

**BETA THALASSEMIA (Autosomal Recessive Disorder)****Ayni Swaleh\*, Ruchira Chaudhary\*\* & Ragini Gothwal\*\*\***

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**ABSTRACT**

*$\beta$ -thalassemia is a diverse group of hereditary disorders in which there is a reduced rate of synthesis of the  $\beta$ -globin chains. Depending upon the genetic defects or deletion lies in transmission of  $\beta$ -globin chain, the body doesn't produce enough of the protein, the red blood cells become defective and cannot carry sufficient oxygen. The resulting anemia is usually severe with several health problems like enlarged spleen, bone deformities, fatigue and requires regular life-long transfusion, therapy and medical supervision. It is not only an important public health problem but also a socio-economic problem of many countries in the regions, it is important to take into consideration about this disorder as it may prove deadly one. Thus the intensity of this disorder can be lowered by diagnosing and taking proper treatments. The present review described the new approaches targeting on the specific mutation in the  $\beta$ -globin genes.*

**Key Words:**  *$\beta$ -Globin Chains, HBB gene, Haemoglobin,  $\beta$ -Thalassemia*

**INTRODUCTION**

$\beta$ -thalassemia is one of the most common monogenic disorders in the world, in  $\beta$ -thalassemia beta globin chain is effected, causing reduced or absent expression of the  $\beta$ -globin gene, leading to an imbalance of  $\alpha$  and  $\beta$ -globin chain. It is not contagious and cannot be passed from person to person by any contact or through blood transfusion.  $\beta$ -thalassemia is a major health problem, immensely affecting, psychological and economic burden on millions of people around the world. This disorder has more prevalence in countries which are located on thalassaemia band, tropical and sub-tropical areas, such as Mediterranean, Asia and India. It is an important disorder that has attracted the attention of medical research towards the various paradigmas of this multifaceted disease (Aessopos *et al.*, 2007).  $\beta$ -thalassemia have been encountered sporadically in every racial group (Sahu *et al.*, 2012). Patients with  $\beta$ -thalassemia on the basis of their  $\alpha$ -globin or  $\beta$ -globin chain imbalance have been classified as  $\beta$ -thalassemia major,  $\beta$ -thalassemia minor and  $\beta$ -thalassemia intermedia. Thalassemia major, also known as Cooley's anemia and Mediterranean anemia, is the most severe form of  $\beta$ -thalassemia, since mutations of both *HBB* alleles results in severely impaired  $\beta$ -globin chain production. In thalassemia major, the excess unpaired  $\alpha$ -globin chains aggregate to form inclusion bodies. These chain inclusion bodies damage RBC membranes, leading to intravascular hemolysis. In addition, there is damage and premature destruction of RBC precursors, causing ineffective erythropoiesis (Cao *et al.*, 2010).

$\beta$ -thalassaemia intermedia have intermediate degree of severity rather than major, that does not require regular blood transfusions. These cases are genetically heterozygous ( $B^0/B$  or  $B^0/B$ ). Patients with  $\beta$ -thalassemia intermedia have mild to moderate anemia and in most cases do not require blood transfusions (Musallam *et al.*, 2012). For instance, individuals with dominantly inherited  $\beta$ -thalassemia or inclusion body  $\beta$ -thalassemia clinically exhibit thalassemia intermedia (Taher *et al.*, 2006). This condition is milder than thalassemia major due to inheritance of a *HBB* mutation associated with reduced beta-globin chain production. Common clinical features include splenic enlargement due to entrapment of damaged RBCs, with risk of iron overload due in part to increased intestinal absorption. Although thalassemia intermedia can be associated with poor growth and bone abnormalities. Patients require regular monitoring because the clinical severity varies widely (Rund *et al.*, 2005).

While  *$\beta$ -Thalassemia minor* is a mild asymptomatic condition in which there is moderate suppression of  $\beta$ -chain synthesis (Raja *et al.*, 2012). It is most common form of  $\beta$ -thalassemia, and is also known as the 'thalassaemia trait', in which affected individuals are asymptomatic (Cao *et al.*, 2010). These subjects are typically heterozygous for  $\beta$ -thalassemia since they carry one normal *HBB* allele and one thalassemia allele either  $B^0$  or  $B^+$ . Asymptomatic patients are usually detected through routine hematologic testing, but in retrospect some newly diagnosed patients are observed to have mild anemia and small RBCs. The primary caution for individuals with thalassemia minor is a potential risk of having children affected with more serious thalassemia if their partner is also a carrier of thalassemia minor (Thein *et al.*, 2005). The  $\beta$ -globin chain is encoded by the *HBB* gene, which encodes an important blood protein called beta globin. A person with  $\beta$ -thalassemia carries a mutation in both copies of the *HBB* gene, completely halting production of the

betaglobin protein. Without beta globin, the important oxygen-carrying protein, hemoglobin, cannot be formed. Although oxygen can be carried by a less efficient form of hemoglobin (Ablhad 2013 and Balne 2013). The gene which spans 1.6 kb on the short-arm of chromosome 11, (11p15.4; MIM: 141900) contains three exons and two intervening sequences (IVS1 and IVS2) and the 5' and 3' untranslated regions (UTRs) (Cao *et al.*, 2010).

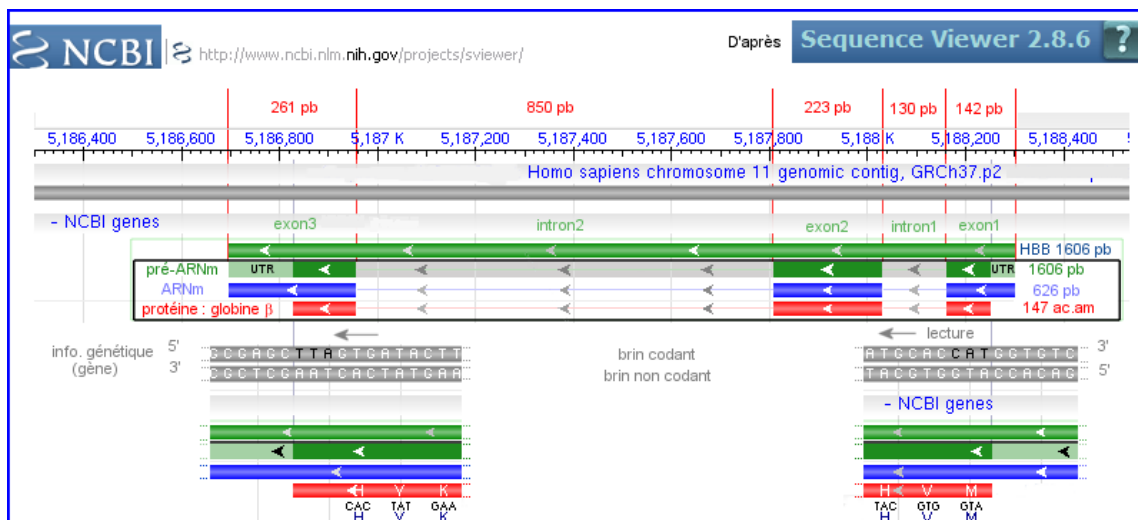


Fig1: Structural feature of the HBB gene, located on chromosome 11 with the position of exons and introns. (<http://jean-jacques.auclair.pagesperso.orange.fr/reticulocytes/gene%20HBB.htm>)

### Molecular genetics of $\beta$ -thalassemia

It has been found that  $\beta$ -thalassemia mutations are relatively population specific, most of the cases are inherited from parents where both have diseased alleles of the *HBB* gene. The combination of large population sizes and high levels of haemoglobinopathies create an issue of major public health concern, with an estimated 17 million  $\beta$ -thalassaemia carriers in India, ~8 million carriers in Pakistan, ~3 million in Bangladesh and ~0.5 million in Sri Lanka (Black *et al.*, 2010). Ten percent of the total world thalasseemics are born in India every year,  $\beta$ -thalassemias are also very heterogeneous at the molecular level, around 180 different mutations which lead to  $\beta$ -thalassemia have been identified and characterized around the world. In the Mediterranean region, over ~50  $\beta$ -thalassemia mutations have been characterized, in which IVS-I-110 (G→A) has high frequencies in the eastern part of the Mediterranean, while the mutation at codon 39 (C→T) is very frequent in western Mediterranean countries (Babu *et al.*, 2016). Some *HBB* alleles with deletion mutations can be common in certain ethnic groups [e.g., the 619-bp deletion in Asian Indians, commonly found *HBB* mutations in Indian population are [COD 8/9(+G), COD 15(G-A), IVS 1:1(G-T), IVS 1:5(G-C), COD 30(G-C), IVS 1:1(GA), COD 41/42 (- TCTT) & (COD 26(G-A))] (Kelkar *et al.*, 2017).

In most cases, mutations are single nucleotide substitutions, deletions, insertions or small oligonucleotides leading to frameshift. Their diversity and the consequent variable degree of globin chain imbalance are the main determinants for milder phenotypes, the coinheritance of homozygosity or compound heterozygosity for mild  $\beta$ -thalassemia alleles being responsible for a consistent residual output of beta chains from the affected  $\beta$ -globin locus (Danjou *et al.*, 2011). Diagnosis can be done by hematological testing and molecular genetic testing. The prevalence of a limited number of mutations in each population has greatly facilitated molecular genetic testing, the most commonly used methods are reverse dot blot analysis or primer specific amplification ARMS PCR, real-time PCR or microarray technology (Ye *et al.*, 2007). Sequence analysis detects mutations in the *HBB* coding region and associated flanking regions. Deletion/duplication analysis detects variable extent of the *HBB* gene or of the beta-globin gene cluster that result in  $\beta$ -thalassemia or in the complex  $\beta$ -thalassemias. To make a clear diagnosis of individual status (Grow *et al.*, 2014) the essential laboratory blood tests for hemoglobin analysis will be performed, including hemoglobin electrophoresis or currently updated technique of automated high performance liquid chromatography (HPLC) (Vanichsetakul *et al.*, 2011). In some cases requiring definite genotypes to be identified, blood tests for molecular assessment at particular globin genes can be conducted any times, regardless timing of blood transfusions. Diagnosis of  $\beta$ -thalassemia using blood tests, including a

Complete blood count (CBC) and special hemoglobin tests. Beta-lowers your RBC count and causes RBCs to be smaller than usual. By evaluating the size and shape of the RBCs present, also called the red cell indices the disease can be diagnosed. A recently added Red Cell Distribution Width Index (RDWI) provide valuable help to the attending physician. RDWI is more advantageous as all the discriminating factors including RBC count, MCV and RDW are incorporated in its formula (Jameel *et al.*, 2017).

### **Therapeutic Approaches**

Treatments for thalassemia depend on the type and severity of the disorder. Treatment for patients with thalassemia major includes Prenatal diagnosis, chronic blood transfusion therapy, iron chelation, splenectomy and bone marrow transplantation (Ghodekar *et al.*, 2010).

### **Prenatal diagnosis:**

In high-risk pregnancies in which both members are defined carriers for beta-thalassemia, prenatal diagnosis is possible by analysis of DNA extracted from fetal cells obtained by amniocentesis, usually performed at approximately 15–18 weeks' gestation, or chorionic villus sampling at approximately 10–12 weeks' gestation. Both disease-causing alleles must be identified before prenatal testing can be performed. If the known *HBB* mutation is present, analysis of globin chain synthesis is performed on a fetal blood sample obtained by percutaneous umbilical blood sampling at approximately 18–21 weeks' gestation (Cao 2010 and Galanello 2010).

### **Blood Transfusions:**

The goals of transfusion therapy are correction of anemia, suppression of erythropoiesis and inhibition of gastrointestinal iron absorption, which occurs in non transfused patients as a consequence of increased, although ineffective, erythropoiesis. The decision to start transfusion in patients with confirmed diagnosis of thalassemia should be based on the presence of severe anemia (Hb < 7 g/dl for more than two weeks, excluding other contributory causes such as infections). Different transfusional regimens have been proposed over the years, but the most widely accepted aims at a pretransfusion Hb level of 9 to 10 g/dl and a post-transfusion level of 13 to 14 g/dl. This prevents growth impairment, organ damage and bone deformities, allowing normal activity and quality of life (Galanello *et al.*, 2010). RBCs live for only about 120 days. So, repeated transfusions are needed to maintain a supply of healthy RBCs, because the hemoglobin in RBCs is an iron-rich protein, regular blood transfusions can lead to a buildup of iron in the blood. This condition is called iron overload. It damages the liver, heart and other parts of the body. To prevent this damage, iron chelation therapy is needed to remove excess iron from the body. Two medicines are used for iron chelation therapy. Deferoxamine is a liquid medicine that's given slowly under the skin, usually with a small portable pump used overnight. This therapy takes time and can be mildly painful. Responsible hematologist will assess and determine which ones are suitable to be used in patient on case by case basis (Vanichsetakul *et al.*, 2011).

### **Splenectomy:**

When the spleen becomes too active and starts to destroy the RBCs, transfusions become less effective. Then it becomes necessary to take the spleen out called "Splenectomy". Other indications for splenectomy are symptoms of splenic enlargement, leukopenia and/or thrombocytopenia and increasing iron overload despite good chelation (Galanello *et al.*, 2010). The susceptibility to overwhelming infections after splenectomy can be reduced by immunization with pneumococcal and meningococcal vaccines before splenectomy and antimicrobial prophylaxis with penicillin after splenectomy. Fever over 38° (101°F) developing in splenectomized patients with no focus of infection requires immediate intravenous broad-spectrum antibiotics (Sarker *et al.*, 2014).

### **Bone Marrow Transplant:**

A bone marrow transplant replaces abnormal or faulty cells with healthy ones from another person (a donor). However, BMT is only available for a minority of patients and bears a significant risk of mortality and morbidity, especially when the donor is unrelated. A definitive cure could only be achieved with bone marrow transplantation (BMT) from related or unrelated donors. In the search for a more general and definitive cure, hematologists have pursued alternative strategies aimed at correcting the defective beta-globin gene by either gene transfer of a normal beta-globin gene or substitution of the defective gene by homologous recombination (La Nasa *et al.*, 2005). However, concerns regarding gene transfer include the need for improved efficiency of gene delivery and mastery of vector stability, viral titers, nononcogenic insertion, the variable expression of globin genes, and the variable contributions of the  $\beta$ -thalassemia phenotype and

other modifiers to the effectiveness of gene transfer. Because of these complications the use of allogenic bone marrow is limited (Sarker *et al.*, 2014).

### Conclusion

From the above information it can be well known that the  $\beta$ -thalassemia is a dangerous disorder which is spreading worldwide, it is not only an important public health problem but also a socio-economic problem of many countries. As  $\beta$ -thalassemia is genetically derived disorder, genetic and cellular targets are potential approaches in management of disease. With the advancement of molecular genetics technique, most of the globin chain gene variants were understood and characterized. So, it is important to take into consideration about this disorder as it may prove deadly one. Thus the intensity of this disorder can be lowered by prenatal diagnosis genetic/marriage counseling, and by prenatal diagnosis of the disease by which proper treatments will be provided.

### REFERENCES

1. Aessopos .A, Kati. M, Farmakis .D, (2007): Heart disease in thalassemia intermedia: a review of the underlying pathophysiology *Haematologica*; 92(5): pp 658-65.
2. Ablahad, A.A, (2013): New Approach for Analysis and Prediction of Genetic Beta Thalassemia Mutations Based On Bioinformatics Bioedit Tools, *Inter Journal of Scientific & Engineering Research*, Issue 12 Vol.4, pp 245-249.
3. Cao, A, Galanello, R., (2010): Beta Thalassemia, *Genetics Medicine*, Vol.12, No.2, pp.61-76.
4. Babu, .S, Shantaram, T.V.M, (2016): "An Incidence of  $\beta$ -Thalassemia in South India-A Review" *2 International Journal of Research in Pharmacy and Biosciences V.3*, pp 1-6.
5. Balne, N.G and Ramachandra, C.S.V.R, (2013): "Role of XmnI restriction site polymorphism and JAK2 gene mutation in  $\beta$ -Thalassemia", *International Research Journal of Biological Sciences Vol. 2(1)*, pp 41-45.
6. Black M.L, Sinha S., Agarwal S., Colah.R, Das. R, Bellgard, M, Bittles, A.H. (2010): A descriptive profile of  $\beta$ -thalassaemia mutations in India, Pakistan and Sri Lanka, *Journal Community Genet 1*: pp 149-157.
7. Danjou, F, Franco, A, Galanello, R. (2010): Beta-thalassemia: from genotype to phenotype *hematologica*; 96:11 pp 1573-1574 2011 doi:10.3324/haematol.2011.055962.
8. Galanello Renzo and Raffaella Origa, Beta-thalassemia: Orphanet Journal of Rare Diseases, 5:11 pp 1-15.
9. Ghodekar, S.R, Grampurohit N.D, Kadam S.S, Thorat R.M. 2010 Thalassemia: A Review *International Journal of Pharma Research and Development*, Vol-2, pp 101-108/014.
10. Grow, K, Minakshi V, Pankaj A, Shiksha S, Ritu, Y. (2014): Beta thalassemia in India: current status and the challenges ahead, *International Journal of Pharmacy and Pharmaceutical Sciences*; 16 (4): pp 28-33.
11. Kelkar, A.J, and Moses, A. (2017): Thalassemia intermedia phenotype resulting from rare combination of c.46delT [Codon 15 (-T)] mutation of beta globin gene and HPFH3, *Clinical Case Reports by John Wiley & Sons Ltd* pp 1-4.
12. Jameel, T, Baig M, Ahmed, I, Hussain, M.B, Alkhamaly MD. (2017): Differentiation of beta thalassemia trait from iron deficiency anemia by hematological indices. *Pakistan Journal of Medical Science*; 33(3) pp 1-5:
13. La, N.G, Argioli, F, Giardini, C. (2005): Unrelated bone marrow transplantation for beta-thalassemia patients: The experience of the Italian Bone Marrow Transplant Group. *Annals of the New York Academy of Sciences*, 1054: pp 186-195.
14. Musallam, K.M, Taher, A.T, Rachmilewitz, E.A. (2012):  $\beta$ -thalassemia intermedia: A clinical perspective. *Cold Spring Harb Perspect Med*; 2: pp 1-15.
15. OMIM. (1986) Hemoglobin-Beta Locus; HBB. John Hopkins University.
16. Rund, D, Rachmilewitz, E. (2005): Beta-thalassemia, *New England Journal of Medical*; 353(11): pp 35-46.
17. Raja, J.V, Rachchh, M.A, Gokani, R.H. (2012): Recent advances in gene therapy for thalassemia. *Journal of Pharmacy and Bioallied Sciences*; 4:1 pp 94-201.
18. Sahu, P.K, Sudhanshu S.P, Kanti, S. (2012): Genotype-phenotype correlation of Beta-thalassemia spectrum of mutations in an Indian population *Hematology Reports*; vol 4:e9 pp 25-27.
19. Sarker, N.R, Ghosh A.K, Saha SK, Shariar, (2014) A, Recent advances in the management of Thalassemia: A Review Update, *Journal of Shaheed Suhrawardy Medical College*; Vol.6, No.1 pp 31-37.
20. Taher, A, Isma'eel, H, Cappellini, MD. (2006): Thalassemia intermedia: revisited, *Blood cells, molecules & diseases*; 37(1): pp 12-20.
21. Thein SL. (2005): Genetic modifiers of beta-thalassemia, *Haematologica*; 90(5): pp 649-60
22. Vanichsetakul, P. (2011): Thalassemia: Detection, Management, Prevention & Curative Treatment, *The Bangkok Medical Journal* pp 113-117.
23. Ye, BC, Zhang Z, Lei Z. (2007): Molecular analysis of alpha/beta-thalassemia in a southern Chinese population. *Genet Test*; 11: pp 75-83.
24. <http://jean-jacques.auclair.pagesperso-orange.fr/reticulocytes/gene%20HBB.htm>.