AG, Perrotta S, Melpignano A, et al. ACE-536 Increases hemoglobin and decreases transfusion burden and serum ferritin in adults with beta-thalassemia: Preliminary results from a phase 2 study. *Blood* 2014;124:53a.
91. Rivella S. Ineffective erythropoiesis and thalassemias. *Curr Opin Hematol* 2009;16:187–94.

92. Libani IV, Guy EC, Melchiori L, et al. Decreased differentiation of erythroid cells exacerbates ineffective erythropoiesis in beta-thalassemia. *Blood* 2008;112:875–885.
93. Rivella S. bthalassemias: paradigmatic diseases for scientific discoveries and development of innovative therapies. *Haematologica* 2015;100: 418–430.
94. Ramos P, Casu C, Gardenghi S, et al. Macrophages support pathological erythropoiesis in polycythemia vera and b-thalassemia. *Nat Med* 2013;19:437–445.
95. Manwani D, Bieker JJ. The erythroblastic island. *Curr Top Dev Biol* 2008;82:23–53.
96. Ginzburg Y, Rivella S. bthalassemia: A model for elucidating the dynamic regulation of ineffective erythropoiesis and iron metabolism. *Blood* 2011;118:4321–4330.
97. Jacobsen RN, Perkins AC, Levesque JP. Macrophages and regulation of erythropoiesis. *Curr Opin Hematol* 2015;22:212–219.
98. Arlet JB, Ribeil JA, Guillem F, et al. HSP70 sequestration by free α -globin promotes ineffective erythropoiesis in b-thalassaemia. *Nature* 2014;514:242–246.
99. Yuan J, Angelucci E, Lucarelli G, et al. Accelerated programmed cell death (apoptosis) in erythroid precursors of patients with severe betathalassemia (Cooley’s anemia). *Blood* 1993;82:374–377.
100. Ghosh S, Thrasher AJ, Gaspar HB. Gene therapy for monogenic disorders of the bone marrow. *Br J Haematol*, in press. doi: 10.1111/bjh.13520.
101. Cavazzana-Calvo M, Payen E, Negre O, et al. Transfusion independence and HMGA2 activation after gene therapy of human b-thalassaemia. *Nature* 2010;467:318–322.
102. Cavazzana M, Ribeil JA, Payen E, et al. Outcomes of gene therapy for b-thalassemia major and severe sickle cell disease via transplantation of autologous hematopoietic stem cells transduced ex vivo with a lentiviral beta globin vector. *Haematologica* 2014;99:S466.
103. Vikas U, Amit M, Mahavir Y, et al. RNA Silencing: An Approach for the Treatment of β -Thalassemia. *J Cell Sci Ther.* 2013; 3:135.
104. Finotti A, Breda L, Lederer CW, et al. Recent trends in the gene therapy of b-thalassemia. *J Blood Med* 2015;6:69–85.
105. Genovese P, Schirotti G, Escobar G, et al. Targeted genome editing in human repopulating haematopoietic stem cells. *Nature* 2014;510:235–240.
106. Modell B, Darlison M. Global epidemiology of haemoglobin disorders and derived service indicators. *Bull World Health Organ* 2008;86:480–487.