From Transfusions to Transformation: Assessing Gene Therapy against Conventional Treatments for Thalassemia

Naisha Vishal Chhabria, Bombay Scottish School, Mahim

Abstract

Thalassemia is an autosomal recessive hemoglobinopathy that results in severe anemia and organ damage, affecting up to 400 million people worldwide. Alpha thalassemia major and beta thalassemia major are the most severe forms of thalassemia. Alpha thalassemia major is fatal before birth, while beta thalassemia major leads to fatality during childhood. Conventional treatments for thalassemia have many drawbacks and can only manage the symptoms without curing the disorder. Gene therapy has recently emerged as a promising treatment for thalassemia, which could serve as a lifelong cure. Some gene therapies aim to cure beta thalassemia by inserting a gene construct for β-globin production, and others by gene editing to reactivate fetal hemoglobin (HbF) synthesis. Some of these therapies have even been approved to treat the disorder. This paper evaluates conventional treatments for thalassemia and examines the potential of gene therapy as a new treatment approach.

Introduction

Thalassemia is an inherited blood disorder characterized by the body's inability to produce adequate amounts of hemoglobin, the protein in red blood cells that carries oxygen (1). The condition leads to chronic anemia, fatigue, growth delays, and complications related to iron overload, such as organ damage, and in the most severe forms, death (1). It results from mutations in the genes responsible for hemoglobin production, primarily affecting the α-globin or β-globin chains (2). There are two main types of thalassemia, categorized by mutation types. Alpha thalassemia is caused by deletion(s) in the α-globin genes, while beta thalassemia is caused by mutation(s) in the β-globin gene (3). The severity of thalassemia can range from mild (requiring little to no treatment) to severe (requiring lifelong medical care or fatal), depending on the type of mutation (3). The severity is determined by both the type of mutations and the number of hemoglobin chains affected (3). Thalassemia is most prevalent in regions where malaria was historically common, including parts of South Asia, the Mediterranean, and the Middle East, as the carrier state (thalassemia trait) provides some protection against malaria (4,5).

Conventional thalassemia treatments have several limitations and are usually a lifelong burden. Until recently, thalassemia management has relied heavily on supportive care, aiming to control symptoms and prevent or address treatment-related complications. Some of these treatments are regular blood transfusions, iron chelation therapy, and bone marrow/stem cell transplantation (6). Patients with severe thalassemia require blood transfusions every 2–4

weeks to maintain hemoglobin levels (3). While lifesaving, this approach leads to iron overload, necessitating additional treatment. Iron build-up in the body from repeated transfusions leads to damage in the heart, liver, and endocrine glands (7). Therefore, iron chelation therapy is often required to address the problem posed by regular blood transfusions. For chelation therapy, patients are prescribed drugs that help remove excess iron in the blood, such as deferoxamine, deferasirox, or deferiprone. These drugs can bind to iron and facilitate their excretion from the body via urination. However, adherence can be challenging (7). Bone marrow and stem cell transplantation is the only established cure for thalassemia (8). This procedure replaces defective hematopoietic stem cells with healthy ones from a compatible donor. However, it carries risks such as graft-versus-host disease, which is when the donor immune cells recognize the recipient cells as foreign and attack them, and is limited by donor availability (8). Despite advances in supportive care, most treatments do not address the underlying genetic cause, and many patients remain dependent on lifelong interventions.

Gene therapy represents a paradigm shift in the treatment of thalassemia. By targeting the genetic root of the disease, the technique offers the potential for a long-term or permanent cure. The two main approaches under clinical investigation are gene addition therapy and gene editing (CRISPR/Cas9) therapy. Gene addition therapy involves introducing a functional copy of the β-globin gene into a patient's hematopoietic stem cells using viral vectors (such as lentiviruses) (9,10). An example is betibeglogene autotemcel (Zynteglo), approved in some regions, which has shown promising results in reducing or eliminating the need for transfusions in many patients (9). On the other hand, gene editing (CRISPR/Cas9) modifies specific mutated DNA sequences within the patient's cells (11). For thalassemia, CRISPR is being used to reactivate fetal hemoglobin (HbF) production by disrupting regulatory genes like BCL11A. The restoration of HbF compensates for the defective adult hemoglobin, potentially curing the disease without the need to insert new genes (11). Both strategies involve collecting a patient's stem cells, genetically modifying them in a lab, and reinfusing them after chemotherapy to clear space in the bone marrow (9,10,11). While gene therapy offers hope, challenges remain, including high costs, long-term safety and monitoring, and access in low-resource settings. Nevertheless, with continued research and broader implementation, gene therapy may soon become the new standard of care for thalassemia.

This review paper examines various potential gene therapy treatments for thalassemia, analyzing their efficacy and comparing them to existing conventional treatments. Different types of thalassemia, categorized by their specific genetic mutations, will be examined alongside the relevant treatment approaches and their effectiveness for varying severities of the disorder. The information in this paper will be supported by various scientific papers, studies, and clinical trials, utilizing statistics for patients of diverse age ranges and geographic locations. Based on this information, the paper will discuss how these gene therapies can be further developed to

benefit patients.

Thalassemia

The thalassemias are a group of autosomal recessive inherited conditions characterized by decreased or lack of synthesis of one of the two polypeptide chains (α or β) that form the normal adult human hemoglobin molecule (5,12). This results in reduced hemoglobin levels in red blood cells and ultimately anemia. The term "thalassemia" is derived from the Greek words "thalassa" (θάλασσα), meaning "sea", and "-emia" (αιμία), which is a suffix meaning "blood" (13). It was first used in 1932 and is named for its initial association with populations living near the Mediterranean Sea, where the condition was first described (13). Approximately 5% of the world population is affected by some mutation of the thalassemia-causing genes. Thalassemia affects almost 4.4 per 10,000 live births worldwide and accounts for almost 3.4% of deaths in children under 5 years old (11). The thalassemia syndromes are named according to the globin chain affected or the abnormal hemoglobin involved. β-globin gene defects may give rise to beta thalassemia, while mutations of the α-globin gene may cause alpha thalassemia (5,14). Alpha thalassemia is more prevalent in Southeast Asia, Africa, and India (5). Beta thalassemia is more prevalent in the Mediterranean, the Middle East, Central, South, and Southeast Asia, and southern China regions (5). These are areas where malaria was, and still is, epidemic, which has led to the theory that thalassemia first evolved as a genetic survival advantage against malaria (15). Due to migration of populations carrying the mutation to non-endemic countries, the disorder has spread worldwide (5,14).

Alpha thalassemia is characterized by reduced or absent production of α-globin chains, essential components of hemoglobin, due to deletions of one or more of the two genes responsible for the production of α-globin chains present on each copy of chromosome 16, *HBA1* and *HBA2* (12). The deletion leads to an imbalance in the α-β-globin ratio, resulting in various clinical manifestations. When just one gene is deleted, individuals are silent carriers and remain asymptomatic with normal bloodwork. The deletion of two genes leads to the alpha thalassemia trait (minor), characterized by microcytosis (abnormally small red blood cells) and minimal or no anemia (12). A three-gene deletion causes HbH disease (alpha thalassemia intermedia), resulting in moderate to severe microcytic anemia, hemolysis, and splenomegaly due to excess β-globin tetramers (12). Finally, deletion of all four genes causes Hb Bart's (alpha thalassemia major), where unstably high levels of gamma-globin tetramers in utero typically result in fatal hydrops fetalis (14). Beta thalassemia results from the deficient or absent synthesis of β-globin chains, leading to an excess of α-chains. β-globin synthesis is controlled by one gene on each chromosome 11 (12). Beta thalassemia occurs from any of more than 200 point mutations and (rarely) deletions of the two genes (12). β-globin production can vary from

normal to completely absent, causing varying severities of the disorder. A one-gene mutation leads to beta thalassemia trait (minor), typically asymptomatic except for microcytosis and mild anemia. Severe mutations affecting both genes cause beta thalassemia major, also known as Cooley's anemia (13). Symptoms are rarely present at birth due to the presence of fetal hemoglobin (HbF), but they begin to develop by six months of age as the infant's body switches from producing HbF to HbA (12, 16). People with beta thalassemia major experience severe anemia and splenomegaly and require regular blood transfusions (12). They have a life expectancy of 17 years and usually die by 30 years (12). If the synthesis of β-chains is less severely reduced, the person has beta thalassemia intermedia. They experience less severe symptoms and do not require lifelong transfusions to survive past 20 years of age (17). In some cases, a patient can have co-inheritance of both alpha and beta thalassemia. In these cases, patients have a milder clinical course due to a less severe α-β chain imbalance (18).

Considering the widespread impact of this disorder, it is paramount to find effective treatments for this condition.

Conventional Treatments for Thalassemia

Thalassemia treatments vary widely based on disease severity and associated complications (Figure 1). People with thalassemia minor typically require little to no treatment (18). Some may require blood transfusions after surgery or childbirth (18). Folic acid supplementation (especially in children) can help to alleviate symptoms like fatigue and muscle weakness by increasing red blood cell production, thus treating mild anemia (19,20). Monitoring lifestyle and diet is sufficient care for thalassemia minor.

Regular blood transfusion is a treatment that has transformed thalassemia major from a fatal childhood disease into a chronic disorder (21). It is the most common treatment for transfusion-dependent thalassemia (TDT), typically in patients with alpha or beta thalassemia major or severe cases of HbH disease (8). Although it is not a routine treatment for patients with thalassemia intermedia, it can have several benefits (6). Blood transfusions provide functional red blood cells to maintain adequate hemoglobin levels in the blood plasma (22). This corrects anemia, which results from ineffective endogenous erythropoiesis (22,23). Blood transfusion therapy is often started after confirmation of the diagnosis of thalassemia major (23). Regular blood transfusions inhibit retardation of growth, bone malformations, and organ impairment, while also improving oxygen transport (21, 23). However, repeated transfusions increase the risk of alloimmunization (the body's production of antibodies against the antigens from the donor individual's blood), and other transfusion-related infections and reactions, including iron overload (14,24). If transfusions are begun in adulthood or late childhood, there is a much greater risk of alloimmunization and delayed hemolytic transfusion reactions (DHTRs) (21).

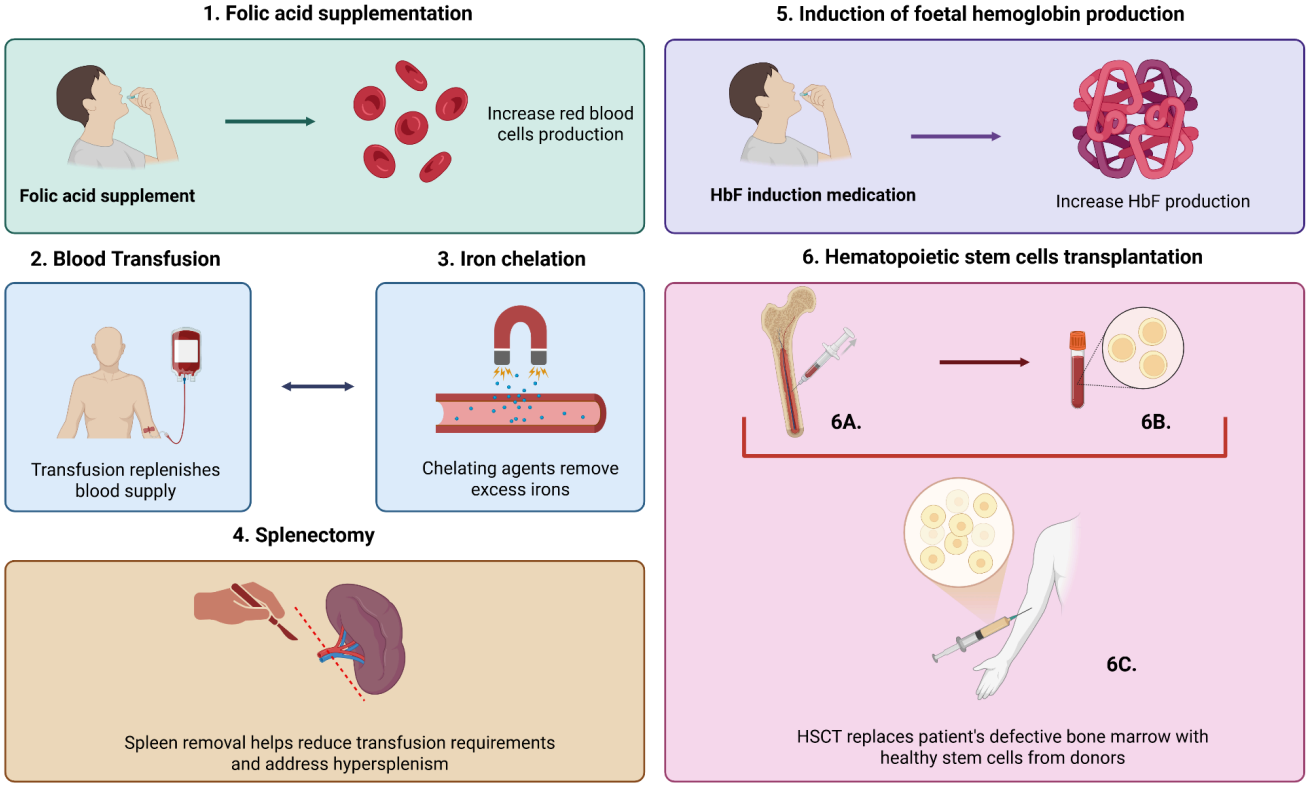

Figure 1. Treatment for thalassemia minor includes folic acid supplementation (1). Treatment for thalassemia intermedia includes blood transfusions (2), iron chelation (3), splenectomy (4), and inducing fetal hemoglobin production (5). Treatment for thalassemia major includes blood transfusions (2), iron chelation (3), splenectomy (4), inducing fetal hemoglobin production (5), and hematopoietic stem cell transplantation (6). ***(1)*** *Folic acid supplementation helps increase red blood cell production and alleviate symptoms like fatigue and muscle weakness.* ***(2)*** *Blood transfusions provide functional red blood cells to maintain adequate hemoglobin levels in the blood plasma. Regular blood transfusions inhibit retardation of growth, bone malformations, and organ impairment and improve oxygen transport.* ***(3)*** *Iron chelation corrects iron overload caused by frequent blood transfusions and thus prevents organ damage resulting from iron overload.* ***(4)*** *Splenectomy is done to treat splenomegaly, which is a symptom of thalassemia that can cause growth retardation and worsening anemia, and various other mechanical disturbances.* ***(5)*** *Inducing fetal hemoglobin (HbF) production is done by oral consumption of drugs like hydroxyurea. HbF has a higher affinity for oxygen than adult hemoglobin, and it may raise Hb levels in transfusion-independent thalassemia and may lower the need for blood transfusions in transfusion-dependent patients.* ***(6)*** *Hematopoietic stem cell transplantation is one of the only widely available curative treatments for thalassemia major. A healthy HLA-matched donor's bone marrow is taken (6A),*

hematopoietic stem cells are sequestered from it (6B), and the extracted stem cells are infused into the patient (6C). It replaces the patient's defective bone marrow with healthy donor marrow.

Iron chelation therapy using drugs such as deferoxamine, deferasirox, and deferiprone corrects iron overload caused by frequent blood transfusions in TDT (14). It removes excess iron from the blood by binding it for excretion via urine or feces (25). This prevents organ damage that can result from excess iron in the body (26). Retrospective observational studies have reported that iron chelation therapy has been associated with lower mortality, especially cardiac-related mortality in patients with TDT (14). This is because chelation therapy prevents cardiac siderosis (intracellular iron deposition in the ventricles and atria of the heart), which causes arrhythmias and heart failure (7,27). Mortality is also greatly reduced when chelation therapy is started at a young age (14).

Transplantation of hematopoietic stem cells (HSCT) is currently one of the only widely available and established, long-term curative treatments for thalassemia major (28). In this treatment, hematopoietic stem cells from healthy individuals are sequestered and transferred to thalassemia patients (23). It replaces the patient's defective bone marrow with healthy donor marrow. Stem cell transplantation is typically carried out using the stem cells of a human leukocyte antigen (HLA) matched sibling donor (29,30). The use of the term hematopoietic stem cells came about with the advent of the use of peripheral blood stem cells (PBSCs) from a donor instead of bone marrow cells (30). However, the risk of developing severe graft versus host disease (GVHD) is highest when PBSCs are donated (31). GVHD is the biggest risk of allogeneic HSC transplantation, with incidences between 20% and 40%, and is a major cause of transplant-related morbidity and mortality (32,33). This happens when the donated cells (graft) view the recipient's cells (host) as threatening or foreign and thus begin to attack them. It typically occurs when the HLAs of the donor and the recipient are too different (32,34,35). GVHD can affect various organs in the body, with acute or chronic symptoms, and can lead to long-term damage (32).

Splenectomy is a common treatment for splenomegaly, a symptom of thalassemia (23). Increased destruction of red blood cells by the reticuloendothelial system, in particular the spleen, along with extramedullary hemopoiesis, results in splenic enlargement (splenomegaly) (36). Hypersplenism may cause growth retardation and worsening anemia and various other mechanical disturbances due to splenomegaly, thus necessitating splenectomy in thalassemia intermedia patients (23). In the case of patients with transfusion-dependent thalassemia major, the purpose of splenectomy is to reduce blood consumption, which reduces transfusion requirements with the ultimate goal of reducing iron overload (36). Splenectomy should be avoided in children below the age of five years due to the high risk of post-operation sepsis, which has a 50% mortality rate (36). Removal of the spleen has been associated with an increase in susceptibility to various infections and an increase in mortality. Therefore, patients are often required to take routine anti-pneumococcal immunisation and prophylactic antibiotics

in the critical 2-4 years after the procedure, or in some cases, for life (36,37).

Inducing the production of foetal hemoglobin (HbF) is an effective treatment for thalassemia intermedia and thalassemia major (23,38). HbF is the predominant form of hemoglobin expressed throughout gestation, and it has a higher affinity for oxygen than adult hemoglobin (HbA) (39). The drug most commonly used for this purpose is hydroxyurea (HU) (24,39). The exact mechanism of HU to induce HbF production is unknown (39). Hydroxyurea is beneficial for thalassemia patients as it may raise Hb levels in transfusion-independent thalassemia and may lower the need for blood transfusions in transfusion-dependent patients (40). The primary adverse side effects of HU are leucopenia and neutropenia, but relatively low incident rates of the same let us conclude that HU is safe and well-tolerated by thalassemia patients (38,40).

As demonstrated above, most of the conventional treatment methods for thalassemia do not provide a definitive cure and can pose serious risks and adverse effects. Thus, novel gene therapy approaches are gaining prominence and proving to be effective in the treatment of this disorder.

Gene Therapy

Gene therapy is a technique that allows for editing of genetic materials with the potential to act as a treatment approach for genetic diseases via correction and modification of altered (mutated) genes (41). These therapies have become possible through advances in genetics and bioengineering, which have enabled the manipulation of vectors for the delivery of extrachromosomal materials to target cells (41). Many gene therapy treatments for thalassemia are currently being examined in preclinical and clinical studies, with a small percentage approved for use in the United States, Europe, and Australia (41). Most of these therapies target hematopoietic stem cells (HSCs) since they are the source for red blood cells, and correction of HSCs could potentially provide lifelong correction during their differentiation into daughter cells. HSCs can be extracted from the patients and corrected via gene therapy *ex vivo* before being delivered back into the patients (42). The editing component of the therapy can be delivered to these cells either via transduction using lentiviral vectors or transfection with non-viral methods (42). In this section, some existing therapies will be examined.

Zynteglo: Betibeglogene Autotemcel or beti-cel is a one-time βA-T87Q-globin gene addition therapy being assessed as a potentially curative mode of gene therapy for people with transfusion-dependent beta thalassemia major with the goal of achieving transfusion independence (43). Patients go through autologous mobilization and harvesting of hematopoietic stem cells, which are transduced *ex vivo* with self-inactivating BB305 lentiviral vectors that insert a gene construct for the β-globin gene and the associated regulatory elements (43). Post *ex vivo* addition, these cells are reintroduced back into the patient, where they replicate and repopulate the blood (43).

In July 2016, Locatelli et al. initiated an open-label, single-dose, phase 3 study to evaluate the efficacy and safety of in adult and pediatric patients with transfusion-dependent beta thalassemia and a non–β0/β0 genotype (44). Post myeloablation with busulfan, patients received beti-cel intravenously. A total of 14 of 15 evaluable patients who were 12 to 50 years of age (93%), as well as 6 of 7 patients younger than 12 years of age (86%), achieved transfusion independence (defined as a weighted average hemoglobin level of 9 g per deciliter or greater in patients who did not receive packed red-cell transfusions for at least 12 months at any time after beti-cel infusion). All patients had at least one adverse event during or after beti-cel infusion, with all being alive at the last follow-up. Most of these were consistent with typical adverse effects following myeloablation with busulfan. Three patients had grade 4 serious hepatic veno-occlusive disease, and one patient had grade 2 nonserious hepatic veno-occlusive disease. Two patients had events of grade 3 thrombocytopenia that the investigators considered to be possibly related to beti-cel. After the infusion of beti-cel, assessments of bone marrow and blood biomarkers during transfusion independence showed improvement in erythropoiesis. The two patients who did not achieve transfusion independence had lower circulating vector copy numbers than those in patients who had transfusion independence. It is possible that the long-term hematopoietic stem cells infused were not adequately transduced, since the population is heterogeneous and transduction is not uniform (44).

As of February 2024, in a similar phase 3 study of Zynteglo by Olson et al., 37/41 patients achieved transfusion independence, and transduction efficiency, pharmacodynamics, transfusion independence rate, and weighted average Hb were similar across genotypes (β0/β0 vs non-β0/β0) and ages (45). This study shows that Zynteglo is also effective in β0/β0 patients. However, Zynteglo is restricted to treating beta thalassemia only. It has been granted conditional marketing authorization in the European Union and has been approved by the FDA (46,47).

BD211 Lentiviral Gene Therapy has a similar approach to Zyntelo, with the goal of one-time addition of βA−T87Q-globin gene for beta thalassemia that uses an optimized lentiviral vector system to deliver the gene into patient-derived hematopoietic stem cells. The product has distinctive technological advantages, including a proprietary insulator design integrated into the lentiviral vector architecture to minimize potential insertional mutagenesis risks and an optimized hemoglobin subunit beta (HBB) gene expression cassette engineered to achieve physiological hemoglobin levels comparable to those observed in healthy individuals. In preclinical studies by Dai et al., CD34+ hematopoietic stem cells were isolated from the peripheral blood of donors following Filgrastim mobilization and were transplanted into Specific pathogen-free (SPF) grade NCG-X mice non-transduced along with the BD211 cells. It resulted in vivo erythroid differentiation of stem cells. BD211 successfully differentiated into various stages of human erythroid cells in vitro and expressed βA−T87Q-globin during the directed differentiation process (48).

In 2021-2022, Li et al. conducted a single-center, single-arm pilot trial evaluating a β-globin

expression-optimized and insulator-engineered lentivirus-modified cell product in the most severe beta thalassemia (β0/β0 TDT) (49). The therapy rebalanced β-globin and α-globin chains in patients who achieved transfusion independence for nearly two years without any notable adverse effects. Two female children were enrolled, infused with BD211, and followed up for an average of 25.5 months. Engraftment of genetically modified hematopoietic stem and progenitor cells was successful and sustained in both patients. No unanticipated safety issues occurred during conditioning or after infusion. No hepatic toxicity or veno-occlusive disease was observed. Both experienced neutropenia and thrombocytopenia for brief periods. Myeloablation-related toxicity was mild, and no serious infections were observed. Both patients achieved transfusion independence for the 25.5-month follow-up. The treatment extended the lifespan of red blood cells by over 42 days. No notable adverse side effects were shown by single-cell DNA/RNA sequencing analysis of the dynamic changes of gene-modified cells, transgene expression, and oncogene activation. Optimized lentiviral gene therapy may be a safe and effective treatment for all β-thalassemia. However, the study was limited to only two patients for a short period of about two years. Longer studies with larger cohorts must be conducted to reach a decisive conclusion about BD211 lentiviral gene therapy for transfusion-dependent beta thalassemia (49).

Exagamglogene Autotemcel (exa-cel/Casgevy) is a nonviral cell therapy designed to reactivate fetal hemoglobin (HbF) synthesis through *ex vivo* clustered regularly interspaced short palindromic repeats (CRISPR) gene editing of the erythroid-specific enhancer region of BCL11A in autologous CD34+ hematopoietic stem and progenitor cells (HSPCs) in beta thalassemia (49). Exa-cel utilizes CRISPR-Cas9 technology to suppress the expression of BCL11A by disrupting an erythroid-specific enhancer of BCL11A. By inhibiting the expression of BCL11A, which serves as a repressor for HbF production, exa-cel enables the resurgence of HbF synthesis. HbF can then compensate for the defective β-globin and restore functional tetrameric hemoglobin complexes along with α-globin in individuals with beta thalassemia (50).

In 2018, Locatelli et al. initiated an open-label, single-group, single-dose, phase 3 study of exa-cel in patients 12 to 35 years of age with transfusion-dependent beta thalassemia and a β0/β0, β0/β0-like, or non–β0/β0-like genotype (51). 52 participants underwent Casgevy through gene editing of CD34+ HSPCs using CRISPR-Cas9 with a guide mRNA. Before the exa-cel infusion, they underwent myeloablation with busulfan. Of these, 35 had evaluable results, of which 32 achieved transfusion independence with a mean duration of 20.5 months, and some patients remained transfusion-free for as long as 40.7 months. After the infusion, there were early and sustained increases in total hemoglobin and fetal hemoglobin levels. The patients who achieved transfusion independence had a mean total hemoglobin level of 13.1±1.4 g per deciliter and a mean fetal hemoglobin level of 11.9±1.9 g per deciliter, with a pancellular distribution of at least 94% of red cells expressing fetal hemoglobin from month 6 onwards. All patients had at least one adverse event after exa-cel infusion. The most serious adverse side

effect was veno-occlusive liver disease attributed to busulfan conditioning, which occurred in five patients. Serious adverse events that were considered by the investigators to be related to exa-cel occurred in two patients. One patient experienced headaches, acute respiratory distress syndrome, and idiopathic pneumonia syndrome, and the second had delayed engraftment and thrombocytopenia, all of which resolved and were also considered to be related to busulfan. Other serious adverse side effects were considered by investigators to be related to busulfan conditioning and not to exa-cel, and no malignancies were observed (51).

In November 2023, the Medicines and Healthcare products Regulatory Agency (MHRA) granted conditional approval to Casgevy (exagamglogene autotemcel; Vertex Pharmaceuticals) for the treatment of transfusion-dependent beta thalassemia intermedia and major in patients 12 years and older for whom a human leukocyte antigen-matched related hematopoietic stem cell (HSC) donor is appropriate, but not available (52). Casgevy is a potentially revolutionary novel therapeutic option for patients suffering from hemoglobinopathies and is the first treatment worldwide to gain regulatory approval involving genomic editing using the CRISPR-Cas9 system in humans (52). Vertex is initiating a comprehensive 15-year follow-up study focusing on malignancies, mortality, and other health outcomes to monitor the safety profile of exa-cel, as part of a post-approval study for it (50). However, with a cost of $2.2 million per patient, its accessibility and affordability by the general population at this moment are a concern (50).

Discussion

As previously discussed, conventional treatments for thalassemia usually remain a lifelong burden and do not cure the underlying genetic cause of the disease. Hematopoietic stem cell transplantation is an effective, curative treatment for beta thalassemia major, but it is limited by a lack of suitable donors and a high risk of graft-versus-host disease (GVHD). These limitations have paved the way for gene therapy, which is considerably more promising, as it can provide curative solutions without being restricted by HLA-matching and GVHD risks. As mentioned above, Zynteglo and Casgevy are both gene therapy approaches for beta thalassemia, which have been approved for use, while BD211 Lentiviral gene therapy has a lot of potential but requires further study. Current preclinical and clinical data indicate great potential for all these approaches to replace conventional approaches to treating thalassemia.

However, further research and improvements are needed. Gene therapy is a complicated technique; therefore, there should be further optimization to improve its accessibility and throughput for application in clinical settings. At the moment, the high cost of these treatments has restricted their mainstream use and made them inaccessible to the general population. Additionally, these therapies are not available in developing nations due to high cost and insufficient infrastructure, and a lack of experienced personnel for administration and regulation. The global focus of gene therapy research has predominantly been in high-income countries, leading to therapies that may not always be optimized for the specific needs or disease burdens

in low and middle-income countries. This is especially concerning because diseases like thalassemia and sickle cell anemia, which have potential gene therapy cures, are more prevalent in these countries.

Most gene therapy treatments for thalassemia are engineered for the treatment of transfusion-dependent beta thalassemia major. In the case of alpha thalassemia major, most patients die in utero due to hydrops fetalis, and there is a small chance of survival to birth if intra-uterine blood transfusions are performed. However, these patients still have a minuscule chance of survival to adulthood. Additional research is required for intra-uterine (*in-utero*) gene therapy approaches so that these fetuses and infants have a chance of survival. Such a novel approach could potentially eliminate fatalities resulting from alpha thalassemia major. In addition, in-utero gene therapy treatment could also benefit beta thalassemia major patients, as this is a preventative therapy that reduces the need for further treatment.

In the case of existing beta thalassemia gene therapy treatments, the long-term effects of these treatments over a person's lifespan are still being documented and have not reached a definitive conclusion, even though these treatments have been approved for administration. Long-term 15-year studies for monitoring and documenting the effects of Zynteglo and Casgevy are still underway. This may be a matter of concern as long-term data regarding malignancies, mortality, autoimmune-like reactions, and other health outcomes are still not available. More research must also be done in this regard. Gene therapy treatments also require myeloablative busulfan conditioning before they can be administered. Busulfan conditioning poses serious risks and side effects, so an alternative that is associated with less risk than busulfan also needs to be found.

Conclusion

Gene therapy marks a transformative advancement in the treatment of thalassemia, offering the potential for a lifelong cure rather than lifelong management. Current approaches, such as Zynteglo, BD211 Lentiviral gene therapy, and Exa-cel, have demonstrated promising outcomes in achieving transfusion independence and restoring effective hemoglobin production in patients with beta thalassemia. However, these therapies remain limited by high costs, restricted accessibility in low-resource settings, and the need for myeloablative conditioning, which carries significant risks. Furthermore, the focus on beta thalassemia highlights a critical gap in research and treatment development for alpha thalassemia, particularly the fatal Hb Bart’s hydrops fetalis. To fully realize the promise of gene therapy, future efforts must prioritize cost reduction, long-term safety monitoring, equitable global access, and the development of prenatal and in-utero interventions. Only then can gene therapy become a universally viable and inclusive cure for all forms of thalassemia.

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