OTHER

Review

Advancing the care of ß-thalassaemia patients with novel therapies

Rayan Bou-Fakhredin[1], Irene Motta[2], Maria Domenica Cappellini[1,2]

[1]*Department of Clinical Sciences and Community Health, University of Milan, Milan, Italy;*
[2]*Department of Internal Medicine, Fondazione IRCCS Ca' Granda, Ospedale Maggiore Policlinico, Milan, Italy*

The β-thalassaemias are a group of inherited disorders of haemoglobin synthesis characterised by chronic anaemia of varying severity. Currently available conventional therapies in thalassaemia have many challenges and limitations. A better understanding of the pathology of β-thalassaemia has led to the development of new treatment options, most of which are currently in clinical trials. These could have the potential of reducing red blood cell transfusion burden, raising haemoglobin levels, and improving patients' overall quality of life. In this review, we will provide an overview of the novel therapeutic approaches that are currently under development to advance the care of β-thalassaemia patients.

Keywords: *β-thalassaemia, clinical trial, novel therapies, quality of life, transfusion burden*.

INTRODUCTION

The thalassaemias constitute one of the most common inherited monogenic diseases in the world. They are characterised by autosomal recessive inherited defects in the production of haemoglobin (Hb). Today, thalassaemia patients are commonly categorised according to their need for blood transfusion. Transfusion-dependent thalassaemia (TDT) patients frequently present with severe anaemia in early childhood and their survival depends on lifelong treatment with transfusions. Non-transfusion-dependent thalassaemia (NTDT) patients commonly present with mild to moderate anaemia in a later stage of childhood, or even in adulthood, and may require occasional or short-course transfusions for the management or prevention of some disease manifestations[1,2]. Over the last decade, the improved management of symptoms and associated comorbidities for β-thalassaemia patients via regular red blood cell (RBC) transfusions, iron chelation therapy (ICT), and supporting therapies has allowed more patients to remain healthy and move into adulthood[3,4]. However, unmet needs in terms of safety, efficacy and adherence to conventional therapies have posed many challenges, limitations and an additional burden not only to the patients themselves, but also to their families[5]. Moreover, advances in the understanding of the disease itself has enabled clinicians and researchers to move forward towards the development of novel therapeutic modalities[6,7]. These can

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Correspondence: Maria Domenica Cappellini
e-mail: maria.cappellini@unimi.it

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be classified into three major categories based on their efforts to address different features of the underlying pathophysiology: correction of the α/β-globin chain imbalance, targeting ineffective erythropoiesis, and targeting iron dysregulation[7] (**Figure 1**). In this review, we will provide an overview of the novel therapeutic approaches that are currently in development to advance the care of β-thalassaemia patients.

CORRECTION OF THE α/β-GLOBIN CHAIN IMBALANCE

Allogeneic haematopoietic stem cell transplantation

Allogeneic haematopoietic stem cell transplantation (HSCT) from a human leukocyte antigen (HLA)-matched sibling donor (MSD), performed in childhood, has been the gold standard for TDT patients for decades[8,9]. Unfortunately, only a minority of patients have siblings as potential donors. Therefore, fully matched unrelated donors (MUD) have been the second choice for cure, with similar results in terms of overall survival (OS). Many key studies on HSCT for TDT patients using MUDs have been conducted, with efforts focused on better engraftment, prevention of graft rejection and toxicity, and targeting conditioning regimens[10-15]. Results from a recent international retrospective study of 1,110 TDT patients, aged ≤25 years, who received grafts from HLA-matched related (61%), HLA-mismatched related (7%), HLA-matched unrelated (23%), and HLA-mismatched unrelated (9%) donors between 2000 and 2016 showed that the highest OS was seen in patients aged ≤6 years. There was no significant difference in OS rates between the groups transplanted from MSDs or MUDs (89 *vs* 87% and 86 *vs* 82%, respectively)[16].

On the other hand, haploidentical transplantation has been performed more frequently during the last decade, with promising results[17-22]. Current practice mainly includes a preconditioning phase and then either selective depletion of T-cell receptor $\alpha\beta^+$/$CD19^+$ graft lymphocytes or the use of post-transplantation cyclophosphamide (post-Cy). The basic advantage of haploidentical donor HSCT is immediate donor accessibility, with no ethnic

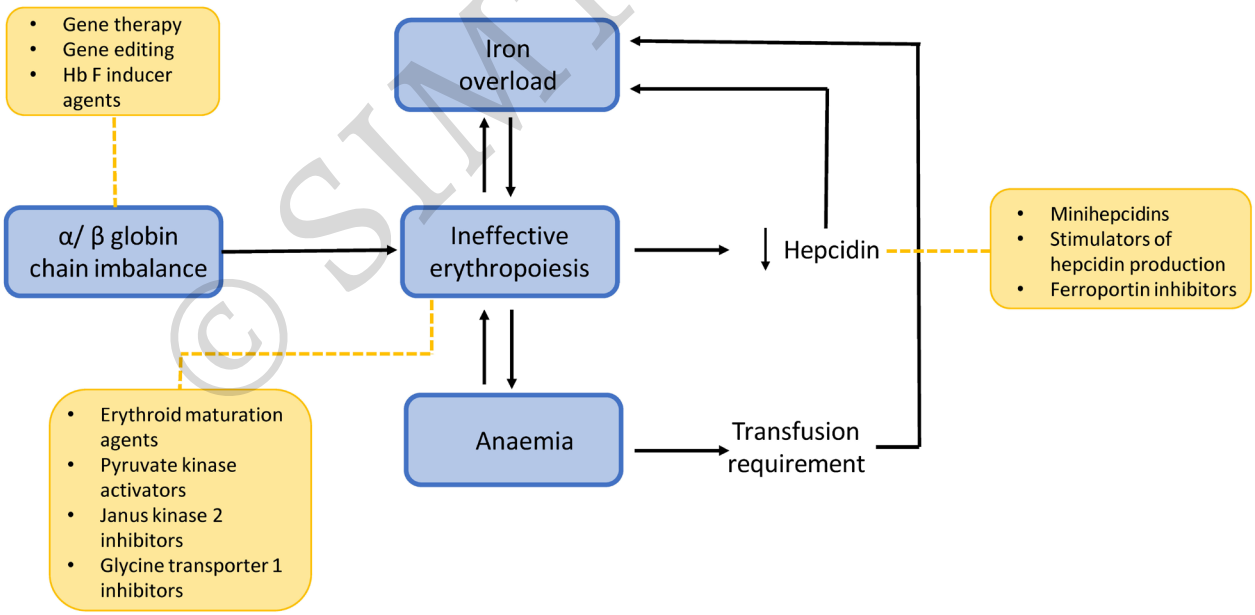

Figure 1 - Summary of novel targets and agents in β-thalassaemia

or racial restrictions. This offers a viable solution for thalassemia patients belonging to ethnic minorities[8].

However, several aspects limit allogeneic HSCT as a curative therapy. First, MSDs are available to only 20-30% of patients, depending on nationality and ethnic groups. Second, HSCT using alternative donors, mainly MUDs, haploidentical donors and cord blood transplants, have not yielded results that can equal those of allogeneic MSDs[23-25]. Third, the probability of real-life, long-term complications such as hypogonadism and thyroid dysfunction after HSCT needs to be considered[26-28]. Further data concerning secondary cancer are required[29]. Moreover several new conditioning regimens are now being evaluated in clinical trials as an effort to improve HSCT outcomes[8,9].

Gene therapy

Gene therapy is a novel and potentially curative treatment strategy for TDT patients that has been designed to correct the underlying α/β-globin chain ratio, thus improving the production of functional Hb, the erythropoiesis, and the chronic anaemia. After isolating haematopoietic stem and progenitor cells (HSPCs), exogenous β-globin genes are incorporated into the host-cell genome using a self-inactivating lentiviral vector. After full or partial myeloablative busulfan conditioning, these genetically modified autologous HSPCs are returned to the patient where they replicate and repopulate in the blood compartment and facilitate normal Hb synthesis[30,31]. In order for gene therapy to be successful in β-thalassaemia, certain requirements have to be met: high-efficiency haematopoietic stem cell (HSC) engraftment and gene transfer, high expression of the β/γ-globin gene and appropriate expression, with minimal to no risk of insertional mutagenesis. The use of gene therapy technology has been proven to be effective and curative in many animal models of β-thalassaemia[32-35] and in proof-of-principle studies in an adult patient with TDT[36,37].

The outcomes of two phase I/II studies (HGB-204 [NCT01745120] and HGB-205 [NCT02151526])[38] conducted on 22 TDT patients (12-35 years of age) who were reinfused with cells transduced *ex vivo* with the LentiGlobin BB305 vector led to the conditional approval in Europe of the gene therapy product betibeglogene autotemcel (Zynteglo, bluebird bio Inc., Cambridge, MA, USA) in June 2019. In these studies, and at a median of 26 months after receiving gene therapy, 12 of the 13 patients who had a non-β°/β° genotype had achieved transfusion independence, and biological markers indicated that ineffective erythropoiesis had been corrected. In 9 patients with β°β° genotype the median annualised transfusion volume was decreased by 73%, and 3 of these patients discontinued RBC transfusions. The adverse events (AEs) attributed to the LentiGlobin BB305 vector were reported and were found to be similar to those associated with autologous HSCT, and no clonal dominance related to lentiviral vector integration was observed[38].

The HGB-207 study (Northstar-2, NCT02906202) and the HGB-212 study (Northstar-3, NCT03207009) are currently ongoing phase III studies using the LentiGlobin BB305 vector for TDT subjects with non-β°β° and β°β° genotypes. Interim data from these studies projected after a median follow-up of 24.3-months showed that 30/34 (88.2%) patients became transfusion independent (6/7 [85.7%] β°/β° and 24/27 [88.9%] non-β°/β°) maintained for a median of 20.6 months)[39]. AEs were similar to those seen following myeloablative conditioning with busulfan. There were no deaths and no evidence of clonal dominance or insertional oncogenesis in these studies[39]. Following these initial positive results, the phase III studies were expanded to also include paediatric patients. Interim results in the paediatric population showed that 27 paediatric patients achieved a similar rate of transfusion independence and showed a similar safety profile to that of adults[40].

Most recently, preliminary data from 44 patients enrolled in the ongoing LTF-303 study (NCT02633943) with a median follow-up of 45.6 months (range: 22.9-76.4) showed that transfusion independence was achieved and sustained in 15/22 (68.2%) patients from the phase I/II studies and in 20/22 (90.9%) of patients treated in the phase III studies. Weighted average Hb during transfusion independence was 10.3 g/dL and 11.8 g/dL in patients in phase I/II and phase III trials, respectively. This was maintained for over three years of follow-up. Moreover, most patients who achieved transfusion independence had a decrease in liver iron concentration (LIC) approaching normal levels. While some serious AEs during LTF-303 study were observed, no deaths, replication-competent lentivirus, or insertional oncogenesis were reported[41].

A decision by bluebird bio to temporarily suspend all clinical trials and marketing of betibeglogene autotemcel was made on 16 February 2021, following two reports of acute myeloid leukaemia (AML) and myelodysplastic syndrome (MDS) in sickle cell patients recruited on the HGB-206 trial[42]. The MDS diagnosis was later revised to transfusion-dependent anaemia and the AML case was reported to be unlikely related to treatment[43]. While the conditional approval of betibeglogene autotemcel was given by the European Medicines Agency (EMA) in 2019, bluebird bio announced on August 9, 2021 that they will discontinue their operations in Europe due to payor challenges[44].

Other gene insertion approaches and vectors have also been evaluated in animal models and validated for clinical investigation. For example, 3 adult and 6 paediatric TDT patients with β^0 or severe $\beta+$ mutations were enrolled and treated in a phase I/II trial (TIGET- BTHAL, NCT02453477) with an intrabone administration of HSCs transduced with the lentiviral vector GLOBE, which removes the HS4 locus control region element. Transfusion reduction was seen in 3 adults and complete independence was achieved in 3/4 evaluated children[45]. Data from the phase I TNS9 clinical trial (NCT01639690), which was conducted on 4 adult TDT patients with β^0/β^+ and β^0/β^0 genotypes treated with autologous $CD34^+$ HSPCs transduced with the TNS9.3.55 lentiviral vector, showed durable and stable gene marking, no evidence of clonal dominance, and no toxicity profile higher than grade 3 in all patients. One patient experienced a significant decrease in transfusion requirements that lasted for more than five years[46,47]. Ongoing and future studies in gene therapy will focus on improving transduction efficiency by optimising protocols preserving biological HSC features and unravelling biological key factors for prediction of favourable outcomes.

Gene editing

Newer approaches have been under development to correct genetic mutations at the DNA level of the cell or to disrupt specific DNA sequences in the genome. *BCL11A*, which is located on chromosome 2, has been shown to be a potential and promising target for genome editing[30]. It acts as a key regulator of the foetal-to-adult Hb switch and HbF silencing[48,49]. Genetic variation in the expression of *BCL11A* and persistence of HbF production was shown to reduce clinical severity in β-thalassaemia[50,51]. Many *in vitro* studies have shown that knock-out gene-editing technology could permanently produce foetal Hb (HbF) in adults with thalassaemia[52-54]. Knock-down studies of *BCL11A* using RNA interference (RNAi) showed it to be an effective method to silence the γ-globin gene[49,55,56]. A new approach to induce HbF by silencing *BCL11A* involves the use of an erythroid-specific promoter to drive a microRNA-adapted short hairpin (shRNA)[53,57]. Other gene-editing approaches to inhibit *BCL11A* have been described and are currently in pre-clinical and clinical development; these include clustered, regularly interspaced, palindromic repeats (CRISPR) in association with Cas9 (CRISPR-Cas9), transcription activator-like effector nucleases (TALENS), and zinc finger nucleases (ZFNs)[58-61].

The first major product under study is CTX011 (CRISPR-Cas9), which is produced using *ex vivo* editing of the erythroid enhancer region of the *BCL11A* in $CD34^+$ HSPCs and reduces erythroid-specific expression of *BCL11A*. Infusion of CTX0011 was shown to increase HbF levels in erythroid cells *in vivo*. CLIMB THAL-111 (NCT03655678) is a phase I/II trial evaluating the safety and efficacy of CTX001 in 45 patients with TDT (age 12-35 years). Interim results from 10 TDT patients who have been followed up for 3.8-21.5 months, and who received CTX001 infusion, showed that engraftment of neutrophils and platelets was achieved at medians of 30 and 38.5 days, respectively; clinically meaningful levels of total Hb, HbF, and F-cell pancellularity were observed early and were maintained over time across all 10 patients (mean total Hb at baseline 10.6 g/dL including 0.2 g/dL HbF, mean total Hb at 21 months 13.3 g/dL including 12.5 g/dL HbF). Patients also stopped transfusions within two months of CTX001 infusion and were transfusion-free up to 23.8 months of follow-up. The safety profile of CTX0011 was also generally consistent with myeloablative conditioning and autologous HSCT[62,63].

The use of ZFN technology is also currently being studied in a clinical trial. THALES (NCT03432364) is a phase I/II clinical trial evaluating the safety, tolerability and efficacy of ST-400 (ZFN) in reducing transfusion requirement (frequency and volume) in 6 adults (18-40 years) with TDT. Preliminary results from 2 patients showed rapid haematopoietic reconstitution following myeloablative conditioning, and elevated HbF levels[64]. These data

are preliminary, and additional patients and longer follow-up will be required to better understand the safety and efficacy of this therapy.

Foetal-haemoglobin-inducing agents

Considerable efforts are also being made to stimulate γ-globin and HbF production by using pharmacological agents. Most notable of them all is IMR-687, a potent, specific, and highly selective small molecule inhibitor of phosphodiesterase (PDE) 9, that increases intracellular cGMP levels and reactivation of HbF[65]. A phase II, randomised, double-blind, placebo-controlled study is currently underway to evaluate the safety, tolerability, pharmacokinetics, and pharmacodynamics of IMR-687 given once daily for 36 weeks in a total of 120 β-thalassemia patients (TDT and NTDT) (NCT04411082), aged 18-65 years of age. The primary objective of this study is to assess the safety and tolerability of IMR-687 in adult subjects with TDT and NTDT. Secondary objectives in the TDT patients include reduction in RBC transfusion burden (TB), iron load rate, mean number of transfusion events, and mean change in ICT dose and serum ferritin levels. Secondary objectives in NTDT patients include change in Hb, mean change in HbF levels, and mean change in ICT dose and serum ferritin levels.

TARGETING INEFFECTIVE ERYTHROPOIESIS

Erythroid maturation agents

Sotatercept or ACE-011 has been shown to correct ineffective erythropoiesis by acting as a ligand trap to inhibit negative regulators of late-stage erythropoiesis in the TGF-β superfamily. Sotatercept was evaluated up to a phase II trial (NCT01571635) on 16 TDT patients and 30 NTDT patients[66]. Most patients with NTDT treated with higher doses of sotatercept achieved sustained increases in Hb level. On the other hand, TDT patients treated with higher doses of sotatercept achieved notable reductions in transfusion requirements. Although sotatercept was efficacious, exhibited an overall good safety profile, and was tolerated by most patients, a decision was made not to take sotatercept forward to phase III trials.

Luspatercept[67,68], an erythroid maturation agent, is a recombinant fusion protein that binds to the transforming growth factor beta (TGF-β) superfamily ligands to inhibit aberrant SMAD2/SMAD3 signalling and promote late-stage erythropoiesis[69,70]. Luspatercept was approved for the treatment of anaemia in adult patients with TD β-thalassemia by the US Food and Drug Administration (FDA) in November 2019 and by the EMA in June 2020[71,72]. The development of this drug was suggested when RAP-536 fusion proteins were used in mouse models with thalassaemia and showed beneficial effects on the reduction of ineffective erythropoiesis, splenomegaly, and iron overload[70,73]. Following on from this, a multicentre, open-label, randomised dose-finding phase II study (NCT01749540, NCT02268409) demonstrated that luspatercept at 0.2-1.25 mg/kg administered subcutaneously every three weeks for at least 5 cycles was effective and well-tolerated in 64 adults with β-thalassaemia[74]. This paved the way for the pivotal phase III BELIEVE trial (NCT02604433) which involved 336 adult patients with TDT or Hb E/β-thalassaemia with no transfusion-free period of >35 days within the 24 weeks before randomisation. Patients were randomized 2:1 to receive luspatercept (1.0 mg/kg, titration up to 1.25 mg/kg) or placebo every three weeks for ≥48 weeks[75]. A significantly greater percentage of patients receiving luspatercept achieved the primary endpoint of a ≥33% reduction in TB from baseline during weeks 13-24, with a reduction of ≥2 RBC units compared with placebo. The percentage of patients achieving a ≥50% reduction in TB *versus* baseline was greater in the luspatercept group than the placebo group for all time points examined. The least squares mean difference in the key secondary end point of mean change from baseline in TB from weeks 13-24 was also in favour of luspatercept[75]. All patient subgroups showed a benefit from luspatercept treatment, including those with the more severe β°/β° genotype[75]. In addition to reductions in TB, patients treated with luspatercept were more likely to achieve clinically meaningful improvements in health-related quality of life as measured by the Transfusion-Dependent Quality of Life (TranQoL) and the 36-item Short Form Health Survey (SF-36) questionnaires, compared with placebo[76]. Moreover, a higher proportion of luspatercept responders achieved clinically meaningful improvements in physical functioning and overall physical component summary compared with placebo. Initial data from 5-year open-label extension phase of the BELIEVE trial, which is currently ongoing, showed that luspatercept treatment was associated with a lower mean cumulative RBC transfusion units and visits through week 48 across

levels of baseline RBC TB. Luspatercept responders also showed meaningful reductions in RBC transfusion units and visits across levels of baseline RBC TB through week 120[77]. In addition, compared with placebo, luspatercept treatment was associated with a significant reduction in serum iron levels, LIC, and myocardial iron levels during the first 96 weeks of treatment[78]. Recruitment is expected to begin soon for a phase IIa study (NCT04143724) to evaluate the safety of luspatercept in paediatric patients who require regular RBC transfusions.

BEYOND (NCT03342404) is a phase II, double-blind, randomised (2:1), placebo-controlled, multicentre study evaluating the efficacy and safety of luspatercept in 145 adult patients with NTDT patients and a Hb level ≤10 g/dL[79]. The BEYOND study met its primary end point of mean increase in Hb of ≥1.0 g/dL from baseline over a continuous 12-week interval during weeks 13-24 in the absence of RBC transfusions with a statistically significant difference in favour of luspatercept. Treatment with luspatercept resulted in clinically significant and sustained improvements in anaemia in adults with NTDT, as measured by Hb levels, with >50% of patients receiving luspatercept achieving and maintaining a mean Hb increase of ≥1.5 g/dL[79]. Improvement in quality of life, as measured by the NTDT-patient-reported outcome Tiredness/Weakness (NTDT-PRO-T/W) domain score, favoured luspatercept and was correlated with increases in Hb levels. Luspatercept was well-tolerated over a prolonged period of time[79].

Pyruvate kinase activators

The enzyme pyruvate kinase (PK) plays a significant role in the last stage of glycolysis in the RBC, and its presence is, therefore, essential for energy production in the RBC. Based on preclinical studies conducted on PK-deficient mice, metabolic disturbance in PK deficiency alters not only the survival of RBCs, but also the maturation of erythroid progenitors, resulting in ineffective erythropoiesis[80]. The use of an oral PK activator known as AG-348 (mitapivat) in healthy subjects has been shown to increase ATP and has been proven to be efficient and safe in PK deficiency patients[81,82]. This same agent showed increased ATP levels, reduced markers of ineffective erythropoiesis, and improved anaemia, RBC survival, and indices of iron overload in mouse models of β-thalassaemia[83]. Interim results from a phase II open-label, multicentre study (NCT03692052) of mitapivat in 15 NTDT adult patients showed an increase in Hb of ≥1.0 g/dL in 11 of 15 patients at 12 weeks with favourable changes in markers of erythropoiesis and haemolysis. No serious AEs were reported[84]. Based on this, two phase III double-blind, randomised, placebo-controlled, multicentre studies evaluating the efficacy and safety of mitapivat (100 mg orally, twice daily) in 240 adult patients with TDT (ENERGIZE-T, NCT04770779) and 171 adult patients with NTDT (ENERGIZE, NCT04770753) are scheduled to begin soon[85].

Janus kinase 2 inhibitors

Preclinical studies have shown that Janus kinase 2 (JAK2) inhibition not only improved ineffective erythropoiesis but also reversed splenomegaly in an NTDT mouse model[86]. More data from other pre-clinical studies on TDT and NTDT mouse models also showed that JAK2 inhibitors are able to reduce splenomegaly[87]. These findings have suggested that the use of ruxolitinib, a JAK1/JAK2 inhibitor, could benefit β-thalassaemia patients. A single-arm, phase IIa study to evaluate the efficacy and safety of the JAK2 inhibitor ruxolitinib (INCB018424; INC424) administered orally at a starting dose of 10 mg twice daily among 30 adults with TDT and splenomegaly has been conducted (NCT02049450)[88]. A decrease in spleen size from baseline was observed in ruxolitinib-treated patients. There was also a mean change in spleen volume from baseline to week 12 (n=26) and week 30 (n=25)[88]. At week 30, one patient who initially had had a 15% decrease in spleen volume at week 12 demonstrated an increase in spleen volume[88]. However, there was no clinically significant improvement in pre-transfusion Hb, thus no related reduction in transfusion requirement. Moreover, although hepcidin levels increased in the ruxolitinib treatment group, no significant changes in iron parameters were observed over time[88]. For all the above-mentioned reasons, the study did not proceed to phase III[88].

Glycine transporter 1 inhibitor

The oral administration of bitopertin (RO-4917838), a small molecule selective inhibitor of glycine transporter 1 (GlyT1), resulted in reduced anaemia and haemolysis, enhanced *in vivo* survival of erythrocytes, and diminished ineffective erythropoiesis in β-thalassaemia mice[89]. Markers of cellular damage induced by reactive oxygen species were also substantially improved. A phase II study

of oral bitopertin in 12 adults with NTDT (NCT03271541) was conducted. The first 8 patients assessed at an 8-week preliminary efficacy analysis showed a reduction in mean total Hb. However, the study failed to show clinically significant improvements in haematologic and chemical biomarkers of disease activity and was,therefore, stopped early[90].

TARGETING IRON DYSREGULATION

Minihepcidins

Minihepcidins are known to restrict iron absorption. The use of minihepcidins on young $Hbb^{th3/+}$ mice led to amelioration of ineffective erythropoiesis, anaemia, splenomegaly, and iron overload[91]. In old $Hbb^{th3/+}$ mice, a combined administration of minihepcidin with the iron chelator deferiprone also improved ineffective erythropoiesis, and anaemia and reversed splenomegaly[91]. A new mouse model ($Hbb^{th1/th2}$), which closely resembles the human TDT phenotype, was generated to assess the efficacy of minihepcidins. In non-transfused $Hbb^{th1/th2}$ mice, administration of high-dose minihepcidin also improved RBC lifespan, and ameliorated ineffective erythropoiesis, anaemia, splenomegaly and iron overload[92]. With these encouraging data, the efficacy and safety of minihepcidins was assessed in several clinical trials. LJPC-401, a synthetic human hepcidin given as a subcutaneous injection, was evaluated in a phase II, multicentre, randomised, open-label study (NCT03381833) in 84 adult patients with TDT. This trial was stopped early for lack of efficacy as determined by interim end point analyses[93]. PTG-300, another injectable hepcidin mimetic, was also evaluated in the phase II TRANSCEND (NCT03802201) trial in 63 adult β-thalassaemia patients. Due to efficacy issues with the drug, however, this study has also recently been stopped.

Transmembrane protease serine 6

Other novel therapeutic approaches to target iron dysregulation include increasing the hepatic synthesis of hepcidin. This can be achieved by suppressing the gene transmembrane protease serine 6 (*TMPRSS6*). In mouse models with NTDT, the inhibition of *TMPRSS6* with small interfering RNA (siRNA) or antisense oligonucleotides caused a rise of hepcidin and an amelioration of anaemia and iron deposition[94,95]. Two major currently ongoing clinical trials are looking at stimulators of hepcidin production. The first is a phase I, randomised, single-blind, placebo-controlled, single-ascending and multiple-dose study (NCT04718844) to investigate the safety, tolerability, pharmacokinetic, and pharmacodynamic response of SLN124 [a GalNAc conjugated double-stranded fully modified siRNA that targets TMPRSS6 messenger RNA (mRNA)] in 112 adults with NTDT (α- and β-thalassaemia) and MDS. The second is phase IIa, randomised, open-label trial (NCT04059406) in 36 adults with NTDT who will be administered with TMPRSS6-LRx (a generation 2+ ligand-conjugated ASO targeting TMPRSS6). Interim data from both of these clinical trials are awaited.

Ferroportin inhibitors

Direct inhibition of the ferroportin receptor represents a novel approach to targeting the hepcidin-ferroportin axis[96]. VIT-2763 is an oral ferroportin inhibitor that has been specifically designed to target the hepcidin-ferroportin axis and ameliorate ineffective erythropoiesis. Preclinical data from NTDT mouse models showed that VIT-2763 decreased organ iron levels and improved haematologic parameters, including Hb level and RBC count, demonstrating amelioration of anaemia and improved erythropoiesis[97]. In addition, findings from a study of combination treatment with VIT-2763 and the iron-chelating agent deferasirox in the same preclinical model of NTDT revealed no negative impact of vamifeport on the efficacy of deferasirox or *vice versa*[98]. Based on this, a phase I, randomised, double-blind, placebo-controlled study was conducted on 72 healthy adult volunteers and showed that VIT-2763 administered at single oral doses up to 240 mg or multiple oral doses up to 120 mg twice daily was well-tolerated compared with placebo. There were no serious or severe AEs or discontinuations due to AEs[99]. A phase IIa trial (VIT-2763-THAL-201) to evaluate the efficacy of multiple doses of VIT-2763 in NTDT patients is currently ongoing. This study will assess improvements in haematologic parameters, including Hb and RBC indices, iron-related markers, and biomarkers for haemopoietic/erythropoietic activity. A phase IIb, multiple-dose, double-blind, randomised, placebo-controlled, parallel-group, multicentre trial in adult TDT patients is also being planned comprising a 24-week treatment phase with VIT-2763. This planned clinical trial will also assess and observe changes in Hb and RBC indices, iron metabolism parameters, and markers of erythropoietic activity[96].

CONCLUSIONS

A new era of novel therapies is emerging to advance care and improve outcomes for thalassaemia patients. It is now a reality that gene therapy can modify HSCs to cure diseases like β-thalassemia. However, major limitations to this approach remain. Manipulation and expansion of HSCs *ex vivo* is challenging and HSCs cannot be targeted *in vivo*. Moreover, current myeloablative regimens are toxic. Until gene therapeutic strategies are proven to be safe in terms of long-term complications, and based on the available scientific data, HSCT using alternative donors (first MUDs and then haploidentical donors), remains the only curative approach for β-thalassaemia patients with no sibling donor. Ultimately, in order for gene therapy to be useful it is important to establish the profile of the patient for whom it is a priority. This is particularly important as it becomes more widely used outside experimental trials conducted in highly selected and equipped centres. Moreover, current costs, limited availability and resources represent a further indication for a rational and progressive decision-making process regarding this innovative treatment. Identifying and selecting patients for gene therapy must be carried out through a consensus decision with a precise set of criteria[100]. Gene disruption via genome editing approaches can be very efficient. Specifically, the base editing approach can correct mutant genes through the achievement of a physiological and tissue specific expression of endogenous genes. However, chromothripsis, a potential phenomenon that is subsequent to the introduction of the double break in the DNA has been described[101] and should be kept in mind as a potential negative outcome. Such damage to the DNA is associated with a very high and dense region of mutations. Future genome editing approaches should aim to improve the targeting of quiescent cells and reducing potential genome toxicity.

The approval of luspatercept has led to the availability of another new treatment modality for TDT patients and may provide long-term sustained reductions in transfusion requirements and the need for ICT. Data from the long-term phase III BELIEVE trial will provide further valuable insights. Moreover, the clinical benefit of luspatercept treatment has now been observed in patients with NTDT from the phase II BEYOND trial, as measured by a meaningful improvement in anaemia. This could implicate the potential use of this agent in NTDT patients in the near future. In conclusion, we currently have several novel therapeutic agents in the field that are currently under development. Once the efficacy and safety of all these novel therapies has been established, long-term, head-to-head, and comparison trials are needed to guide the integration of these novel therapies into the standard care of β-thalassaemia. Moreover, cost-effectiveness and health-economic studies are warranted to select the best therapy for the individual patient in addition to demonstrating the cost/benefit of a particular therapy in prolonging a patient's life, while taking into consideration the resources available.

CONFLICTS OF INTEREST

RB reports no conflicts of interest. IM reports receiving honoraria from Sanofi-Genzyme and Amicus Therapeutics. MDC has been or is a current consultant for Novartis, Celgene Corp (Bristol Myers Squibb), Vifor Pharma and Ionis Pharmaceuticals, and has received research funding from Novartis, Celgene Corp (Bristol Myers Squibb), La Jolla Pharmaceutical Company, Roche, Protagonist Therapeutics, and CRISPR Therapeutics.

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