

BLA Clinical Review Memorandum

Application Type	Biologics license application (BLA)
STN	125717/0
CBER Received Date	20-Sep-2021
PDUFA Goal Date	19-August-2022
Division / Office	OTAT
Priority Review (Yes/No)	Yes
Reviewer Name(s)	Karl Kasamon, MD
Review Completion Date / Stamped Date	19-August-2022
Supervisory Concurrence	Tejashri Purohit-Sheth, MD
Applicant	bluebird bio, Inc.
Established Name	betibeglogene autotemcel
(Proposed) Trade Name	ZYNTEGLO
Pharmacologic Class	Autologous Hematopoietic Stem and progenitor Cells (HSPC)
Formulation(s), including Adjuvants, etc.	Autologous CD34+ Hematopoietic Stem Cells Transduced with Lentiviral Vector, BB305 LVV, Encoding the Human β^{A-T87Q} -Globin Gene.
Dosage Form(s) and Route(s) of Administration	Betibeglogene autotemcel is a cell suspension for infusion. Single cell dose of $\geq 5.0 \times 10^6$ CD34+ cells/kg to be provided via intravenous (IV) infusion
Dosing Regimen	Single treatment
Indication(s) and Intended Population(s)	Treatment of adult and pediatric patients with β thalassemia who require regular red blood cell (RBC) transfusions
Orphan Designated (Yes/No)	Yes

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GLOSSARY

AE	adverse event
allo-HSCT	allogeneic hematopoietic stem cell transplantation
AML	acute myeloid leukemia
AUC	area under the curve
beti-cel	Betibeglogene autotemcel
BLA	biologics license application
BT	Breakthrough Therapy
C	Conditioning
CALD	cerebral adrenal leukodystrophy
CFR	Code of Federal Regulations
c/dg	copies per diploid genome
CI	confidence interval
CMC	chemistry, manufacturing, and controls
CR	complete response
CSR	clinical study report
D1	Day 1
EAP	Efficacy Analysis Population (also transplant population, TP)
eCTD	electronic Common Technical Document
EFS	event-free survival
EMA	European Medicines Agency
EOP2	End-of-Phase 2
EPO	erythropoietin
ES	Executive Summary
EU	European Union
FDA	Food and Drug Administration
GCP	Good Clinical Practice
G-CSF	granulocyte-colony stimulating factor
GVHD	graft-versus-host disease
Hb	hemoglobin
HbA	hemoglobin A (i.e., adult hemoglobin)
HbA2	hemoglobin A2 (i.e., minor variant of adult hemoglobin)
Hb ^{AT87Q}	hemoglobin containing β^{A-T87Q} -globin
<i>HBB</i>	β -globin gene
HbE	hemoglobin E
HbF	fetal hemoglobin
HGVS	Human Genome Variation Society
HIV	human immunodeficiency virus
HLA	human leukocyte antigen
HPC-A	hematopoietic progenitor cells obtained by apheresis
HRQoL	health-related quality of life
HSC	hematopoietic stem cell
HSCT	hematopoietic stem cell transplant
ICF	informed consent form
ISA	Integration Site Analysis
ISE	integrated summary of efficacy

ITT	intent-to-treat
IV	intravenous
LVV	lentiviral vector
M	mobilization
M24	Month 24
<i>MECOM</i>	MDS1 and EVI1 locus (gene chromosomal location 3q26.2)
MedDRA	Medical Dictionary for Regulatory Activities
NE	neutrophil engraftment
OBE	Office of Biostatistics and Epidemiology
OCOD	Office of Communication Outreach and Development
OS	Overall Survival
OSE	Office of Surveillance and Epidemiology
PD	pharmacodynamics
PeRC	Pediatric Review Committee (CDER)
PI	package insert
PK	pharmacokinetics
PMR	postmarketing requirement
PREA	Pediatric Research Equity Act
qPCR	quantitative polymerase chain reaction
RBCs	red blood cells
REMS	risk evaluation and mitigation strategy
RTF	refuse to file
SAE	serious adverse event
SAP	Statistical Analysis Plan
SCD	sickle cell disease
SEP	Successful Engraftment Population
SIN	self-inactivating
SGE	Special Government Employee
SmPC	summary of product characteristics
SMQ	Standardized MedDRA Queries
SOC	System Organ Class
TDT	transfusion-dependent β -thalassemia
TI	transfusion independence
TIF	Thalassemia International Federation
TR	transfusion reduction
TRM	transplant-related mortality
US	United States
<i>VAMP4</i>	Vesicle Associated Membrane Protein 4 gene
VAS	visual analog score
VCN	vector copy number
VOD	veno-occlusive liver disease

1. Executive Summary

Bluebird bio submitted this Biologics License Application (BLA), STN 125717, for the licensure of betibeglogene autotemcel (referred to as beti-cel, with trade

name of ZYNTEGLO) for the treatment of adult and pediatric patients with β -thalassemia who require regular red blood cell (RBC) transfusions.

Beti-cel is comprised of autologous hematopoietic stem cells (HSCs) transduced with BB305 lentiviral vector (LVV) encoding β^{A-T87Q} -globin suspended in cryopreservative. Being a β -globin, β^{A-T87Q} -globin is expected to lead to reconstitution of hematopoiesis of functional red blood cells (RBCs) and mitigation of the sequelae of β -thalassemia. The recommended regimen is a single beti-cel dose of $\geq 5.0 \times 10^6$ CD34+ cells/kg administered intravenously (IV) after full myeloablative conditioning with busulfan.

β -thalassemia is a group of inherited hemoglobinopathies caused by mutations in the β -globin gene leading to reduced or absent expression of β -globin in erythropoietic cells. The resulting non-alpha globin chain imbalance in erythrocyte progenitors causes precipitation of unpaired alpha-globin chains, leading to destruction of erythroid precursors and ineffective erythropoiesis and peripheral hemolysis. β -thalassemia leads to increased iron absorption and progressive iron overload. Transfusion-dependent β -thalassemia (TDT) is the most severe form with life-long anemia requiring frequent red blood cell (RBC) transfusions, and is complicated by organopathy related to iron overload, reduced quality of life, and shortened survival. Allogenic hematopoietic stem cell transplantation (AHSCT) using HLA-matched, related donors results in the best outcomes, but few patients have such donors available. Despite supportive care with transfusions, iron chelators, and luspatercept (an erythroid maturation agent), there continues to be a significant unmet need for patients with this disease.

To support safety and effectiveness of beti-cel for TDT, the applicant conducted two Phase 1/2 as well as the ongoing Phase 3 trials HGB-207 and HGB-212. These are single-arm, open label, multicenter trials to evaluate beti-cel in patients with β thalassemia who required regular RBC transfusions. These two Phase 3 study designs are largely parallel. However, HGB-207 enrolled two age cohorts, Cohort 1 (≥ 12 years of age) and Cohort 2 (< 12 years of age), and the studies differed in TDT genotype eligibility. HGB-207 enrolled subjects with non- β^0/β^0 genotype, whereas HGB-212 enrolled those with β^0/β^0 and another genotype that has the same degree of clinical severity as the β^0/β^0 genotype. Upon completion of parent studies, subjects were to enroll into the non-interventional long-term, follow-up Study LTF-303, for up to 15 years from time of gene therapy administration.

Due to changes in product manufacturing, only the Phase 3 studies used a version of beti-cel similar to the commercial product, therefore only Phase 3 efficacy data were reviewed. Similarly, the safety review focused on the phase 3 trials but also considered supportive data from one of the Phase 1/2 trials (HGB-204).

The clinical studies with beti-cel enrolled 51 subjects, of whom 43 started mobilization, and of these, 41 received myeloablation and beti-cel. All subjects had β thalassemia with transfusion dependence requiring ≥ 100 mL/kg/year of RBCs over 2 years preceding enrollment (or ≥ 8 transfusions of pRBCs per thalassemia guidelines, per year in the two years preceding enrollment [subjects ≥ 12 years]). HGB-207 subjects' median age was 15 years (range 4-34), 52.2% female, 56.5% were Asian, 34.8% White, and 8.7% other. HGB-212 subjects' median age was 12.5 years (range 4 - 33), 44% were female, 39% Asian, 56% White, and 5.6% unreported.

The primary efficacy endpoint was specified as the proportion of subjects meeting the definition of transfusion independence (TI), which was defined as a weighted average Hb ≥ 9 g/dL without any pRBC transfusions for a continuous period of ≥ 12 months at any time during the study after drug product infusion. For HGB-207, the success criterion for the primary efficacy endpoint was the lower bound of the 2-sided 95% exact CI $\geq 30\%$ for Cohort 1 (subjects ≥ 12 years old). No success criterion was pre-specified for Cohort 2. In HGB-212, the success criterion was the lower bound of the 2-sided 95% exact CI $\geq 30\%$.

In Study HGB-207, 20 of 22 (91%) TI-evaluable subjects achieved TI at any time post beti-cel. Fourteen out of 15 TI-evaluable subjects (93%, 2-sided 95% CI of 68.1 to 99.8) achieved TI at any time post infusion in Cohort 1 and 6 out of 7 TI-evaluable subjects (86%, 2-sided 95% CI of 42.1 to 99.6) achieved TI at any time post infusion in Cohort 2. In Study HGB-212, 12 out of 14 TI-evaluable subjects (86%; 2-sided 95% CI of 57.2 to 98.2) achieved TI at any time post beti-cel. The success criterion for both studies has been met. The clinical efficacy endpoint was supported by secondary endpoints, including observed duration of TI in HGB-207 of 20.4 months (range 15.7-21.6) and a weighted average Hb during TI of median 11.7 (range 9.5 to 12.8 g/dL). Clinical efficacy endpoints were corroborated by pharmacodynamic data which showed sustained β^{A-T87Q} -globin expression of median 8.8 g/dL (range 0.3 to 12.4) at Month 24 (N=30) and remained durable through last follow up at Month 36.

Studies HGB-207 and HGB-212 served as the primary source of safety data from a total 43 subjects who started mobilization, of whom 41 received beti-cel. Supportive safety data were also reviewed from an additional 19 subjects who started mobilization and 18 who were dosed with beti-cel in Phase $\frac{1}{2}$ study HGB-204. In the Phase 3 trials, median age was 13 (range 4 - 34) years; 49% were females; 49% were Asian, 44% White, 5% Other, 2% Not Reported. The median duration of follow-up was 27.2 (range 4.1 - 48.2) months. Serious adverse events (SAEs) occurred in 37% of patients as of last follow-up. The most common SAEs ($> 3\%$) were pyrexia, thrombocytopenia, liver veno-occlusive disease, febrile neutropenia, neutropenia, and stomatitis. There were no deaths.

The most common non laboratory adverse events (AEs) ($\geq 20\%$) were mucositis, febrile neutropenia, vomiting, pyrexia, alopecia, epistaxis, abdominal pain,

musculoskeletal pain, cough, headache, diarrhea, rash, constipation, nausea, decreased appetite, pigmentation disorder, and pruritus. All beti-cel recipients had at least one laboratory-based AE of \geq grade 3 neutropenia, thrombocytopenia and leukopenia. Most of these AEs could be attributed to the myeloablative chemotherapy that is a prerequisite for beti-cel administration. Delayed platelet engraftment with prolonged thrombocytopenia was an observed risk of beti-cel treatment, although only one SAE of bleeding was attributed to beti-cel due to delayed platelet engraftment.

The reviewed safety data do not warrant a Risk Evaluation and Mitigation Strategies (REMS). However, in addition to product labeling, and routine pharmacovigilance, a clinical safety post-marketing requirement (PMR) study is being required to assess the long-term risk of hematologic malignancies related to insertional oncogenesis.

The BLA provides substantial evidence of safety and effectiveness for beti-cel for treatment of adult and pediatric patients with β -thalassemia who require regular red blood cell transfusions, based on two adequate and well controlled trials. The overall benefit-risk profile favors regular approval of beti-cel in adult and pediatric patients with beta thalassemia who require regular red cell transfusions. The clinical team recommends traditional approval of the BLA.

1.1 Demographic Information: Subgroup Demographics and Analysis Summary

Of the 51 subjects screened for the phase 3 studies HGB207 and HGB-212, 43 were enrolled and underwent mobilization and apheresis, 41 received conditioning chemotherapy and were treated with beti-cel. There were 20 (49%) females; 49% were Asian, 44% White, 7% other/not provided; 2% were Latino. Median age was 13 (range 4, 34), with 39% < age 12, 27% \geq 12 to < 18 and 34% \geq 18 years old. A total of 44% were treated in USA, 54% in European Union, and 2% in Thailand.

1.2 Patient Experience Data

Quality-of-life outcomes were assessed using the (b) (4) parent general core and general core); for adolescents: (b) (4) (parent general core and general core) and (b) (4) (Youth version; (b) (4); and for adults: (b) (4) and (b) (4)

Reviewer Comment

The Applicant did not seek a labeling claim based on QOL data and these data were not incorporated in the PI. The data were not evaluated as part of the application review, given the limitations of QOL assessments in uncontrolled, open-label trials. As with time-to-event endpoints, interpretation of patient-reported outcomes is challenging in uncontrolled clinical trials, because it is unclear to what extent the outcomes can be attributed to the treatment effect of the regimen vs. to

underlying disease and patient characteristics. Furthermore, one of the tools used was generally designed and intended to be used in patients with advanced cancers, and its validity in patients with hemoglobinopathy remains to be demonstrated.

Data Submitted in the Application

Check if Submitted	Type of Data	Section Where Discussed, if Applicable
<input checked="" type="checkbox"/>	Patient-reported outcome	6.1.11.2, 6.2.11.2, and 6.5.11
<input type="checkbox"/>	Observer-reported outcome	
<input type="checkbox"/>	Clinician-reported outcome	
<input type="checkbox"/>	Performance outcome	
<input type="checkbox"/>	Patient-focused drug development meeting summary	
<input type="checkbox"/>	FDA Patient Listening Session	
<input type="checkbox"/>	Qualitative studies (e.g., individual patient/caregiver interviews, focus group interviews, expert interviews, Delphi Panel)	
<input type="checkbox"/>	Observational survey studies	
<input type="checkbox"/>	Natural history studies	
<input type="checkbox"/>	Patient preference studies	
<input type="checkbox"/>	Other: (please specify)	
<input type="checkbox"/>	If no patient experience data were submitted by Applicant, indicate here.	
Check if Considered	Type of Data	Section Where Discussed, if Applicable
<input type="checkbox"/>	Perspectives shared at patient stakeholder meeting	
<input type="checkbox"/>	Patient-focused drug development meeting	
<input type="checkbox"/>	FDA Patient Listening Session	
<input type="checkbox"/>	Other stakeholder meeting summary report	
<input type="checkbox"/>	Observational survey studies	
<input type="checkbox"/>	Other: (please specify)	

2. Clinical and Regulatory Background

2.1 Disease or Health-Related Condition(s) Studied

Transfusion dependent Beta β -thalassemia (TDT) manifests with profound anemia that typically emerges within the first year of life. Without RBC transfusions, mortality by age 5 is as high as 80%. Most patients with TDT require RBC transfusion every 2- 5 weeks; however, transfusions lead to iron overload and organ damage, with resultant sequelae of endocrinopathies, cardiomyopathy, and

cirrhosis. Chronic anemia can lead to growth retardation, skeletal abnormalities, leg ulcers, and hepatosplenomegaly. Fluctuating Hb levels between transfusions lead to episodes of fatigue and malaise. RBC transfusions can be associated with viral infections such as hepatitis. Even with standard supportive care with transfusions and iron chelators, patients experience diminished quality of life with only a 55% probability of survival to age 30.

2.2 Currently Available, Pharmacologically Unrelated Treatment(s)/Intervention(s) for the Proposed Indication(s)

Chronic transfusion therapy and iron chelation are the mainstay treatment for patients with TDT and lessen the degree of anemia and suppress ineffective erythropoiesis, but rarely alleviate completely the fatigue and bone marrow pains of the patients. Regular RBC transfusions can lead to infections such as viral hepatitis or human immunodeficiency virus (HIV), alloimmunization, and reactions to mismatched blood components. Despite chelation therapy, transfusion related iron overload as a consequence of transfusions can result in endocrine, cardiac, and hepatic comorbidities and shorten lifespan. More recently, luspatercept-aamt was approved to treat anemia in adults and leads to reduced transfusion requirements. Allogeneic-HSCT is a potentially curative therapy for patients with β -thalassemia but is associated with risks such as graft rejection, GVHD, and severe infections. Moreover, most patients are not candidates for allo-HSCT due to lack of matched donors or disease complications. According to some sources, a significant (14%) transplant-related mortality (TRM) may accompany allo-HSCT¹.

2.4 Previous Human Experience with the Product (Including Foreign Experience)

Clinical study reports and datasets for most early studies have been submitted to the BLA and will be reviewed herein. On 29 May 2019, beti-cel (Zynteglo) was granted conditional approval in the E.U. and U.K. Zynteglo was indicated in Europe for patients ≥ 12 -year-old with non- β^0/β^0 -genotype TDT lacking Human Leukocyte Antigen (HLA)-matched related donor. Since EU approval, a single patient was treated with Zynteglo in Europe. Zynteglo is being withdrawn from the European market for business reasons.

2.5 Summary of Pre- and Post-submission Regulatory Activity Related to the Submission

December 19, 2012	Original Investigational new drug (IND) application submitted
January 17, 2013	IND application allowed to proceed
January 31, 2013	Fast Track Designation granted / Breakthrough Designation denied
March 18, 2013	Orphan Drug Designation granted (ODD #13-3905)

1 Caocci G, Orofino MG, Vacca A, Piroddi A, Piras E, Addari MC, Caria R, Pilia MP, Origa R, Moi P, La Nasa G. Long-term survival of beta thalassemia major patients treated with hematopoietic stem cell transplantation compared with survival with conventional treatment. Am J Hematol. 2017 Dec;92(12):1303-1310. doi: 10.1002/ajh.24898. Epub 2017 Sep 25. PMID: 28850704.

January 29, 2015	Breakthrough Therapy Designation granted
March 25, 2015	Type B, end of phase II (EOP2) Meeting
July 9, 2015	Type B, chemistry manufacturing controls (CMC) Meeting
May 31, 2016	Type B, Initial Comprehensive Multidisciplinary BTM meeting
July 11, 2018	Type B Meeting to obtain feedback on the potential registration pathway for beti-cel for treatment of patients with non- β^0/β^0 genotype TDT.
November 30, 2018	Rare Pediatric Disease Designation granted (RPD-2018-193).
March 5, 2019	Type B Meeting, Written Responses
September 19, 2019	Type B Meeting CMC November 7, 2019 Pre-biologics license application (BLA) Meeting
September 22, 2020	Type B Meeting
July 22, 2021	Type B Meeting (2 nd Pre-BLA)
September 20, 2021	Rolling Submission Part 2 Quality (CMC) and Clinical Modules
November 18, 2021	BLA filed
January 14, 2022	Major Amendment to BLA
January 18, 2022	Mid-Cycle Communication
February 14-18, 2022	Pre-license Inspection
June 10, 2022	Advisory Committee Hearing
August 19, 2022	Action Due Date

2.6 Other Relevant Background Information

Protocol amendment for Study HGB-207

Version 6 submitted 10 Jun 2021, updated clinical work-up criteria, procedure, and follow-up in the section describing integration site analyses, introduction of optional archival and genetic testing on bone marrow samples, added considerations for vaccines as concomitant medications, and removal of the statement that a subject may be withdrawn from the study if they have undetectable vector copy number (VCN) in peripheral blood cells for 2 consecutive measurements at least 1 month apart.

Protocol amendment for Study HGB 212

Version 6 submitted 10 Jun 2021, updated clinical work-up criteria, procedure, and follow-up in the section describing integration site analyses, introduction of optional archival and genetic testing on bone marrow samples, added considerations for vaccines as concomitant medications, and removal of the statement that a subject may be withdrawn from the study if they have undetectable VCN in peripheral blood cells for 2 consecutive measurements at least 1 month apart.

The proposed regulatory pathway for beti-cel is Section 351(a) of the Public Health Service Act (PHSA). bluebird bio has submitted a Biologics License Application (BLA) in accordance with 21 CFR 601, subpart C, seeking to demonstrate substantial evidence of effectiveness on a clinically meaningful endpoint. bluebird

bio has initiated submission of the BLA (BLA 125717) on a rolling basis with final Module 4 and associated summaries submitted in November 2019 and completed the rolling submission of the BLA with clinical section in September 2021. Bluebird bio has requested and been granted a priority review designation for the BLA due to the seriousness of the disease treated by beti-cel and the improvement in effectiveness it provides over other treatments.

3. SUBMISSION QUALITY AND GOOD CLINICAL PRACTICES

3.1 Submission Quality and Completeness

The submission was adequately organized and integrated to accommodate the conduct of a complete clinical review without unreasonable difficulty. Inadequacies were resolved via use of information requests (IRs).

3.2 Compliance With Good Clinical Practices And Submission Integrity

The applicant stated that the trials were completed in multiple centers overseas and in the USA under IND 15324, in accordance with the regulations specified in 21 CFR 312, and were compliant with Good Clinical Practice (GCP), international ethical and scientific quality standards for the design, conduct, recording, and reporting of clinical trials involving human subjects (including Title 21, United States (US) Code of Federal Regulations (CFR) Parts 50, 54, 56 and 312 Subpart D; the International Conference on Harmonisation (ICH) Guideline on Good Clinical Practice (GCP; E6); and the ethical principles outlined in the Declaration of Helsinki). The clinical trials included provisions for obtaining informed consent by all study subjects, and for ethical treatment of study subjects.

Four bioresearch monitoring (BIMO) inspections were completed, 3 clinical investigator inspections and one sponsor inspection. No significant inspectional findings impacting data integrity were noted in the completed EIRs, and the preliminary draft summary reports. The inspections performed by BIMO are listed below:

Table 1 Bioresearch Monitoring Inspection Summary

Site Number	Protocol Number	Location	FDA Form 483 Issued	Review Status	Inspection Classification
103	207, 212	The Children's Hospital of Philadelphia	No	*Inspection Complete Review Pending	No Action Indicated*
104	207, 212	UCSF Benioff Children's Hospital Oakland	No	*Inspection Complete Review Pending	No Action Indicated*
110	207, 212	Ann and Robert H. Lurie Children's Hospital of Chicago	No	Inspection Complete Review Complete	No Action Indicated
Sponsor	N/A	Bluebird bio, Inc. Cambridge, MA	No	Inspection Complete Review Complete	No Action Indicated

*Final review of the EIRs and final inspection classification are still pending.

Source: BIMO inspection review Memo

3.3 Financial Disclosures

No significant financial interests or conflicts were identified that could potentially bias the conduct of the studies. A complete list of clinical investigators was provided, and no investigators had disclosable financial interests/arrangements or submitted Form FDA 3455.

Table 2 Financial Disclosures

Covered clinical study (name and/or number): HGB-207
Was a list of clinical investigators provided? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request list from applicant)
Total number of investigators identified: <u>39</u>
Number of investigators who are sponsor employees (including both full-time and part-time employees): <u>0</u>
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): <u>0</u>

<p>If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)):</p> <p style="padding-left: 40px;">Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: _____</p> <p style="padding-left: 40px;">Significant payments of other sorts: _____</p> <p style="padding-left: 40px;">Proprietary interest in the product tested held by investigator: _____</p> <p style="padding-left: 40px;">Significant equity interest held by investigator in sponsor of covered study: _____</p> <p style="padding-left: 40px;">Is an attachment provided with details of the disclosable financial interests/arrangements? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request details from applicant)</p> <p style="padding-left: 40px;">Is a description of the steps taken to minimize potential bias provided? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request information from applicant)</p>
<p>Number of investigators with certification of due diligence (Form FDA 3454, box 3): <u>0</u></p> <p style="padding-left: 40px;">Is an attachment provided with the reason? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request explanation from applicant)</p>
<p>Covered clinical study (name and/or number): HGB-207</p>
<p>Was a list of clinical investigators provided? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No</p>
<p>Total number of investigators identified: 39</p>
<p>Number of investigators who are sponsor employees (including both full-time and part-time employees): 0</p>
<p>Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): 0</p>
<p>If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)):</p> <p style="padding-left: 40px;">Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study:</p> <p style="padding-left: 40px;">Significant payments of other sorts:</p> <p style="padding-left: 40px;">Proprietary interest in the product tested held by investigator:</p> <p style="padding-left: 40px;">Significant equity interest held by investigator in sponsor of covered study:</p> <p style="padding-left: 40px;">Is an attachment provided with details of the disclosable financial interests/arrangements? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request details from applicant)</p> <p style="padding-left: 40px;">Is a description of the steps taken to minimize potential bias provided?</p>

<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request information from applicant)
Number of investigators with certification of due diligence (Form FDA 3454, box 3): 0 Is an attachment provided with the reason? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request explanation from applicant)

Covered clinical study (name and/or number): HGB-212
Was a list of clinical investigators provided? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request list from applicant)
Total number of investigators identified: <u>80</u>
Number of investigators who are sponsor employees (including both full-time and part-time employees): <u>0</u>
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): <u>0</u>
If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)): Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: _____ Significant payments of other sorts: _____ Proprietary interest in the product tested held by investigator: _____ Significant equity interest held by investigator in sponsor of covered study: _____ Is an attachment provided with details of the disclosable financial interests/arrangements? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request details from applicant) Is a description of the steps taken to minimize potential bias provided? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request information from applicant)
Number of investigators with certification of due diligence (Form FDA 3454, box 3): <u>0</u> Is an attachment provided with the reason? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request explanation from applicant)

Covered clinical study (name and/or number): HGB-204
Was a list of clinical investigators provided? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request list from applicant)

Total number of investigators identified: <u>47</u>
Number of investigators who are sponsor employees (including both full-time and part-time employees): <u>0</u>
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): <u>0</u>
<p>If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)):</p> <p style="padding-left: 40px;">Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: _____</p> <p style="padding-left: 40px;">Significant payments of other sorts: _____</p> <p style="padding-left: 40px;">Proprietary interest in the product tested held by investigator: _____</p> <p style="padding-left: 40px;">Significant equity interest held by investigator in sponsor of covered study: _____</p> <p style="padding-left: 40px;">Is an attachment provided with details of the disclosable financial interests/arrangements? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request details from applicant)</p> <p style="padding-left: 40px;">Is a description of the steps taken to minimize potential bias provided? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request information from applicant)</p>
<p>Number of investigators with certification of due diligence (Form FDA 3454, box 3): _____</p> <p style="padding-left: 40px;">Is an attachment provided with the reason? <input type="checkbox"/> Yes <input type="checkbox"/> No (Request explanation from applicant)</p>

Covered clinical study (name and/or number): LTF-303
Was a list of clinical investigators provided? <input checked="" type="checkbox"/> Yes <input type="checkbox"/> No (Request list from applicant)
Total number of investigators identified: <u>71</u>
Number of investigators who are sponsor employees (including both full-time and part-time employees): <u>0</u>
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): <u>0</u>

If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)):

Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: _____

Significant payments of other sorts: _____

Proprietary interest in the product tested held by investigator: _____

Significant equity interest held by investigator in sponsor of covered study: _____

Is an attachment provided with details of the disclosable financial interests/arrangements? ☐ Yes ☐ No (Request details from applicant)

Is a description of the steps taken to minimize potential bias provided? ☐ Yes ☒ No (Request information from applicant)

Number of investigators with certification of due diligence (Form FDA 3454, box 3): 0

Is an attachment provided with the reason? ☐ Yes ☐ No

4. SIGNIFICANT EFFICACY/SAFETY ISSUES RELATED TO OTHER REVIEW DISCIPLINES

4.1 Chemistry, Manufacturing, and Controls

The BLA contained adequate description of manufacture process and characterization of beti-cel. Autologous hematopoietic cells are obtained by apheresis from the subject in order to manufacture the drug product. At the manufacturing facility, CD34+ cells are selected from the apheresed material and transduced with BB305 LVV and incubated with recombinant human cytokines and washed. The washed, transduced cells are the drug substance. Over the course of the beti-cel development program, important CMC issues included issues related to LVV potency and drug product potency. The Agency held meetings with the Applicant, with feedback for strategies to rectify the identified issues. The CMC review team is proposing post marketing commitments for the Applicant, which will include additional assessments of (b) (4) of beti-cel parameters such as (b) (4). Additional (b)(4) testing will be performed, as well as evaluation of certain (b) (4) leachable compounds, and (b) (4) testing of the bag (through (b) (4)).

Of note, Beti-cel manufacturing evolved between the version administered in Study HGB-205 (generation 0 product version), to HGB-204 (Generation 1 product version), and that used in HGB-207 and HGB-212 (Generation 2 product versions). Improvements in (b)(4) led to increases in the

percentage of vector-positive [%LVV+] cells, and integrated copies of the BB305 LVV sequence in beti-cel². Please see CMC memo for further details.

4.2 Assay Validation

See CMC memo

4.3 Nonclinical Pharmacology/Toxicology

The non-clinical data package included a series of in vitro experiments as well as in vivo animal studies, either using a test article manufactured according to a base manufacturing process or a refined manufacturing process. The pharmacokinetic and biodistribution properties of BB305 LVV-transduced β -thalassemic mouse BMCs were evaluated in vivo in a pivotal combined therapeutic POC, pharmacology, single-dose toxicity and genotoxicity study in the β -thalassemic mice. Considering the nature of the final beti-cel (autologous CD34+ hematopoietic stem cells transduced with BB305 lentiviral vector), the test articles used during non-clinical studies were surrogates and consisted of either thalassemic mouse bone marrow cells transduced with BB305 LVV, or human healthy donor HSCs transduced with BB305 LVV, administered to β -thalassemic (Hbbth1/th1) mice and immunodeficient (b) (4) mice, respectively. No test article-related mortality occurred, mean PLT counts were lower for animals treated with beti-cel compared with the control animals. This was attributed by the clinical pathologist to abnormal erythrocytes in control animals, specifically, when mice received beti-cel, their murine model of thalassemia was partially corrected. Control mice retained model thalassemia phenotype, with frequent red cell fragments and microcytes, which confounded platelet counter device.

Developmental and reproductive toxicology studies were not conducted because there were no adverse findings in the male and female reproductive tissues of mice administered murine beti-cel seen in the studies performed. No carcinogenicity/tumorigenicity studies with beti-cel were done and were not warranted based on the known safety profile. Overall, nonclinical studies did not identify any safety concerns that could not be addressed in the product label, and the nonclinical data support approval of the license application. Please see PT memo for details.

4.4 Clinical Pharmacology

Beti-cel contains autologous HSCs modified to express the transgenic β^{A-T87Q} -globin, the primary pharmacodynamic effect after engraftment and differentiation of the HSCs is the production of β^{A-T87Q} -globin. After infusion of beti-cel, LVV VCN in peripheral blood rapidly increases and plateaus at approximately Month 6, remaining stable until last follow up. This is associated with Hb^{AT87Q} also increasing steadily and then stabilizing by approximately Month 6. Hb^{AT87Q} levels are the chief

2 Locatelli, F., et al. Betibeglogene Autotemcel Gene Therapy for Non- β^0/β^0 Genotype β -Thalassemia. New England Journal of Medicine 386, 415-427 (2021).

contributor to unsupported hemoglobin levels in subjects who achieve transfusion independence after treatment (minor hemoglobin variants make up the rest). Median unsupported total Hb levels were > 10 g/dL during the observation period. Please see Clinical Pharmacology memo for details.

4.4.1 Mechanism of Action

The mechanism of action consists of engraftment in the bone marrow (BM) of the transduced CD34+ HSCs and differentiation, to produce RBC containing biologically active β^{A-T87Q} -globin that will combine with α -globin to produce functional Hb. This is predicted to correct the anemia and ineffective erythropoiesis that underpin TDT.

4.4.2 Human Pharmacodynamics (PD)

Beti-cel infusion is followed by rapid increase in LCC VCN in peripheral blood over the first few months, then the VCN reaches a plateau. The VCN levels generally remained stable as of the data cut off data of all studies, although variability of PB VCN kinetic profiles between subjects was high. Population PD analyses also indicated that beti-cel %LVV+ cells was the most important covariate impacting PB VCN levels. There was no correlation observed between total CD34+ cell dose and Hb^{A^{T87Q}} in peripheral blood at either Month 6 or Month 24, indicating that the lowest cell dose evaluated to date was adequate for effective reconstitution of HSCs in treated subjects.

Please see Clinical Pharmacology memo for details.

4.4.3 Human Pharmacokinetics (PK)

Please see pharmacology memo for details. As beti-cel is an ex vivo genetically modified autologous HSC-based product, traditional absorption, distribution, metabolism and excretion (ADME) studies are not relevant.

4.5 Statistical

The statistical reviewer verified that the primary study endpoint analyses cited by the applicant were supported by the submitted data. Please see the statistical review memorandum for details.

4.6 Pharmacovigilance

Bluebird bio has proposed to conduct a non-interventional post-authorization registry study (REG-501) evaluating the long-term safety and efficacy of beti-cel in patients with β -thalassemia for up to 15 years after treatment with beti-cel. However, upon review of the BLA and in discussion with Office of Biostatistics and Pharmacovigilance (OBPV), the review team determined that the proposed registry study would be insufficient to assess the postmarketing safety of beti-cel, particularly with respect to insertional oncogenesis. Consequently, the Applicant will be required to conduct a postmarketing study that includes specific testing and

evaluation for insertional oncogenesis as a Postmarketing Requirement. See OBPV memorandum for additional details.

5. SOURCES OF CLINICAL DATA AND OTHER INFORMATION CONSIDERED IN THE REVIEW

5.1 Review Strategy

The review focused on efficacy data derived from the ongoing Phase 3 studies HGB-207 (non- β^0/β^0 genotype) and HGB-212 (β^0/β^0 genotype), which used beti-cel manufactured with Generation 2 process. Supportive safety data were evaluated from the phase 1/2 study HGB-204 (which administered a lower dose of beti-cel manufactured with generation 1 process). Study HGB-205 evaluated 4 TDT subjects using a version of the beti-cel that could not be deemed comparable to the commercial product by CMC. As such, data from this study

were excluded from analysis. All subjects were encouraged to enroll into long term follow up study, Study LFT-303. Therefore, safety and efficacy data from LFT-303 were reviewed.

Insertional oncogenesis is a potential risk of LVV based therapy and reports of hematologic malignancy emerged in the development programs of other LVV products manufactured by the Applicant. These included myelodysplastic syndrome (MDS) with insertional oncogenesis in BLA 125755 and reports of acute myeloid leukemia (AML) and potential MDS in IND 15905. For these reasons, the FDA requested submission of safety data to this BLA from the following:

- IND 15905 is for sickle cell disease (SCD) using an LVV based drug product that shares the same LVV vector as beti-cel, though has some manufacturing differences compared to beti-cel.
- BLA 125755 is for cerebral adrenal leukodystrophy (CALD) with another LVV-based product manufactured using a related LVV vector.

To gain understanding and appreciation of potential insertional oncogenesis risks with beti-cel, high-level review of pertinent data from BLA 125755 and IND 015905 was performed during the review of this BLA.

5.2 BLA/IND Documents That Serve as the Basis for the Clinical Review

The review focused on Modules 1, 2 and 5. This included clinical study reports for Studies HGB-204, HGB-205, HGB-207, HGB-212, and LTF-303, along with case report forms, and data submitted in response to numerous information requests. Primary efficacy analyses were verified, and other analyses were performed by the review team using JMP 16 software.

The clinical review was primarily based on the Phase 3 trials HGB-207 and HGB-212, with efficacy data cut off of March 09, 2021, with 41 treated subjects. The protocols are described in Section 6.1 and 6.2, respectively. The team also reviewed relevant documents including pre-BLA review memos, meeting summaries, study protocols.

A major amendment which contained the safety data from BLA 125755 that is described in Section 5.1, and which contained a substantial amount of new information, added three months to the review clock.

5.3 Table of Studies/Clinical Trials

Clinical trials comprising the clinical development program which were submitted to the BLA include:

- Two completed Phase 1/2 studies (Studies HGB-205 and HGB-204)
- Two ongoing Phase 3 studies (Studies HGB-207 and HGB-212)
- One ongoing long-term follow-up study (Study LTF-303)
- Supportive safety data from HGB-206 in SCD

The table below contains the main clinical trials whose data was submitted to BLA:

Table 3 Clinical Studies Evaluating beti-cel in Subjects with TDT

<i>Study type</i>	<i>Study Identifier</i>	<i>Study Objective</i>	<i>Study Design</i>	<i>Dosage; Route of Administration</i>	<i>Number of Subjects</i>	<i>Study Status</i>
Phase 1/2	HGB-204	Safety and efficacy	Non- randomized open label, multi-site, single dose uncontrolled	$\geq 3.0 \times 10^6$ autologous transduced CD34+ hematopoietic stem cells/kg; Intravenous infusion	19	Completed
Phase 3	HGB-207	Efficacy and safety	Non- randomized, open label, multi-site, single Dose, uncontrolled	$\geq 5.0 \times 10^6$ autologous transduced CD34+ hematopoietic stem cells/kg; Intravenous infusion	24	Ongoing
Phase 3	HGB-212	Efficacy and safety	Non- randomized, open label, multi-site, single dose, uncontrolled	$\geq 5.0 \times 10^6$ autologous transduced CD34+ hematopoietic stem cells/kg; Intravenous infusion	19	Ongoing
Long-term follow-up	LTF-303	Long-term follow-up safety and efficacy	Multi-site, long-term follow-up	N/A: Subjects dosed in parent studies	51	Ongoing

Source: adapted from 125717/0002/m2/27-clin-sum, synopses-indiv-studies.pdf, page 1-2

*: HGB-205 was not reviewed due to inability to compare drug product administered. High level review of safety was performed

5.4 Consultations

5.4.1 Advisory Committee Meeting

Due to the complexities related to potential risk of insertional oncogenesis and hematologic malignancies with ZYNTGLO, and in light of hematologic malignancies reported after treatment with products manufactured with the same or related LVV for other diseases, an Advisory Committee (AC) Meeting was

convened on June 9-10, 2022. The review team posed the following voting question to the panel:

Do the benefits of beti-cel outweigh the risks for the treatment of subjects with transfusion-dependent β -thalassemia? The results of the vote were as follows: Yes=13; No=0; Abstain=0.

Additional, discussion questions included:

1. Hematologic malignancies have not occurred in transfusion-dependent β -thalassemia (TDT) subjects treated with beti-cel. However, the beti-cel lentiviral vector (LVV) is similar to the vector used in sickle cell disease (SCD) and is related to the vector used for cerebral adrenoleukodystrophy (CALD), and there have been cases of hematologic malignancies in both SCD and CALD subjects in other studies. In this setting, what is the likelihood that the constellation of delayed platelet reconstitution, abnormal marrow morphology findings, and insertion site analyses will predict future development of hematologic malignancies in TDT patients treated with beti-cel?

Summary of Discussion: Committee members highlighted the substantial differences between products, vectors, and disease states, as well as absence of any evidence of insertional oncogenesis to date with beti-cel. The committee reached agreement that while the etiology and significance of delayed hematopoietic reconstitution and cytopenias in beti-cel recipients is unclear, the differences between beti-cel versus eli-cel, as well as the underlying pathophysiology of beta-thalassemia vs. SCD and CALD, lessen their concern of hematologic malignancy risk in patients with beta-thalassemia.

2. Please discuss whether patients with TDT should be screened for potential germline and somatic mutations predisposing to hematologic malignancy prior to administration of beti-cel. What screening tests, if any, for such mutations would you recommend?

Summary of Discussion: The majority of the panel generally did not recommend screening for germline somatic mutations prior to product administration. A suggestion was to evaluate subclone evolution with next-generation sequencing (NGS), although a sensitivity of 0.2-0.5% would be needed, and such high sensitivity may not be feasible with available panels. Baseline bone marrow testing with standard cytogenetics and germline/somatic mutation evaluation was also suggested before treatment, to be subsequently reassessed in the event of cytopenias.

3. Please discuss the adequacy of the proposed postmarketing pharmacovigilance program, including the long-term follow-up study and registry study and discuss additional recommendations for safety monitoring for hematologic malignancies.

Summary of Discussion: The committee was unable to reach a consensus regarding multiple different safety monitoring recommendations. Assays discussed included detailed phenotyping to include more rare sub-clones, differentiating between original and expanded products, as well as baseline cells before and after transduction. Also mentioned was bone marrow analysis at baseline and possibly when primary endpoints related to neutrophil and platelet engraftments are not reached.

4. Please discuss recommendations for specific testing for hematologic malignancies following administration of beti-cel, to include frequency of testing, in patients with TDT.

Summary of Discussion: The panel mentioned tracking the importance of transduction efficiency, tracking the integration sites, clonal hematopoiesis in subclones and primitive stem cells, as well as next generation sequence (NGS) tests for driver mutations and consideration of FISH for its greater sensitivity. Some committee members also mentioned performing pre-implantation integration site analysis on a small sample of CD34+ drug product cells to later determine if the frequencies of specific gene integration events have increased relative to what was transplanted.

Reviewer Comment

The AC members opined that the hematologic malignancies observed in SCD subjects who received a similar LVV-based product are not informative regarding the risk of hematologic malignancy in patients with β -thalassemia who received ZYNTGLO. The AC members also opined that the pathophysiologic characteristics of SCD may predispose patients to increased risk of hematologic malignancy. Additionally, the AC members considered the Applicant's product for the treatment of CALD to be distinct from ZYNTGLO, and as such, the MDS cases observed in the CALD clinical program were not informative regarding the risk of insertional oncogenesis with ZYNTGLO. Ultimately, the expert panel unanimously concluded that ZYNTGLO had a favorable benefit-risk profile for the treatment of patients with β -thalassemia.

5.4.2 External Consults/Collaborations

Analysis of the potential role of LVV in the development of hematologic malignancy after treatment with LVV based products, and assessment of any relationship between beti-cel and other LVV based products, is very complex. Therefore, the Agency consulted Lucy A. Godley, MD, PhD, a multidisciplinary expert, to serve as special government employee (SGE). Dr. Godley has extensive expertise with stem cell transplantation, the treatment of patients with bone marrow diseases and malignancies and has a special interest in the molecular basis of hematologic malignancies. She is an active laboratory researcher studying DNA methylation in neoplastic cells, as well as hereditary bone marrow cancers. She has received

numerous awards for her research; she was determined to have no relevant conflict of interest.

Dr. Godley advised the review team advice on issues such as germline mutation screening for predisposition to malignancy, the utility of baseline bone marrow biopsies, the specifics of follow up testing, as well as integration site analysis (ISA). She was instrumental in our interpretation, understanding of the complexities of LVV integration and its impact on genes within the genome of affected study subjects.

5.5 Literature Reviewed (if applicable)

- Caocci G, Orofino MG, Vacca A, Piroddi A, Piras E, Addari MC, Caria R, Pilia MP, Origa R, Moi P, La Nasa G. Long-term survival of beta thalassemia major patients treated with hematopoietic stem cell transplantation compared with survival with conventional treatment. *Am J Hematol.* 2017 Dec;92(12):1303-1310. doi: 10.1002/ajh.24898. Epub 2017 Sep 25. PMID: 28850704.
- Akpek G, Pasquini MC, Logan B, Agovi MA, Lazarus HM, Marks DI, Bornhaeüser M, Ringdén O, Maziarz RT, Gupta V, Popat U, Maharaj D, Bolwell BJ, Rizzo JD, Ballen KK, Cooke KR, McCarthy PL, Ho VT. Effects of spleen status on early outcomes after hematopoietic cell transplantation. *Bone Marrow Transplant.* 2013 Jun;48(6):825-31. doi: 10.1038/bmt.2012.249. Epub 2012 Dec 10. PMID: 23222382; PMCID: PMC3606905.
- Pelizzo G, Guazzotti M, Klersy C, Nakib G, Costanzo F, Andreatta E, Bassotti G, Calcaterra V. (2018). PLOS ONE, 13 (8) 1-13. Spleen size evaluation in children: Time to define splenomegaly for pediatric surgeons and pediatricians <https://doi.org/10.1371/journal.pone.0202741>
- Brunson A, Keegan THM, Bang H, Mahajan A, Paulukonis S, Wun T. Increased risk of leukemia among sickle cell disease patients in California. *Blood.* 2017 Sep 28;130(13):1597-1599. doi: 10.1182/blood-2017-05-783233. Epub 2017 Aug 22. PMID: 28830890; PMCID: PMC5620417.

6. DISCUSSION OF INDIVIDUAL STUDIES/CLINICAL TRIALS

6.1 Trial #1

HGB-207 (NCT02906202)

This is an ongoing trial titled: A Phase 3 Single Arm Study Evaluating the Efficacy and Safety of Gene Therapy in Subjects with Transfusion-dependent β -Thalassemia, who do not have a β^0/β^0 Genotype, by Transplantation of Autologous CD34+ Stem Cells Transduced Ex Vivo with a Lentiviral β^A -T87Q-Globin Vector in Subjects ≤ 50 Years of Age. HGB-207 was initiated on 08 August 2016, and the data are reviewed at an interim data cut-off date of 09 March 2021.

6.1.1 Objectives (Primary, Secondary, etc.)

Primary: Evaluate the efficacy of treatment with beti-cel in subjects ≤ 50 years of age with transfusion dependent thalassemia (TDT) who do not have a $\beta 0/\beta 0$ genotype at the β -globin (HBB) gene.

Secondary: Evaluate the safety of treatment with beti-cel in subjects ≤ 50 years of age with TDT who do not have a $\beta 0/\beta 0$ genotype at the HBB gene.

6.1.2 Design Overview

6.1.3 Population

Subjects with TDT under ≤ 50 years of age at time of consent, with history of ≥ 100 mL/kg/year of packed red blood cells (pRBCs) in the 2 years preceding enrollment or were managed under standard thalassemia guidelines with ≥ 8 RBC transfusions per year (subjects ≥ 12 years). Subjects were without a human leukocyte antigen (HLA)-matched family donor, or matched unrelated donor if required by regional regulatory authority. Subjects with a mutation characterized as either $\beta 0$ or IVS-I-110 on both HBB alleles were to be excluded (i.e., genotypes of $\beta 0/\beta 0$, $\beta 0/\text{IVS-I-110}$, or $\text{IVS-I-110}/\text{IVS-I-110}$).

Reviewer Comment

Earlier Phase 1/2 studies administering a product version with less efficient transduction and lower dose suggested that TDT subjects not homozygous for $\beta 0$ alleles (i.e., those with non- $\beta 0/\beta 0$ genotypes) were the most likely to achieve TI after engraftment. This is due to the innate production of at least some endogenous hemoglobin, which would then be supplemented by additional expression of the Hb^{AT87Q} gene. The non $\beta 0/\beta 0$ group includes the following genotypes: $\beta 0/\beta +$, $\beta 0/\beta \text{E}$, $\beta +/\beta +$, and $\beta +/\beta \text{E}$. Conversely, it was felt that in subjects with $\beta 0/\beta 0$ disease, transduced Hb^{AT87Q} levels might reduce transfusion needs but might not achieve transfusion independence. For this reason, separate Phase 3 studies were launched for these TDT genotypes and HGB-207 included subjects with non- $\beta 0/\beta 0$ genotypes. HGB-207 also excluded the $\beta 0/\text{IVS-I-110}$ or $\text{IVS-I-110}/\text{IVS-I-110}$ genotypes, because they behave like $\beta 0/\beta 0$ genotype, expressing practically no β -globin.

6.1.4 Study Treatments or Agents Mandated by the Protocol

The protocol stipulated hypertransfusion to achieve a Hb level of ≥ 11 g/dL during mobilization and apheresis. HSC were mobilized using granulocyte colony stimulating factor (G-CSF) and plerixafor; a total of two mobilization cycles could be performed if needed. Each mobilization cycle may include up to three apheresis procedure days. No more than two consecutive apheresis procedure products may be sent for each transduction; each transduction produces an individual drug product lot. Mobilization cycles must be separated by at least two weeks.

Beti-cel consists of an autologous CD34+ cell-enriched population that contains HSC transduced with BB305 LVV encoding the β^{A-T87Q} -globin gene. Each beti-cel lot was prepared individually for each subject using their autologous cells, and >1 beti-cel lot may have been required to reach the minimum cell dose required. All subjects received beti-cel at a dose of $\geq 5.0 \times 10^6$ CD34+ cells/kg as a single IV dose on a single day, as stipulated per protocol.

The ITT population included 24 subjects who initiated any study procedure such as GCSF mobilization and were assessed for demographics and safety. The efficacy analysis population included those who underwent infusion of beti-cel after myeloablation (N=23). Parameters related to beti-cel administration are listed below:

Table 4 HGB-207 beti-cel Parameters in EAP

Parameter	Statistic	Cohort 1 (N=15)	Cohort 2 (N=8)	Total (N=23)
# of beti-cel lots administered	n (%)	12 (80)	7 (88)	19 (83)
1	n (%)	3 (20)	1 (12)	4 (17)
2				
Cell dose CD34+cells x10 ⁶ /kg	N Median (Range)	15 7.4 5.0, 19.4	8 9.5 7.2, 19.9	23 8.1 5.0, 19.9
VCN in beti-cel (weighted average per subject c/dg) ^a	N Median (Range)	15 3.4 2.2, 5.6	8 2.7 1.9, 4	23 3.26 1.9, 5.6
%LVV+ Cells in beti-cel (weighted average per subject) ^a	N Median (Range)	15 82.0, 53.0, 90	8 68.5 34.0, 81.0	23 79.3 34.0, 90
beti-cel VCN/ beti-cel % LVV + Cells (weighted average per subject) ^a	N Median (Range)	15 4.4 3.4, 6.4	8 4.2 2.8, 7.6	23 4.4 2.8, 7.65

Source: Reviewer calculations ADSL dataset.

Abbrev.: beti-cel, beti-cel; LVV, lentiviral vector; VCN, vector copy number

^a If a subject had multiple lots of beti-cel, the VCN, %LVV+ cell in beti-cel, and beti-cel VCN/beti-cel %LVV+ cells were calculated per lot then the fractions of dose in each lot/total dose of all lots were used as weight to get weighted average per subject prior to statistical considerations.

To ensure successful engraftment, the total cell dose in the beti-cel was required to meet a minimal cell dose of 5.0×10^6 CD34+ cells/kg. Of 23 subjects, 5 (22%) required 2 cycles of mobilization/ apheresis.

The median cell dose administered in Cohort 1 was slightly greater than in Cohort 2 but ranges largely overlapped. On the other hand, median beti-cel VCN and the median beti-cel %LVV+ Cells in Cohort 1 are both higher than seen in Cohort 2, although ranges overlap, indicating that the majority of the subjects in Cohort 1

received beti-cel that contained a greater proportion of transduced cells than the beti-cel produced for subjects in Cohort 2. However, the median beti-cel VCN/beti-cel %LVV+ Cell ratios are similar between the cohorts, suggesting that the number of integrations per transduced cell is similar between the beti-cel produced for Cohort 1 and Cohort 2.

Reviewer Comment

The differences in the dose of beti-cel between the cohorts appear small. The impact is unclear, although beti-cel dose was selected in part based on minimal CD34+ cells needed for timely neutrophil engraftment (NE) from auto HSCT literature. Since all subjects exceeded the minimal dose, and experienced NE without need for back up cells, the differences in doses are not considered significant.

6.1.5 Directions for Use

Beti-cel was to be given after a minimum of 48 hours after completion of the busulfan conditioning regimen in order to achieve complete washout of busulfan. Beti-cel was to be administered on Day 1 via IV infusion. Vital sign monitoring including electrocardiogram (ECG), pulse oximetry, and blood pressure measurements, were to be employed during drug-product infusion. Additionally, vital signs (excluding ECG) were to be measured for two hours after infusion. Infusion reactions, including anaphylaxis, were to be managed according to the medical judgment of the investigator.

6.1.6 Sites and Centers

The study is being conducted at 9 study centers in France, Germany, Italy, United Kingdom, Thailand, and USA.

Reviewer Comment

The study sites appropriately were located in regions of the world with higher prevalence of TDT, such as the Mediterranean (Italy) and South-East Asia (Thailand), and places where people from these regions have immigrated.

6.1.7 Surveillance/Monitoring

The Applicant ensured appropriate monitoring procedures were performed before, on, and after the study. Furthermore, Study HGB-207 utilized an independent Data Monitoring Committee (DMC) comprised of members with appropriate scientific and medical expertise. Study candidates underwent a battery of screening procedures within 90 days of mobilization. This included detailed review of records from the prior two years on transfusions and hospitalizations. In the pre-conditioning phase, 30 days before busulfan, subjects were hypertransfused to a hemoglobin level of $\geq 11\text{g/dL}$. Conditioning with busulfan took place between Day -6 to -3, after which subjects had beti-cel infusion on Day 1 and remained in the hospital post infusion until neutrophil engraftment.

The applicant ensured appropriate monitoring procedures were performed before during and after the study. Please see the appendix for Study HGB-207 schedule of events.

After the Month 24 Visit, consenting subjects were enrolled in long-term follow-up Study LTF-303, to be followed for up to an additional 13 years. It included an independent Data Monitoring Committee (DMC).

6.1.8 Endpoints and Criteria for Study Success

Primary Endpoint:

- The proportion of subjects who meet the definition of transfusion independence (TI). TI is defined as a weighted average hemoglobin (Hb) \geq 9 g/dL without any pRBC transfusions for a continuous period of \geq 12 months at any time during the study after beti-cel infusion.

Secondary Endpoints:

- Characterization of subjects achieving TI (proportion of subjects who meet the definition of TI at Month 24; duration of TI; time from beti-cel infusion to achievement of TI; and weighted average Hb during TI)
- Characterization of transfusion reduction from 12 months post-beti-cel infusion through Month 24 compared to annualized requirement during 2 years prior to enrollment
- Weighted average nadir Hb during the 2 years prior to enrollment compared to weighted average nadir Hb from 12 months post-beti-cel infusion through Month 24
- Unsupported total Hb levels over time
- Characterization of use of iron chelation and/or therapeutic phlebotomy among all subjects
- Evaluation of the change in iron burden over time, including liver iron content by magnetic resonance imaging (MRI), cardiac T2* on MRI, and change in serum ferritin at baseline to Month 12 and Month 24
- Evaluation of health-related quality of life (HRQoL) based on validated tools:
 - for pediatrics: (b) (4) for adolescents: (b) (4) (parent general core and general core) and (b) (4) (Youth version; (b) (4) ; and
 - for adults: (b) (4)

Exploratory Endpoints:

- Assessment of growth and puberty parameters, bone density, diabetes, endocrine evaluations, and neurocognitive development (pediatric subjects <18 years of age)
- Assessment of change in dyserythropoiesis

- Correlations of pre-treatment variables (e.g., beti-cel vector copy number [VCN]) with response (e.g., peripheral blood VCN, hemoglobin A [HbA]T87Q)
- Measures of health resource utilization (including comparing annualized number of transfusions, number of hospitalizations, and number of days hospitalized, from 12 months post-beti-cel infusion through Month 24 with the annualized corresponding parameters during the 2 years prior to enrollment)
- Length of in-patient hospital stay from initiation of conditioning to discharge

Safety Endpoints:

- Success and kinetics of HSC engraftment
- Incidence of transplant-related mortality through 100 days and through 365 days post-beti-cel infusion
- Overall survival (OS)
- Detection of vector-derived replication competent lentivirus (RCL) in any subject
- Monitoring of laboratory parameters
- Frequency and severity of clinical adverse events (AEs)
- Incidence of acute and/or chronic graft-versus-host disease (GVHD)
- The number of subjects with insertional oncogenesis (myelodysplasia, leukemia, lymphoma, etc.)

Exploratory Endpoint:

The number of subjects with clonal predominance

Pharmacodynamics:

- β^{A-T87Q} -globin expression over time, including Month 6, Month 9, Month 18 and Month 24, as measured by assessing the ratio of β^{A-T87Q} -globin to all β -like globin chains in whole blood, and the ratio of α -globin to all β -like globin chains, in whole blood; and correlation of β^{A-T87Q} -globin expression at early time points post beti-cel infusion to β^{A-T87Q} -globin expression at later time points, as well as clinical outcomes.
- VCN in peripheral blood over time, including Month 6, Month 9, Month 12, Month 18, and Month 24
- Exploratory pharmacodynamic (PD) endpoints including the relationship between measures of myeloablation and PD and clinical outcomes.

6.1.9 Statistical Considerations & Statistical Analysis Plan

Analyses followed the statistical analysis plan (SAP Version 4.0, dated 18 December 2020). Per the SAP, an interim analysis was planned to support the regulatory submission of beti-cel for the treatment of TDT.

The following subject populations were to be evaluated and used for presentation and analysis of the data:

- Intent-to-Treat (ITT) population: All subjects who initiate any study procedures, beginning with mobilization by G-CSF and/or plerixafor
- Efficacy Analysis Population (EAP): All subjects who receive beti-cel
- Successful Engraftment Population (SEP): All subjects with successful neutrophil engraftment (NE) after beti-cel infusion.
- Study prospectively defined cohort 1 for participants ≥ 12 years of age, and cohort 2 for those < 12 .

The ITT population is the primary population for the safety analysis. The EAP is the primary population for efficacy, PD, and transplant parameter endpoints. The SEP was to be used to provide supportive data for subjects who engraft; however, the EAP and SEP were the same for this interim CSR, thus additional analysis of the SEP was not performed.

Tabulations were produced for appropriate demographic, baseline, efficacy, and safety parameters. For categorical variables, summary tabulations of the number and percentage of subjects within each category of the parameter are presented. For continuous variables, the number of subjects, mean, standard deviation (SD), median, minimum, and maximum values are presented. Descriptive summary statistics as well as 2-sided 95% confidence intervals (CIs), as appropriate, are presented on selected parameters, and the exact CIs for proportions were calculated using the Clopper-Pearson exact method. Longitudinal data (collected serially over time on study and follow-up) are presented by appropriate time intervals, such as monthly, quarterly and so forth, depending on the nature of the data. Two years of retrospective pre-study enrollment data were collected for each subject in the study, so that each subject may serve as his/her own control for the parameters of pRBC transfusion requirements, weighted average nadir hemoglobin (Hb) concentrations, and in-patient hospitalizations. For these parameters, baseline was annualized over the 2 years prior to study entry (date of informed consent). For other efficacy parameters as well as for PD parameters, baseline was defined as the most recent measurement prior to conditioning; the conditioning start date was defined as the first date of busulfan administration. For safety parameters, including shifts in key laboratory parameters, the most recent value prior to mobilization was used as baseline assessment.

The primary efficacy endpoint of TI was analyzed as a point-estimate of the proportion of subjects achieving TI at any time during the study, with a 2-sided 95% CI calculated using the Clopper-Pearson exact binomial method. The success criterion for Cohort 1 was proposed as a point estimate of 60% (9 out of 15 subjects), which would yield a lower 1-sided 97.5 exact confidence bound of 32.3%, exceeding the 30% minimal criterion. And the success criterion for Cohort 2 was proposed as a point estimate of 62.5% (5 out of 8 subjects).

Secondary efficacy endpoints were descriptively analyzed in summary tables where data are available; all efficacy data were presented in listings.

All subjects starting mobilization (i.e., the ITT population) were evaluated for safety. All AEs were listed and summarized for the following time periods: 1) from informed consent/assent up to the start of mobilization; 2) from start of mobilization up to start of conditioning; 3) from start of conditioning through neutrophil engraftment (NE); 4) from NE through Month 24 Visit; 5) from beti-cel infusion through Month 24 Visit; 6) from informed consent/assent through Month 24 Visit. Additionally, survival status, laboratory results, and insertional oncogenesis (insertional mutagenesis resulting in oncogenesis) events were summarized.

Reviewer Comment

Please refer to Statistical review for detailed information regarding the statistical analysis plan.

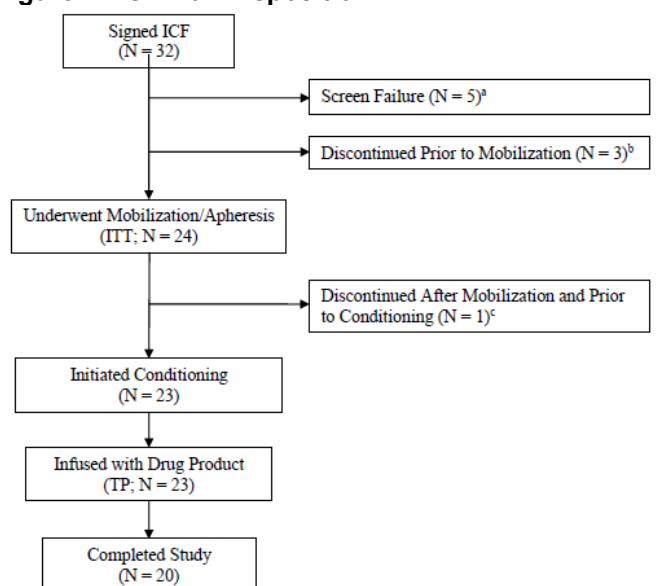
6.1.10 Study Population and Disposition

At time of interim analysis, 32 subjects signed informed consent/assent, of whom 5 subjects were screen failures. Three subjects were excluded due to advanced liver disease, one was excluded due to severe cardiac iron overload, with cardiac T2* < 10 msec measured by MRI, and one due to having $\beta 0$ on both HBB alleles. Three additional subjects withdrew consent prior to mobilization and therefore were not part of the ITT population.

Study disposition illustrated in Figure 1 below shows that 24 subjects were mobilized with G-CSF and plerixafor (ITT population). Of these 24 subjects, one subject (Subject (b) (6)) underwent 1 cycle of mobilization/apheresis with G-CSF and plerixafor but discontinued before conditioning because of pregnancy. The remaining 23 subjects were conditioned with busulfan and administered beti-cel (EAP).

All 23 subjects who were infused with beti-cel achieved successful neutrophil engraftment (NE) (SEP), 23 subjects had completed at least their Month 12 Visit, and 20 subjects had completed the study (Month 24 Visit). Data are presented for the overall ITT population as well as by protocol-defined Cohorts.

Figure 1 HGB-207 Disposition



Abbrev.: ICF, informed consent form; ITT, Intent-to-Treat; MRI, magnetic resonance imaging; TP, Transplant Population

^a Reason for screen failure: 3 due to advanced liver disease, 1 due to cardiac T2* < 10 msec measured by MRI, and 1 due to an ineligible genotype.

^b Reason for discontinuation prior to mobilization: 3 withdrew consent.

^c Reason for discontinuation after mobilization and prior to conditioning: 1 due to pregnancy.

Source: Copied from BLA Submission, HGB-207 Study Report (SR) page 95

6.1.10.1 Populations Enrolled/Analyzed

HGB-207 populations included intention to treat (ITT), which contained all subject signing ICF who underwent any part of the study, e.g. mobilization, Efficacy Analysis Population (EAP) who underwent conditioning, and infusion of beti-cel, and successful engraftment population (SEP) which required that subjects have neutrophil engraftment after beti-cel infusion, defined as the first of 3 consecutive absolute neutrophil count laboratory values $\geq 0.5 \times 10^9$ /L obtained on different days after a post-transplant value of $< 0.5 \times 10^9$ /L within 42 days after infusion.

6.1.10.1.1 Demographics

The ITT population included 24 subjects, 10 of whom were ≥ 18 years of age (41.7%), 6 were ≥ 12 to < 18 years of age (25.0%), and 8 were < 12 years of age (33.3%) at the time of consent/assent. The overall age distribution is the result of the protocol-defined age requirements and staggered enrollment.

Most subjects were Asian (14/24, 58.3%); 8/24 (33.3%) were White (8/24, 33.3%); and 2 (8.3%) subjects identified their race as Other (1 Asian Pakistani and 1 Caucasian/Thai). The ITT was balanced by sex (13/24, 54.2% females).

Table 5 HGB-207 Demographics

Parameter	Statistic	Cohort 1 (≥ 12 YO) (N=16)	Cohort 2 (<12 YO) (N=8)	Overall (N=14)
Age at ICF (years)	N	16	8	24
	Median	20	8	15

	(Range)	(12, 34)	(4,11)	(4,34)
Age at ICF				
≥18 years	n (%)	10 (62.5)	0	10 (41.7)
≥12 years to < 18 years	n (%)	6 (37.5)	0	6 (25)
< 12 years	n (%)	0	8 (100)	8 (33.3)
Sex				
Male	n (%)	7 (43.8)	4 (50)	11 (45.8)
Female	n (%)	9 (56.3)	4 (50)	13 (54.2)
Race				
Asian	n (%)	10 (62.5)	4 (50)	14 (58.3)
White	n (%)	6 (37.5)	2 (25)	8 (33.3)
Other	n (%)	0	2 (25)	2 (8.3)
Ethnicity				
Hispanic/Latino	n (%)	0	1 (12.5)	1 (4.2)
Not Hispanic/Latino	n (%)	10 (100)	6 (75)	22 (91.7)
Not provided	n (%)	0	1 (12.5)	1 (4.2)

Source: Reviewer calculations ADSL dataset

Reviewer Comment

Number of subjects was small, limiting conclusions regarding subgroup analysis based on demographics. There were no African American subjects in this study, and Asian subjects comprised the majority of subjects. This reflects the general epidemiology of beta thalassemia, which is particularly prevalent in the Mediterranean, Middle East, Africa, central Asia, the Indian subcontinent, and the Far East. Cohort 2 was comprised of pediatric subjects < 12 years of age and enrolled 8 subjects.

6.1.10.1.2 Medical/Behavioral Characterization of the Enrolled Population

His section will discuss several baseline thalassemia related subject characteristics, and these will be discussed in this section. The age at which thalassemia was diagnosed and age upon commencement of transfusions was also documented for all subjects. Subjects were diagnosed at a median age of 12 months, range of 0 to 84 months, and median age of first RBC transfusion was 6 months, with range of 3 to 84 months. The protocol included strict inclusion criteria that established the phenotype and severity of the disease with respect to number and volume of RBC transfusions the subject required within the 24 months preceding signature of ICF. Liver function might be negatively impacted by the ubiquitous transfusions that TDT subjects depend on lifelong, and this might in turn predispose to development of complications of the myeloablative conditioning regimen (with busulfan) that is a prerequisite for beti-cel. Liver function testing as well as imaging ascertainment of liver iron content (LIC) was therefore performed on all subjects with MRI. The median LIC was 5.45 (range of 1-41), the three subjects who had the highest LIC (over 16 mg/g) also underwent liver biopsy. Similarly, cardiac iron overload is common and thus candidates were screened

with ventricular ejection fraction evaluation by echocardiogram at baseline. Median EF was 66%, with range of 54 to 77%. Twenty-three of 24 (96%) of subjects were taking chelator therapy upon signature of ICF. Finally, splenomegaly is a prevalent sequela of TDT which may impact to platelet³ (and even neutrophil) engraftment, therefore, spleen volume was assessed at baseline in all subjects, as was a history of splenectomy. The study includes adult and pediatric subjects as young as 4 years of age, and spleen size is directly proportional to age, between 0 and 18 years of age⁴. However, the study does not attempt to correct or normalize splenic size to age nor body size. Median splenic size was 342 cm³ (range 68 to 1165), The pediatric cohort having smaller spleens than the older group; 5/24 (79%) of the subjects were status post splenectomy. Baseline characteristics are listed below:

Table 6 HGB-207 Subject Baseline Medical Characteristics

Parameter	Statistic	Cohort 1 (N=16)	Cohort 2 (N=8)	Total (N=24)
Diagnosis age (months)	N Median (Range)	16 12.0 (3, 84)	8 13.0 (0, 36)	24 12.0 (0, 84)
Age at 1 st RBC transfusion (months)	N Median (Range)	16 11 (4, 84)	8 16.0 (3, 36)	24 12.0 (3, 84)
Transfusion dependent on established regimen (months)	N Median (Range)	16 90 (6, 216)	8 17.5 (3, 48)	24 42 (3, 216)
Splenectomy	n (%)	5 (31.3)	0	5 (20.8)
No Splenectomy	n (%)	11 (68.6)	7 (100)	19 (79.2)
Spleen size	N Median (Range)	11 416 (230, 1165)	7 161 (68, 381)	18 342 (68, 1165)
Chelation onset (years)	N Median (Range)	16 4.50 (0.5, 17.0)	8 2.50 (1.3, 4.0)	24 3.00 (0.5, 17.0)
Liver Iron Content (mg/g) per MRI	N Median (Range)	15 7.2 (1, 41)	8 3.8 (1.8, 9.0)	23 5.3 (1, 41)

Source: Calculated by reviewer from ADSL dataset

3 Akpek G, Pasquini MC, Logan B, Agovi MA, Lazarus HM, Marks DI, Bornhaeüser M, Ringdén O, Maziarz RT, Gupta V, Popat U, Maharaj D, Bolwell BJ, Rizzo JD, Ballen KK, Cooke KR, McCarthy PL, Ho VT. Effects of spleen status on early outcomes after hematopoietic cell transplantation. Bone Marrow Transplant. 2013 Jun;48(6):825-31. doi: 10.1038/bmt.2012.249. Epub 2012 Dec 10. PMID: 23222382; PMCID: PMC3606905.

4 Pelizzo G, Guazzotti M, Klersy C, Nakib G, Costanzo F, Andreatta E, Bassotti G, Calcaterra V. (2018). PLOS ONE, 13 (8) 1-13. Spleen size evaluation in children: Time to define splenomegaly for pediatric surgeons and pediatricians <https://doi.org/10.1371/journal.pone.0202741>

Reviewer Comment

The older age Cohort 1 subjects started their first RBC transfusion slightly earlier than the pediatric Cohort 2, but Cohort 1 did not become completely dependent on an established transfusion regimen until later (90 months) vs. Cohort 2, who were dependent on an established RBC regimen by age 17.5 months.

Table 7 HGB-207 Baseline Transfusion Requirements of Subjects by Cohort

Parameter	Statistic	Cohort 1 (N=16)	Cohort 2 (N=8)	Total (N=24)
Volume RBC transfused (ml/kg/year)	N Median (Range)	16 192 (118- 251)	8 214 (142- 274)	24 201 (119- 274)
# RBC transfusions per year	N Median (Range)	16 17 (12, 37)	8 16 (12, 19)	24 16 (12, 37)
Weighted Avg. nadir Hg prior to transfusion (g/dl)	N Median (Range)	16 9.6 (7.5, 11.0)	8 9.6 (8.9, 10.2)	24 9.6 (7.5, 11.0)

Source: Reviewer Calculations from ADEF1 dataset

HGB-207 enrolled TDT subjects with all β thalassemia genotypes except β^0/β^0 . Eligibility included subjects homozygous for β^+ mutations, compound heterozygous for β^E , β^+ , or β^0 produce some β -globin in variable quantities (i.e., β^E/β^+ , β^E/β^0 , β^+/β^+ , β^+/β^0). Cohort 2 shows a higher prevalence of subjects with the β^+/β^0 genotype compared to Cohort 1, while Cohort 1 has a higher prevalence of subjects with the β^E/β^0 genotype. Subject genotypes of those in the EAP are summarized below.

Table 8 HGB-207 Genotype Summary

Parameter	Statistic	Cohort 1 (N=16)	Cohort 2 (N=8)	Total (N=24)
β^E/β^0	n (%)	5 (31.3)	1 (12.5)	6 (25.0)
β^+/β^0	n (%)	7 (43.8)	6 (75.0)	13 (54.2)
β^+/β^+	n (%)	4 (25.0)	1 (12.5)	5 (20.8)

Source: Reviewer calculations from ADSL dataset.

Reviewer Comment

Cohort 2 has relatively more subjects with β^+/β^0 genotype compared to Cohort 1, while Cohort 1 has a higher prevalence of β^E/β^0 genotype participants. The younger age at which subjects in Cohort 2 achieve an established transfusion dependence may be due to higher prevalence of β^+/β^0 .

6.1.10.1.3 Subject Disposition

Of 32 candidates who signed the consent/assent document five were screen failures (three with severe liver disease, one due to cardiac disease, and one with

β0 on both hemoglobin subunit beta (HBB) alleles). Three others withdrew consent before mobilization. Of the 24 eligible subjects in the ITT population who were mobilized with GCSF and plerixafor, one discontinued study before she was conditioned due to pregnancy, leaving 23 subjects who underwent conditioning with busulfan and administered beti-cel in the efficacy analysis population. While 23 subjects completed their Month 12 Visit, only 20 subjects had completed the study (Month 24 Visit) to the data lock date. Data are presented for the overall ITT population as well as by protocol-defined Cohorts: Cohort 1 and Cohort 2.

6.1.11 Efficacy Analyses

The goal of gene therapy for TDT is achievement of permanent discontinuation of pRBC transfusions by achieving a near- normal total Hb level. This is defined by transfusion independence.

6.1.11.1 Analyses of Primary Endpoint(s)

The primary efficacy endpoint was the proportion of subjects who achieved TI, which is defined as the weighted average hemoglobin (Hb) ≥ 9 g/dL without any pRBC transfusions for a continuous period of ≥ 12 months at any time during the study after beti-cel infusion. TI is defined as maintaining a weighted average Hb ≥ 9 g/dL without any pRBC transfusions for a continuous period of ≥ 12 months at any time during the study after beti-cel infusion, with the start of TI defined when subjects achieve an Hb ≥ 9 g/dL with no transfusions in the prior 60 days.

Subjects evaluable for TI are defined as subjects who have achieved TI, will not achieve TI in the study, or completed their parent study. Two subjects had insufficient follow up past infusion of beti-cel to be TI evaluable. Of 22 subjects evaluable for TI, 20 (90%) achieved TI at any time. Cohort 1 included 14 of 15 (93%) subjects who achieved TI at any time. In Cohort 2, 6 of 7 (86%) evaluable subjects achieved TI at any time. The results and calculated two-sided 95% confidence intervals are summarized below:

Table 9 HGB-207 Primary Efficacy Endpoint per Cohort (TI at any time)

Parameter	Statistic	Cohort 1 (N=15)	Cohort 2 (N=8)	Total (N=23)
TI evaluable	N	15	7	22
Achieve TI at anytime	n (%) 2-sided 95% CI	14 (93) 68.1, 99.8	6, (86) 42.1, 99.6	20 (91) 70.8, 98.9

Source: Reviewer calculations from ADEF2 dataset

Reviewer Comment

The primary efficacy endpoint analysis indicates that the majority of subjects had a response leading to transfusion independence for ≥ 12 months in both cohorts. A greater percentage of TI evaluable subjects in cohort 1 reached TI than in Cohort 2. It is the case that one subject in Cohort 2 was not TI evaluable with 13 months of follow up at data lock and did maintain an unsupported Hb of 10.3 g/dL, thus

was likely to reach TI by study end. Even so, this will make the TI rate in Cohort 2 seven of eight (86%), which is somewhat lower than 14/15 (93%) in Cohort 1. It is possible that this is due to small numbers of subjects. However, PD data indicate that of TI evaluable subjects, Cohort 2 does have lower Hb levels (median 12.1 g/dL) than Cohort 1 (median 10g/dL). One explanation may be that Cohort 2 had lower PB VCN and Hb^{AT87Q} levels at Month 6 compared to Cohort 1. Another reason is that Cohort 2 includes the youngest children, whereas Cohort 1 includes adults. Children normally express lower total Hb levels than adults.

6.1.11.2 Analyses of Secondary Endpoints

Complications of iron overload lead to shorter survival and present major challenges in managing patients who require lifelong regular/frequent transfusions. Therefore, besides transfusion independence, even reduction in transfusion burden is expected to provide clinically meaningful benefit for these patients. These secondary efficacy analyses included characterization of subjects achieving TI. Transfusion independence was maintained at Month 24 in 18 of 20 (90%), which included 14 of 15 (93%) subjects in Cohort 1 and 4/5 (80%) subjects in Cohort 2. Secondary endpoint analyses are outlined below.

Table 10 HGB-207 Additional Characterization of TI endpoint

Parameter	Statistic	Cohort 1 N=14	Cohort 2 N=6	Total N=20
TI Duration (months)	Median (range)	20.5 (19.2 - 21.6)	19.9 (15.7 - 20.8)	20.4 (15.7 - 21.6)
Weighted Hgb avg. during TI	Median (range)	12.1 (11.3 - 12.8)	10 (9.5 - 11.5)	11.7 (9.5 - 12.8)
Time from beti-cel to last pRBC before TI (months)	Median (range)	0.94 (0.5 - 2.2)	0.8 (0.5, 2.4)	0.87 (0.5 - 2.4)
Time from beti-cel to TI status (months)	Median (range)	15.4 (15.0 - 17.9)	10 (14.8 - 19.4)	15.4 (14.8 - 19.4)

Source: reviewer calculations from ADEF2, ADTTE data sets

Study HGB-207 also evaluated the reduction in transfusion requirements (mL/kg pRBCs transfused from 12 months post-beti-cel infusion through Month 24 [approximately a 12-month period] compared to the annualized mL/kg pRBC transfusion requirement during the 2 years prior to enrollment). In addition, Study HGB-207 compared the annualized number and volume of pRBC transfusions from 12 months post-beti-cel infusion through Month 24 compared to the annualized number and volume of transfusions during the 2 years prior to enrollment. The time from beti-cel infusion to last pRBC transfusion; and time from last pRBC transfusion to Month 24 were also evaluated.

As of data lock date, all subjects completed at minimum 12 months of follow-up post beti-cel infusion but not to Month 24, hence analysis was performed from beti-

cel to last follow-up. One subject in Cohort 1, and one in Cohort 2 failed to achieve TI following beti-cel. The results for reduction in transfusion requirements are listed below:

Table 11 HGB-207 Percent Reduction in RBC transfusions between Month 12 post beti-cel and last follow up compared to baseline (EAP)

Parameter	Statistic	Cohort 1 (N=15)	Cohort 2 (N=8)	Total (N=23)
<50%	n (%) 2-sided 95% CI	0 0,21.8	1 (12.5) 0.3, 52.7	1 (4.3) 0.1, 21.9
≥50%	n (%) 2-sided 95% CI	15 (100) 78.2, 100	7 (87.5) 47.3, 99.7	22 (95.7) 78.1, 99.9
≥60%	n (%) 2-sided 95% CI	14 (93.3) 68.1, 99.8	7 (87.5) 47.3, 99.7	22 (95.7) 72.0, 98.9
≥75	n (%) 2-sided 95% CI	14 (93.3) 68.1, 99.8	7 (87.5) 47.3, 99.7	21 (91.3) 72.0, 98.9
≥90	n (%) 2-sided 95% CI	14 (93.3) 68.1, 99.8	7 (87.5) 47.3, 99.7	21 (91.3) 72.0, 98.9
≥100	n (%) 2-sided 95% CI	14 (93.3) 68.1, 99.8	7 (87.5) 47.3, 99.7	21 (91.3) 72.0, 98.9

Source: reviewer calculations from ADEF1 dataset, CI calculations with The Clopper-Pearson Exact method is used to calculate the 2-sided 95% CI for the proportion of subjects meeting this criterion

Two subjects (b) (6) from Cohort 1, and (b) (6) from Cohort 2) who did not achieve TI had baseline transfusion needs of 192.9 and 208.0 mL/kg/year, and these were reduced to 93.6 and 161.9 mL/kg/year, respectively, from 12 months post-beti-cel infusion to last follow-up, for a reduction of 52% and 22%, respectively, compared to baseline.

A third subject (b) (6) achieved 100% reduction in RBC requirements but has only had a total of 13 months of follow up and has not yet met criteria for TI. Outcomes for these 3 subjects are summarized below:

Table 12 HGB-207 RBC Transfusion Reduction from 12 Months Post-beti-cel Infusion through Last Follow-Up, Among Subjects who failed to achieve TI, or are not TI evaluable

Subject ID	Baseline Annualized transfusion volume (ml/kg/yr)	Baseline Weighted average Hgb Nadir (g/dl)	Follow up Annualized transfusion volume (ml/kg/yr)	Follow-up Weighted average Hgb Nadir (g/dl)	% change from baseline	Follow up Duration (months)
Cohort 1 (b) (6)	192.9	9.5	93.6	7.9	-52	24
Cohort 2 (b) (6)	208.0	9.7	161.9	8.9	-22	25
Cohort 2 (b) (6)	207.9	10.2	0	10.1	-100	13

Source: Reviewer calculation from ADEF1 dataset

The overall results of transfusion reduction from 12 months post-beti-cel through last follow-up are listed below, broken down by cohort:

Table 13 HGB-207 RBC Transfusion Reduction from 12 Months Post-beti-cel Infusion through Last Follow-Up (EAP)

Group	N Median (Range)	Baseline Annualized transfusion volume (ml/kg/yr)	Baseline Weighted average Hgb Nadir g/dl)	Follow up Annualized transfusion volume (ml/kg/yr)	Follow up Weighted average Hgb Nadir (g/dl)	% change from baseline	Follow up Duration (months)
Cohort 1	N Median (Range)	15 192.9 (152.3, 251.3)	15 9.6 (7.5, 11.0)	15 0 (0, 93.5)	15 12.5 (7.9, 12.2)	15 -100 (-100, - 52)	15 24.3 (23, 28)
Cohort 2	N Median (Range)	8 213.6 (142.1, 274.4)	8 9.6 (8.9,10.2)	8 0 (0,161.9)	8 10.1 (8.9,11.3)	8 -100 (-100 - 7)	8 23.7 (13, 25)
Total	N Median (Range)	23 207.9 (142.1, 274.4)	23 9.5 (7.5,11.0)	23 0 (0, 161.9)	23 11.8 (7.9, 13.2)	23 -100 (-100, - 7)	23 24.3 (13, 28)

Source: Reviewer Calculations from ADEF1 dataset

Two TI-evaluable subjects did not achieve TI, one from each cohort. These 2 subjects had the lowest PB VCN values of all TI-evaluable subjects and were also producing the lowest amounts of Hb^{AT87Q} at Month 6. These subjects' beti-cel VCNs and %LVV+ Cells values were below the median values for all treated subjects, although not the lowest in the study. The reason for the relatively large decrease between beti-cel VCN and PB VCN for these subjects is believed to be due to a lower contribution of transduced long-term HSCs in the drug product for these subjects, the applicant was not able to provide an explanation. One of these two subjects did have leukocytosis three weeks before apheresis and also an upper respiratory tract infection was reported on Day 2 of mobilization, though the impact of this on the collected cells is not clear.

Reviewer Comment

With at least 12 months of follow up, most subjects achieved TI, and by definition a 100% reduction from baseline transfusion requirements, with exception of the two subjects who failed to achieve TI, plus the one who achieved a 100 % reduction in transfusion requirements but has only a total of 13 months follow up from beti-cel infusion.

The weighted nadir hemoglobin of the cohorts at Baseline, and on follow up post therapy with beti-cel for the overall population increased from 9.5 gm/dl at Baseline on transfusions, to 11.8 gm/dl in follow-up, suggesting robust hemoglobin synthesis in the treated population, and this also noted within the each of the two

cohorts. The two subjects who failed to achieve TI had slightly lower nadir hemoglobin levels during follow up than at Baseline. Per protocol, transfusions were to be avoided for Hb ≥ 9 g/dL unless the need is medically justified, but it was recommended that subjects receive RBC transfusions if Hb < 7 g/dL, and for clinically symptomatic anemia, irrespective of Hb level.

Two subjects did fail to achieve TI. These subjects tended to have somewhat low beti-cel VCNs and %LVV+ Cells values, and had the lowest PB VCN values of all subjects, however, the reason for these observations is not clear. It is possible that individuals might have differences in the susceptibility of their HSPCs to transduction by LVV.

Unsupported Total Hemoglobin

Unsupported total Hb levels are defined as those measurements with no RBC transfusion in the prior 60 days, and these were assessed over time at Month 6, Month 9, Month 12, Month 18 and 24. The table below contains results by TI status.

Table 14 HGB-207 Unsupported Total Hb Over Time, By Cohort (TI Subjects)

Group	Statistic	Month 6	Month 12	Month 18	Month 24
Cohort 1 Overall	N	15	15	13	13
	Median	11.8	12.3	12.3	12.6
	(Range)	8.4, 13.3	8.4, 13.1	11.2, 13.5	10.9, 13.3
Cohort 1 only TI	N	14	14	13	13
	Median	11.8	12.3	12.3	12.6
	(Range)	11.1, 13.3	10.7, 13.1	11.2, 13.5	10.9, 13.3
Cohort 2 Overall	N	7	6	5	4
	Median	9.8	10	10.2	10.3
	(Range)	9.3, 12.1	9.7, 12.5	9.5, 11.7	9.5, 11.8
Cohort 2 only TI	N	6	5	5	4
	Median	9.8	10	10.2	10.3
	(Range)	9.3, 12.1	9.7, 12.5	9.5, 11.7	9.5, 11.8
Total	N	22	21	18	17
	Median	11.5	11.9	11.8	12.5
	(Range)	8.4, 13.3	8.4, 13.1	9.5, 13.5	9.5, 13.3
Total who had TI	N	20	19	18	17
	Median	11.7	12.1	11.8	12.5
	(Range)	9.6, 13.3	9.7, 13.1	9.5, 13.5	9.5, 13.3

Source: Reviewer calculations from ADLB dataset

Reviewer Comment

Overall, the hemoglobin level appears stable, with a possible trend towards slight increase in median levels of unsupported hemoglobin over time, with the values at Month 24 exceeding earlier time points in all subgroups.

Iron Overload

Characterization of use of iron chelation and/or therapeutic phlebotomy among all subjects was also analyzed to include: proportion of subjects who have not received iron chelation therapy for ≥ 6 months following beti-cel infusion; time from last iron chelation use to last follow-up; proportion of subjects using therapeutic phlebotomy; and annualized frequency of phlebotomy use per subject following beti-cel infusion. Chelation was held before mobilization and after treatment, to optimize mobilization, and prevent possible iatrogenic engraftment delays from certain chelators. Time to resumption of chelators and further management of iron overload was left to the investigator. These results are summarized below.

Table 15 HGB-207 Iron Chelation Therapy Post beti-cel Infusion (EAP)

Parameter	Statistic	Cohort 1	Cohort 2	Total
Subjects with ≥ 6 Months Follow Up	n (%)	15 (100)	8 (100)	23 (100)
Subjects who stopped chelation post beti-cel infusion	n (%)	11 (73.3)	3 (37.5)	14 (60.9)
Months from stopping chelation to last follow up	N Median (Range)	11 23.3 0, 24.6	3 13.0 4, 23.1	14 23.1 0, 24.6
Subjects who stopped iron for ≥ 6 months post beti-cel	n (%)	9 (60)	2 (25)	11 (47.8)
Subjects who used Phlebotomy therapy post beti-cel	n (%)	7 (46.7)	0	7 (30.4)
Annualized phlebotomy procedures from beti-cel to last follow up	N Median (Range)	7 2.6 0.5, 12.5	0	7 2.6 0.5, 12.5

Source: Calculated by reviewer from ADSL dataset

As iron overload is a ubiquitous complication seen in subjects with TDT, and one which might respond to effective therapy that may obviate the need for transfusions and promote normalization of iron stores, iron stores were evaluated sequentially in liver and heart. Invasive monitoring (biopsy) was avoided.

Reviewer Comment

Almost twice as many Cohort 1 subjects stopped chelation than Cohort 2 subjects. And subjects in Cohort 1 remained off of chelation almost twice as long as Cohort 2, and a larger % of Cohort 1 remained off chelation for at least 6 months. Since Cohort 2 subjects were younger than cohort 1, one would predict that they had accumulated less iron in their shorted exposure to transfusions and would need less chelation. Cohort 2 had a larger improvement (decrease) in their median ferritin levels from baseline to Month 24 on study (shown in the next

section). Since resumption and duration of chelation/phlebotomy was at the investigator's prerogative, perhaps Cohort 2 subjects' lower ferritin values led to less aggressive iron reduction therapy on the study. Phlebotomy was only used in Cohort 1 subjects. This was likely largely due to their higher unsupported Hb levels compared with Cohort 2. (i.e., the lower Hb in Cohort 2 was a contraindication for phlebotomy).

Study HGB-207 subjects had iron assessment with liver iron content (LIC) and cardiac T2* by MRI, and serum ferritin, serum iron, and transferrin in peripheral blood. Analysis compared TI versus non-TI subjects with respect to iron stores post beti-cel infusion. Serum levels of parameters that indicate iron loads were also evaluated, but are fraught with confounding from acute inflammation, and suboptimal reliability in general.

The magnitude of change in hepatic iron burden over time assessed with magnetic resonance imaging (MRI) LIC values at baseline compared with those at Month 12 and Month 24 was analyzed for HGB-207 subjects. After beti-cel treatment, resumption of chelation was at investigator's discretion and all subjects did not restart chelation after beti-cel infusion. The reviewer performed the analyses using data contained in ADEF3 dataset, reaching the following conclusions:

- Baseline LIC values were highly variable among subjects.
- Median LIC values trended upwards between baseline and Month 12.
- Median LIC values tended to decrease towards baseline by Month 24 for most subjects. This trend was clearer in TI subjects than those who did not achieve TI
- Six subjects had liver biopsy at screening showing elevated LIC (7 mg/g to 16.8 mg/g), no follow-up biopsies were performed after beti-cel infusion.

Reviewer Comment

Iron overload tends to progress between enrollment and month 12 post beti-cel. Following this point, LIC appears to level off and then trend towards baseline. Although the group who did not achieve TI was very small (N=2), making drawing of conclusions difficult, the group (N=17) who achieved TI, had improved LIC trend compared to the non-TI group. The group with TI had a median 9% change from baseline in LIC, whereas those who did not achieve TI had a 38% median change from baseline. Initiation of phlebotomy was at the investigator's discretion in the study.

Cardiac Iron Burden

According to the sponsor, LIC greater than 15 mg/g dry weight is associated with an increased risk of cardiac disease and early death, whereas levels between 7 and 15 mg/g enhance the risk of hepatic fibrosis and endocrine complications.

Additional evaluation of iron burden over time was performed by measuring the change in cardiac T2* on MRI from baseline to Month 12 and Month 24. T2* of the

ventricular septum measured with MRI is reciprocal to cardiac iron burden, thus permitting noninvasive assessment of iron overload in this important organ. The Reviewer drew the following conclusions after performing analyses of T2 over time from baseline to month 12 and month 24: There was a trend of increasing iron (i.e., decreasing cardiac T2*) starting from baseline to Month 12 and Month 24, even in those achieving TI.

Serum ferritin values were also monitored over time, to reflect body iron burden. Reviewer analysis of ferritin trends over time led to the following conclusion:

- Ferritin levels at baseline were highly variable
- Ferritin levels peaked at Month 3
- Ferritin levels then started dropping towards of below baseline. Most (16 of 20 subjects with Month 24 data) had ferritin levels at Month 24 that were lower than baseline.

Reviewer Comment

Similar to LIC, ferritin levels increase following beti-cel infusion, possibly due to ferritin being an acute phase reactant. After peaking, the ferritin levels trended towards the baseline, and even below the baseline levels. TI achievers tended to have a greater decrease in ferritin at month 24 compared to baseline, though the number of non-TI achieving subjects was very small at N=2.

Health Related Quality of Life

The Applicant included patient reported outcome (PRO) measures in the clinical study design. Health-Related Quality of Life (HRQoL) was assessed by questionnaires as a secondary endpoint. Quality of life assessments were collected at Screening, Month 3 (beginning with HGB-207 Protocol Version 1.5), Month 6, Month 12, Month 18, and Month 24 Visits. The tools evaluated health-related quality of life (HRQoL) over time including Month 12 and Month 24 as compared to baseline, using the following tools: (b)(4) ; parent general core and general core); (b)(4) (parent general core and general core) and (b)(4) (Youth version; (b)(4)

Results of all the tools were evaluated but will only be summarized here to highlight the relatively small changes in the scores relative to the range of the median and standard deviation of the mean values. For example, considering the (b)(4) tool, the results showed small changes, and the significance of which is difficult to interpret. Among 9 subjects with available data for physical component summary of (b)(4) , the overall results from baseline to Month 24 suggested an increase in the score from median of 52.6 (range 46.5-61.2) to 54.8 (range 45.2-61), a change of 0.48 (range -4.3 to 7). However, the difference is quite small. Among those who had TI, the score increased by 2.57, but again, the range was -9.4 to 7.6).

Reviewer Comment

Small numbers of subjects with evaluable QOL data and modest changes make drawing inferences from these outcomes challenging, especially given the limitations of QOL assessments in uncontrolled, open-label trials. As with time-to-event endpoints, interpretation of patient-reported outcomes is challenging in uncontrolled clinical trials, because it is unclear to what extent the outcomes can be attributed to the treatment effect of the regimen vs. to underlying disease and patient characteristics. Furthermore, the Applicant did not seek a labeling claim based on QOL data and these data were not incorporated in the PI. Finally, one of the tools used was generally designed and intended to be used in patients with advanced cancers, and its validity in patients with hemoglobinopathy remains to be demonstrated.

6.1.11.3 Subpopulation Analyses

Subgroup analyses included evaluation of outcomes in young children (vs. adolescents/ adults) and efficacy differences between males vs. females. Because TDT subjects continue to accumulate iron from ongoing RBC transfusions despite chelation therapy, iron loads in organs may be expected to rise with age. Younger pediatric subjects might therefore be expected to have lower iron stores. Beti-cel might affect iron overload parameters differently in different age cohorts. Analysis to compare iron metrics between study Cohort 1 (subjects ≥ 12 years old) and Cohort 2 (subjects < 12 years old) was undertaken.

HGB-207 used a number of parameters to assess iron overload. One of the tools used was cardiac T2* score measured per MRI, expressed in milliseconds (ms). Cardiac T2* evaluates the ventricular septum iron content and is **reciprocal** of iron load. Candidates were ineligible for the study if their cardiac iron overload were not severe, therefore, a cardiac T2* < 10 ms (indicative of very high iron), was exclusionary.

Cardiac Iron Burden Between Cohort 1 (age ≥ 12 to ≤ 50 years) and Cohort 2 (< 12 years old).

At baseline, Cohort 1 (N=15) cardiac T2* score was median 36.4ms (20.6, 50.9), mean 37.0 (7.3), whereas Cohort 2 (N=8) had a median cardiac T2* score of 37.4ms (23.3, 56.8), and mean 38.4ms (11.6). Therefore, baseline cardiac T2* scores were comparable, presumably due to impact of ongoing iron chelation.

At month 24, Cohort 2 subjects (N=4) had a median cardiac T* score of 36ms (29.7, 38.6, Mean 35.2ms (3.9), whereas Cohort 1 (N=15) had Median cardiac T* score of 33.3ms (14.5, 46.5), mean 33.2ms (8.6). Both cohorts experienced a modest decrease in cardiac T2* score (i.e., a slight *increase* in cardiac iron load). However, because cardiac T2* scores dropped more in Cohort 1 vs. Cohort 2,

cardiac iron load had increased slightly in Cohort 1 as compared to Cohort 2 between baseline and Month 24. Please see table below:

Table 16 HGB-207 Cardiac T2 Trends from Baseline to Month 24 Score per Cohort

Parameter	Statistic	Cohort 1 (Age ≥12 to ≤50)	Cohort 2 (< 12YO)
Baseline	N	15	8
CARDIAC	Median (range)	36.4 (20.6, 50.9)	37.4 (23.2, 56.8)
T2* SCORE	Mean (SD)	37.0 (7.3)	38.4 (11.6)
Month 24	N	15	4
CARDIAC	Median (range)	33.3 (14.5, 46.5)	36.3 (29.7, 38.6)
T2* SCORE	Mean (SD)	33.2 (8.6)	35.2 (3.9)

Source: Reviewer calculation from ADEF3 dataset

Reviewer Comment

It is not clear why the older subjects in Cohort 1 would experience a larger increase in cardiac iron stores while on study, between baseline and Month 24. Admittedly, the changes between baseline and Month 24 in both groups appear small and of dubious clinical significance. Cohort 1 subjects tended to undergo a more aggressive iron lowering regimen with earlier and more prevalent resumption of chelators and about 30% of Cohort 1 even underwent phlebotomy, as compared to none of cohort 2 subjects. Admittedly, the T2 differences are small, and the population size, especially in cohort 2, is limited. Also, a 24-month duration of follow up may be insufficient to capture the impact of beti-cel on cardiac iron burden.*

Liver Iron Content (LIC)

Hepatic iron toxicity is an important sequela of TDT. Baseline LIC was substantially higher in cohort 1 than cohort 2, likely in keeping with longer duration of dependence on RBC transfusion among the older (cohort 1) group. Between baseline and Month 24, LIC decreased modestly in Cohort 1 subjects. However, LIC increased between baseline and Month 24 in Cohort 2 participants. The reason for this is unclear, however, Cohort 1 subjects tended to resume chelation earlier after beti-cel than Cohort 2, and some Cohort 1 subjects also underwent phlebotomy. These data are tabulated below:

Table 17 HGB-207 LIC Trends from Baseline to Month 24 Score per Cohort

Parameter	Statistic	Cohort 1 (>12 years)	Cohort 2 (< 12 years)
Baseline	N	15	8
LIC	Median (range)	7.2 (1, 41)	3.8 (1.8, 9)
	Mean (SD)	10 (10.5)	4.2 (2.3)
Month 24	N	14	5
LIC	Median (range)	5.5 (1.4, 24.9)	4.1 (2.4, 7.1)
	Mean (SD)	7.8 (7)	4.7 (1.8)

Source: Reviewer calculation from ADEF3 dataset

Laboratory iron Assessment with Ferritin

Ferritin correlates directly with iron stores, although it is also an acute phase reactant. In HGB-207 subjects, median ferritin levels decreased in both cohorts between Baseline and Month 24, more notably in Cohort 2 than Cohort 1, despite the more aggressive chelation and phlebotomy regimen implemented in Cohort 1. One explanation is that ferritin, due to being an acute phase reactant, might not accurately reflect iron stores. Results are summarized in the following table:

Table 18 HGB-207 Ferritin Trends from Baseline to Month 24 Score per Cohort

Parameter	Statistic	Cohort 1 (>12 YO)	Cohort 2 (< 12YO)
Baseline Ferritin	N	15	8
	Median (range)	4611.2 (784, 22516.9)	3677.5 (1467.4, 5413.5)
	Mean (SD)	5971.2 (5523.9)	3464 (1447.6)
Month 24 Ferritin	N	15	5
	Median (range)	3957.3 (211, 18970.8)	1240.4 (788.8, 6256.2)
	Mean (SD)	4785.9 (5180.9)	2156.4 (2303.2)

Source: Reviewer calculation from ADLB dataset

Reviewer Comment

Overall, the younger subjects of Cohort 2 tended to have at least slightly lower baseline iron burden per cardiac T2, LIC and ferritin than Cohort 1. Between Baseline and Month 24, Cohort 1 experienced a slightly greater worsening cardiac iron burden than Cohort 2; conversely, Cohort 1 appeared to have greater improvement in liver iron burden than Cohort 2. Both cohorts' ferritin levels improved, Cohort 2 improving more than Cohort 1. A longer follow up will likely be needed to better understand beti-cel impact on iron levels in TDT patients.*

The primary efficacy endpoint hinges on maintenance of a specific hemoglobin concentration, and regulation of hemoglobin levels in post-pubertal humans is impacted by sex hormones, particularly testosterone in males. Male subjects may be expected to have higher hemoglobin expression than female subjects, therefore an efficacy endpoint based on a specific hemoglobin level, un-adjusted for sex, might give males an advantage in meeting the TI endpoint.

Table 19 HGB-207 Proportion of Subjects Who Have Achieved TI at Any Time, by Sex (EAP)

Group	Statistic	Males (N=11)	Females (N=12)	Total (N=23)
TI non evaluable	N	0	1	1
TI evaluable	N	11	11	22
Subjects with TI at anytime	n (%) 2-sided 95% CI	11 (100) 71.5, 100	9 (82) 48.2, 97.7	20 (91) 70.8, 98.9

Source: Reviewer calculations from ADEF2

The analysis suggests that a higher proportion of males achieved TI than females.

Reviewer Comment

The proportion of males who achieved TI at any time was 100%, compared with 82% of females. However, given the small numbers of subjects of each sex, this discrepancy may be by chance. One explanation for the greater prevalence of TI achievers among males is that normal males, as a group, have physiologically higher Hb than females. As this study defines TI based on maintaining a single arbitrary Hb value for both sexes, (i.e., weighted average Hb of ≥ 9 g/dL), a greater % of the male subjects will have Hb above this modest Hb threshold than the % of female subjects.

6.1.11.4 Dropouts and/or Discontinuations

During the COVID-19 pandemic, according to the statistical analysis plan (SAP), subjects were not required to visit clinical trial sites and may have miss scheduled visits for assessments per study protocol. As a result, some study visits and lab collections have not been performed for some subjects. Some study visits were conducted in an alternate manner to include assessments being collected locally, some assessments were collected at unscheduled or out-of-window visits or at the following scheduled site visit, and some assessments were missed assessments. For the primary endpoint (TI) when the Hb at the end of the 12-month period needed to confirm TI is not available given the visit has been cancelled due to COVID-19, the last observation carried forward method will be used to impute the missing value provided 1) the weighted Hb from t0 up to the latest observed Hb is ≥ 9 g/dL, 2) the subject has at least 6 months of observed Hb from t0 to last follow-up, and 3) the subject has remained off pRBC transfusions from t0 to last follow-up. Missed scheduled visits prior to TI confirmation will not be imputed for the primary analysis. As a sensitivity analysis, the lowest value observed post t0 will be used to impute any missing values during the TI period. The above imputation rules will only be applied if the success criterion for TI cannot be reached due to missing Hb data from COVID-19 for regulatory submission. Eight subjects had 23 altered or virtual study visits in lieu of scheduled site. Per the SAP, imputation for missed Hb assessments could be used if the success criteria for TI could not be reached due to COVID-19. However, as of 09 March 2021, the missing Hb lab data are limited and did not impact the TI derivation. The COVID-

19 imputation algorithm described in the SAP therefore was not ultimately applied in the analysis for this interim HGB-207 CSR.

Subjects who enrolled in HGB-207 but discontinued prior to drug product infusion were followed for at least 30 days after any invasive study procedure (e.g., mobilization, liver biopsy) before withdrawal, and ongoing AEs were followed for 30 days. One subject from Cohort 1 discontinued after start of mobilization but before myeloablation and beti-cel infusion.

6.1.11.5 Exploratory and Post Hoc Analyses

Exploratory endpoints included the PD endpoint of β^{A-T87Q} -globin expression over time, including Month 6, 9, 18 and 24, and correlation of β^{A-T87Q} -globin expression at early time points post beti-cel infusion to β^{A-T87Q} -globin expression at later time points, as well as clinical outcomes. Analyses of these are discussed in the following section.

Hb^{AT87Q} in Peripheral Blood

Successful treatment with beti-cel requires transgene expression (β^{A-T87Q} -globin production) in the appropriate target cell population (cells of the erythroid lineage). Transgene expression requires successful transduction of HSCs, engraftment of those transduced HSCs in the subject, differentiation of transduced HSCs with subsequent erythroid compartment reconstitution, transcription and translation of the LVV-inserted transgene in cells of the erythroid lineage, and combination of β^{A-T87Q} -globin with α -globin to form functional Hb (Hb^{AT87Q}) in mature RBCs. To measure the Hb^{AT87Q} level, the ratio of the β^{A-T87Q} -globin peak area to total peak area for all β -like-globins (such as βA -T87Q, βA , γA , γG , and δ -globins) was determined by RP-HPLC, and then this ratio was multiplied by the total Hb (g/dL) measured in a second blood sample to obtain a g/dL estimate. The levels of Hb^{AT87Q} are listed below:

Table 20 Hb^{AT87Q} Over Time Overall (EAP)

Group	Statistic	Month 6 Hb ^{AT87Q} (g/dL)	Month 12 Hb ^{AT87Q} (g/dL)	Month 18 Hb ^{AT87Q} (g/dL)	Month 24 Hb ^{AT87Q} (g/dL)
Cohort 1	N	13	15	15	15
	Median (Range)	9.3 3.3, 10.6	9.3 3.6, 10.6	9.3 0.2, 10.6	9.4 0.9, 11.4
Cohort2	N	7	8	5	5
	Median (Range)	6.9 1.1, 10.5	7.4 0.9, 9.1	8.4 5.0, 10	7.4 0.3, 8.6
Total	N	20	23	20	20
	Median (Range)	8.7 1.1, 10.6	8.6 0.9, 10.6	9.3 0.2, 10.5	8.8 0.3, 11.4

Source: Reviewer calculations from ADPD dataset.

Note: Assessments assigned to the scheduled visit per midpoint windowing.

Note: Subjects (b) (6) in Cohort 1 did not have an available Month 6 Hb^{AT87Q} value due to a

central lab processing error. This was noted as a protocol deviation.

Note: Subject (b) (6) in Cohort 2 did not have an available Month 6 Hb^{AT87Q} value, and Subjects (b) (6) in Cohort 2 did not have available Month 18 Hb^{AT87Q} values due to COVID-19 restrictions.

Note: Month 24 Hb^{AT87Q} data for Subject (b) (6) and Month 3 Hb^{AT87Q} data for Subject (b) (6) were switched in the database and thus incorrectly entered. This error was corrected after the database lock date.

Immediately post beti-cel infusion, Hb^{AT87Q} levels are low as transduced HSCs engraft and initiate erythropoiesis. However, as transduced HSCs give rise to progeny carrying the transgene and go on to produce RBCs, Hb^{AT87Q} levels increase and eventually plateau. In general, Hb^{AT87Q} levels plateaued by Month 6 after beti-cel infusion at a median of > 8 g/dL. According to the Applicant, three main factors influence Hb^{AT87Q} levels in a subject. Firstly, PB VCN shows a correlation with Hb^{AT87Q}, and in fact, the 4 subjects with the lowest Hb^{AT87Q} levels at Month 6 are those with the lowest PB VCN at Month 6 (Subjects (b) (6)). Secondly, pRBC transfusions are known to suppress endogenous hematopoiesis. Subject (b) (6) had sufficient levels of Hb^{AT87Q} and endogenous Hb to allow this subject to be transfusion-free from approximately Month 3 to Month 14, and Hb^{AT87Q} levels in this subject plateaued approximately between Months 6 and 12. However, this subject could not sustain a total Hb above 8 g/dL, and restarted pRBC transfusions at approximately Month 14, after which time Hb^{AT87Q} levels declined. Thirdly, similar to the effect of transfused blood on Hb^{AT87Q}, levels of endogenous Hb production can affect levels of Hb^{AT87Q}, with higher levels of endogenous Hb leading to lower levels of Hb^{AT87Q}. Subjects (b) (6), all had similar PB VCN at Month 6, with varying expression of Hb^{AT87Q} at Month 6. The difference in levels of Hb^{AT87Q} between these subjects is inversely related to the levels of endogenous Hb each subject produced, with the highest Hb^{AT87Q} levels seen in the subject with lowest endogenous Hb.

Reviewer Comment

Beti-cel treatment leads to production of Hb^{AT87Q} and total hemoglobin in most subjects. Persistent expression of Hb^{AT87Q} in the subjects achieving TI explains the mechanism of action behind beti-cel's efficacy. This appears to be related to the PB VCN. Moreover, among subjects who achieve TI, Hb^{AT87Q} complements any endogenous hemoglobin variants and corrects anemia sufficiently to obviate need for transfusion. The Hb^{AT87Q} levels appear to be inversely proportional to the quantity of endogenous Hb produced.

6.1.12 Safety Analyses

6.1.12.1 Methods

Key materials used for the safety review included:

- The BLA application electronic submission

- Applicant submissions in response to the review team's information requests
- Proposed labeling for beti-cel
- Published literature
- Prior regulatory history

The clinical review of safety was primarily based upon analysis of 24 subjects in Study HGB-207 at the primary data cutoff of 09 March 2021. Analysis datasets (ADaM datasets) were used for the safety analysis. Analyses by the clinical reviewer for safety were performed using JMP 16. All narratives and relevant case report forms (CRFs) were reviewed for all serious adverse events (SAEs) that occurred in the primary safety population. Adverse events (AEs) were coded using the Medical Dictionary for Regulatory Activities (MedDRA) version 23.0, and AE severity was graded using the National Cancer Institute's (NCI's) Common Terminology Criteria for Adverse Events (CTCAE) version 4.03. Concomitant medications were coded using the World Health Organization (WHO) Drug Dictionary March 2016 or later. Some AEs are presented throughout this review as grouped terms as defined by the review team. The complete list of FDA's grouped terms is presented in APPENDIX. Unless otherwise specified, all analyses and tables were generated by the FDA clinical reviewer.

The safety analysis set included all subjects treated with any dose of beti-cel. All AEs were collected from the signing of the informed consent form (ICF) until 24 months after beti-cel infusion. Serious adverse events (SAEs) were defined as any AEs that met at least one of the following criteria: fatal, life threatening, required inpatient hospitalization or prolongation of existing hospitalization, resulted in persistent or significant disability, resulted in congenital anomaly or birth defect, or resulted in any other medically important serious event. SAEs were collected from the time of screening. Treatment-emergent adverse events (TEAEs) were defined as all AEs occurring after initiation of beti-cel administration through the Month 24 visit after beti-cel infusion.

Reviewer Comment

The Applicant's methodology for determining ADRs differs from the one the reviewer team used. The Applicant defined ADRs as: 1) an AE that is consistent with the pharmacology of the drug, temporality, and the consistency of the pattern of symptoms across studies, or if the AE was assessed by the Applicant as related to beti-cel. This definition is subjective. Furthermore, beti-cel therapy is necessarily preceded by myeloablative conditioning chemotherapy; therefore, it is often difficult to parse out the causality of AEs. For the above-mentioned reasons and in order to decrease bias in this uncontrolled study, the reviewer considered any AE that occurred after initiating beti-cel treatment as an ADR. Otherwise, the safety reporting methods appear acceptable.

Overview of Adverse Events

Overall Exposure:

The safety population included the ITT subjects, N=24, defined as those who initiated any study procedure starting with mobilization. Select endpoints are also presented by Cohort 1 (≥ 12 years of age) and Cohort 2 (< 12 years of age). No subjects in the ITT population were excluded from the safety analyses.

Relevant Characteristics of the Safety (ITT) Population

Safety population demographic characteristics for HGB-207 are summarized by treatment in the table below. Subjects were reasonably matched by sex with 15 females and 12 males. The majority of patients in the safety population were white and identified as non-Hispanic/Latino.

Table 21 Demographics in HGB-207 ITT Safety population (N=24)

Parameter	Statistic	Cohort 1 (≥ 12) N=16	Cohort 2 (< 12) N=8
Males	n (%)	8 (42%)	4 (50%)
Females	n (%)	11 (58%)	4 (50%)
Age	Median (range)	20 (12, 35)	8 (4,11)
White Race	n (%)	6 (32)	9 (25%)
Asian Race	n (%)	12 (63)	4 (50%)
Other Race	n (%)	1 (5)	2 (25%)
Hispanic ethnicity	n (%)	0	1 (13%)
Non-Hispanic Ethnicity	n (%)	19 (100%)	6 (75%)
Not reported Ethnicity	n (%)	0	1 (13%)

Source: Reviewer Calculations from ADSL dataset

Reviewer Comment

1. Adequacy of the safety database

The demographics of the safety population are consistent with those of the intended patient population. Due to the nature of the product requiring myeloablative chemotherapy conditioning, the study has no control population. The safety database is relatively small, owing to the rarity of TDT; however, it is considered sufficient for safety evaluation of beti-cel for TDT.

2. Demographics Comment

The safety dataset was mostly comprised of Asian subjects, due to the regions of the World most affected by TDT, and immigration patterns. Safety results are generalizable to the beta thalassemia patient population given that the majority of patients are Asian, with Whites from the Mediterranean region a close second demographic. In addition, the safety population was

comprised of younger patients with a mean age of 31-32 years. Because of the significant morbidities associated with TDT, many patients do not have a normal lifespan and suffer from premature death; also subjects over 50 years old were excluded.

Exposure to plerixafor and G-CSF for Mobilization/Apheresis

All 24 subjects underwent mobilization of stem cells to allow for manufacture of beti-cel using G-CSF and plerixafor. Average daily dose ranges were: 1) G-CSF 5.0 to 10.2 µg/kg/day for splenectomized subjects and 6.8 to 11.6 µg/kg/day for subjects with an intact spleen, and 2) plerixafor 0.22 to 0.28 mg/kg/day for all subjects per the protocol recommended dosing. G-CSF was adjusted for spleen presence due to response differences. After G-CSF for 5 to 7 days, subjects underwent apheresis. A single cycle of mobilization was sufficient in 19 subjects (79%) to reach the target CD34+ cell dose, whereas the other 5 subjects (21%) required 2 cycles of mobilization. One subject discontinued from the study after one cycle of mobilization upon discovering that she was pregnant and was not conditioned or given beti-cel.

Exposure to busulfan for conditioning

A total of 23 subjects received conditioning with busulfan, of whom 4 had daily dosing for 4 days and 9 subjects received busulfan every 6 hours for up to 4 days. Thirteen of 23 subjects had estimated average busulfan AUC within the protocol recommended range (daily AUC of 3800 to 4500 µM*min). One subject had AUC below the recommended range and 9 were above this range. Average daily busulfan dose and estimated average AUC for subjects in Study HGB-207 are presented in the table below:

Table 22 HGB-207 Busulfan exposure per cohort

Parameter	Statistic	Cohort 1 (N=15)	Cohort 2 (N=8)	Total N=23
Average Daily Dose (mg/kg/day)	Median (Range)	3.5 (2.6, 5.0)	3.6 (3.1, 4.6)	3.5 (2.6, 5.0)
Estimated Average Daily AUC (µM*min)	Median (Range)	4544.9 (3708,7847)	4145.3 (3958, 7497)	4337.4 (3708,8947)

Source: Reviewer Calculations from ADPP dataset

Exposure to beti-cel

Per protocol, dose was $\geq 5.0 \times 10^6$ CD34+ cells/kg on Day 1. Of 23 subjects who received beti-cel, 19 subjects (83%) received one beti-cel lot, and four subjects (17%) received two beti-cel lots. The reason for multiple lots was that sometimes a single apheresis did not generate sufficient cell collection and had to be repeated. Subjects received a median (min, max) cell dose of 8.10 (5.0, 19.9) $\times 10^6$ CD34+ cells/kg, with median (min, max) beti-cel VCN of 3.263 (1.9, 5.6) c/dg and median (min, max) %LVV+ Cells of 79.28% (34%, 90%).

Duration of Safety Follow-up

Safety was monitored in HGB-207 for up to 24 months. Median length of follow-up in Study HGB-207 for the 23 treated subjects was 24.3 (range 13.0, 27.5) months, and 20 subjects completed the Month 24 Visit.

Reviewer Comment

HGB 207 subjects had adequate dosing of busulfan, although one subject had a target AUC, that was slightly lower and 9 had AUCs exceeding the target AUC. The subjects had adequate exposure to the study drug, as all met the minimal recommended dose of beti-cel.

Adverse Events

The system organ classes (SOC) with the most AEs in this study was Blood and lymphatic system disorders (24/24; 100%) and Gastrointestinal disorders (24/24, 100%).

Most commonly experienced AE, noted in $\geq 30\%$ of subjects, included thrombocytopenia (24/24, 100%), neutropenia (18/24, 75%), stomatitis (18/24, 75%), anemia (17/24, 71%), vomiting (16/24, 67%), nausea (14/24, 58.3%), leukopenia (13/24, 54%), pyrexia (13/24, 54%), procedural pain (11/24, 46%), headache (10/24, 42%), cough (10/24, 42%), epistaxis (10/24, 42%), febrile neutropenia (9/24, 38%), abdominal pain (9/24, 38%), diarrhea (9/24, 38%), alanine aminotransferase increased (9/24, 38%), and alopecia (8/24, 33%). The AEs are reported below:

Table 23 Non laboratory based TEAEs Experienced by 10% or More Subjects D1 to Month 24. N=23.

System Organ Class	Group Term	Subjects With All Gr AEs	% ALL Gr AEs	% Gr 3 and Gr4
Blood and lymphatic system disorders	Febrile neutropenia	9	39%	35%
Gastrointestinal disorders	Abdominal pain	9	39%	4%
Gastrointestinal disorders	Anal hemorrhage	3	13%	0%
Gastrointestinal disorders	Constipation	6	26%	0%
Gastrointestinal disorders	Diarrhea	7	30%	0%
Gastrointestinal disorders	Dyspepsia	3	13%	4%
Gastrointestinal disorders	Gingival bleeding	3	13%	4%
Gastrointestinal disorders	Mucositis	18	78%	61%

Gastrointestinal disorders	Nausea	7	30%	4%
Gastrointestinal disorders	Vomiting	12	52%	0%
General disorders and administration site conditions	Chest pain	3	13%	0%
General disorders and administration site conditions	Fatigue	3	13%	0%
General disorders and administration site conditions	Pyrexia	13	57%	17%
Hepatobiliary disorders	Veno occlusive liver disease	4	17%	13%
Infections and infestations	Folliculitis	3	13%	0%
Infections and infestations	Pneumonia	3	13%	4%
Infections and infestations	Sepsis	4	17%	17%
Infections and infestations	URTI	8	34%	0%
Infections and infestations	Viral infection	5	22%	0%
Injury, poisoning and procedural complications	Transfusion reaction	4	17%	0%
Metabolism and nutrition disorders	Decreased appetite	5	22%	1%
Musculoskeletal and connective tissue disorders	Musculoskeletal pain	6	26%	0%
Nervous system disorders	Headache	7	30%	0%
Respiratory, thoracic and mediastinal disorders	Cough	10	44%	0%
Respiratory, thoracic and mediastinal disorders	Dyspnea	3	13%	0%
Respiratory, thoracic and mediastinal disorders	Epistaxis	10	44%	22%

Respiratory, thoracic and mediastinal disorders	Oropharyngeal pain	4	17%	0%
Skin and subcutaneous tissue disorders	Alopecia	7	30%	0%
Skin and subcutaneous tissue disorders	Pigmentation disorder	5	22%	0%
Skin and subcutaneous tissue disorders	Pruritus	6	26%	0%
Skin and subcutaneous tissue disorders	Rash	4	17%	0%

Source: Calculated by reviewer from ADLB dataset. Please see Appendix for details of Group Terms

Reviewer Comment

AEs such as alopecia are expected with cytotoxic chemotherapy. Likewise, gastrointestinal events including mucositis, and nausea/vomiting are known complications of chemotherapy myeloablation. The incidence and extent of these AEs is the expected side effect profile of busulfan. Infections and febrile neutropenia are also common and expected events during post myeloablation period. These occurred early and were attributed by the reviewer to conditioning.

Endocrine and reproductive systems: Two subjects experienced Infertility (i.e., Hypogonadism and Androgen deficiency). TDT subjects with chronic iron overload have increased risk for endocrinopathies, and these may be exacerbated by conditioning chemotherapy.

Hepatobiliary Disorders

Myeloablative conditioning with busulfan has been associated with liver toxicity, including veno-occlusive disease (VOD) and consequently, subjects were prophylaxed and monitored for this complication. Four subjects experienced TEAEs of VOD (17.4%).

Reviewer Comment

The draft label includes recommendations for prophylactic therapy against VOD prior to conditioning chemotherapy. VOD will be mentioned in the label, and suggestion for prophylaxis, to ensure optimal care for patients.

6.1.12.3 Deaths

No deaths occurred.

6.1.12.4 Nonfatal Serious Adverse Events

Thirty SAEs in HGB-207 were experienced by 14 subjects. SAEs that occurred in > 1 subject each were pyrexia, thrombocytopenia, and liver veno occlusive disease

(VOD) (3/24; 12.5% each). Subjects experienced SAEs throughout the various stages of the study as follows: time to signing of informed consent form (ICF) to < time of mobilization (M) (2/24, 8.3%), M to < time of conditioning (C) (3/24, 12.5%), C to < time of neutrophil engraftment (NE) (4/23, 17.4%), NE to Month 24 (M24) (11/23, 47.8%), and Day 1 (D1) to M24 (12/23, 52.2%).

Twelve of 23 treated subjects (52.2%) experienced 24 treatment-emergent SAEs, with more than 1 subject experiencing SAEs of VOD (3/23, 13.0%), thrombocytopenia and pyrexia (2/23, 8.7% each) after beti-cel infusion. Of the 24 treatment-emergent SAEs, 15 were attributed to conditioning by the investigator. The reviewer agrees with the attributions of these SAEs, except the one SAE of thrombocytopenia whose description follows:

Subject (b) (6)

5-year-old female experienced a Grade 3 SAE of epistaxis on Day 69, who was evaluated in the emergency department (ED) with intermittent nosebleed lasting 2.5 hours with 3-day history of nocturnal nosebleeds. On exam, the subject was not actively bleeding, appeared pale, and had a blood pressure of 83/55, heart rate of 121 bpm, body temperature of 36.9°C, respiratory rate of 22 and oxygen saturation (SpO2) of 100%. Laboratory results showed hemoglobin of 7.1 g/dl and platelet count of 26×10^9 cells/L. During the ED visit, the subject had 2 episodes of epistaxis that were terminated following less than 20 minutes of pressure. She was hospitalized for ~ 24 hours for the management of epistaxis. The subject received oxymetazoline as well as pRBCs and platelets. On Day 70, the subject's hemoglobin and platelets had improved to 9.7 g/dL and 126×10^9 cells/L, respectively. Her blood pressure was 89/54, heart rate was 93 bpm, body temperature and the event of epistaxis was considered resolved and the subject was discharged from the hospital with her usual prescribed concomitant medications. On Day 80, the subject achieved platelet engraftment. The investigator considered the event related to conditioning therapy with busulfan. No action was taken with beti-cel as a result of the event of epistaxis and the subject continued in the study.

Reviewer Comment

One of the 24 SAEs is considered by this reviewer to be attributable to beti-cel rather than conditioning.

Subject (b) (6) experienced an event of epistaxis due to thrombocytopenia on Day 69 through Day 70. This reviewer considers the underlying SAE to be severe thrombocytopenia, which was complicated by epistaxis that required hospitalization and thus met criteria for Serious Adverse Event. This reviewer attributes this SAE to beti-cel because it occurred at Day 69, that is, in the setting of delayed platelet engraftment (platelet engraftment was reached at Day 80).

All 30 SAEs resolved as of this interim analysis, and none have led to study discontinuation. One SAE was considered possibly related to beti-cel by the

investigator and was reported as a SUSAR: a 22-year-old female (Subject (b) (6)) experienced an SAE of Grade 3 Thrombocytopenia on Day 114, which resolved on Day 163. SAE summary is in the table below:

Table 24 HGB-207 Listing of Serious Adverse Events (ITT) (N=24)

Subject	Preferred Term	AE Start Day	AE End Day	Period AE started	TE
(b) (6)	Hypotension	11	12	C to <NE	Y
	Hypoxia	34	36	NE to M24	Y
	VOD	34	84	NE to M24	Y
	Epistaxis	69	70	NE to M24	Y
	Pyrexia	44	46	NE to M24	Y
	Pneumonia viral	53	54	NE to M24	Y
	Viral infection	198	199	NE to M24	Y
	Bacterial sepsis	149	162	NE to M24	Y
	Pyrexia	-116	-115	ICF to <M	
	Gastroenteritis viral	-153	-149	ICF to <M	
	Transfusion reaction	23	24	NE to M24	Y
	VOD	23	37	NE to M24	Y
	Contusion	505	522	NE to M24	Y
	Vascular device infection	-60	-55	M to <C	
	Pyrexia	37	44	NE to M24	Y
	Femur fracture	676	677	NE to M24	Y
	Neutropenic sepsis	9	10	C to <NE	Y
	Thrombocytopenia	114	163	NE to M24	Y
	Appendicitis	200	204	NE to M24	Y
	Febrile neutropenia	13	17	C to <NE	Y
	Neutropenia	10	23	C to <NE	Y
	Stomatitis	13	16	C to <NE	Y
	Sepsis	13	16	C to <NE	Y
	Thrombocytopenia	12	50	C to <NE	Y

(b) (6)	Atrial fibrillation	345	359	NE to M24	Y
	Hypokalemia	-315	-307	M to <C	
	Lower respiratory tract infection	14	25	C to <NE	Y
	Thrombocytopenia	-314	-312	M to <C	
	VOD	14	54	C to <NE	Y
	Catheter site hemorrhage	-313	-313	M to <C	

Source: Reviewer calculations, ADAE dataset. Abbr. Treatment Emergent = TE, veno occlusive disease = VOD.

Narratives for subjects in the ITT population who experienced an SAE are below. All were reviewed, but we excluded some which were considered uninformative. For example, viral gastroenteritis that occurred 153 days before beti-cel infusion.

Hepatic Veno Occlusive Disease

Three subjects had treatment-emergent SAEs in the Hepatobiliary disorders SOC:

- Subject (b) (6) had a non-serious Grade 3 AE of VOD on Day 26 and a Grade 4 SAE of VOD on Day 34, and in that setting had additional non-serious AEs of hepatomegaly, cholelithiasis, AST increased, and bilirubin increased. He required peritoneal drain placement. VOD was considered resolved on Day 84.
- Subject (b) (6) had a Grade 4 SAE of VOD on Day 23. The SAE was considered resolved on Day 37.
- Subject (b) (6) had a Grade 4 SAE of VOD on Day 14 and in that setting had a non-serious AE of ALT increased. VOD was considered resolved by Day 54.

The VOD events were assessed as related to conditioning with busulfan and resolved after appropriate treatment.

SAEs Attributed to beti-cel.

Below are discussed the two SAEs which the review team ascribed to beti-cel.

- 22-year-old female was reported to have an SAE of thrombocytopenia on Day 114. Platelet engraftment occurred on Day 53, but her platelet counts subsequently remained below 35×10^9 cells/L. The Investigator reported that the subject had increased susceptibility to bruises but no overt bleeding. On Day 163, the subject's platelet count reached 50×10^9 cells/L and the event of thrombocytopenia was considered resolved. The investigator assessed the event of thrombocytopenia as meeting the serious criterion of a life-threatening event and was possibly related to beti-cel, but not related to plerixafor, conditioning with busulfan, or study procedure.

Reviewer Comment

Such protracted, high-grade thrombocytopenia after a delayed platelet engraftment is of concern, and was appropriately attributed to beti-cel, rather than busulfan. The subject had platelet counts in the $20\text{--}35 \times 10^9/\text{L}$ range after Day 53 platelet engraftment until Day 163. According to the Applicant, the event was serious because it was determined a medically important event. Because many clinical factors impact a clinician's judgement of a patient, the reviewer will accept the investigator's determination that based on direct patient evaluation in the clinic on around Day 114, this met the criterion of a serious event.

- The other SAE attributed to beti-cel was an SAE of thrombocytopenia complicated by epistaxis on Day 69, discussed above, in section 6.1.12.4.

6.1.12.5 Adverse Events of Special Interest (AESI)

AESIs for LVV products include insertional oncogenesis, clonal proliferation, and presence of replication competent lentivirus. In addition, earlier clinical evidence suggests that beti-cel is associated with prolonged platelet engraftment. The Applicant proposed events of interest as HIV infection, autoimmune disease, infections, malignancies, bleeding events and graft vs host disease (GVHD). None of these categories of events was reported except one bleeding event caused by thrombocytopenia attributed to beti-cel, which is described in the last paragraph of section 6.1.12.4, immediately before this section. There were other bleeding events which occurred shortly after myeloablation and were expected reactions of busulfan.

Replication Competent Lentivirus (RCL)

Another recognized safety concern for LVV products is the potential for generation of RCL during manufacture or after transplantation, thus blood of all subjects was sampled for RCL at Months 3, 6, 12, and 24; with the more rigorous co-culture assays used to evaluate for any false positives. A qPCR assay for the detection of VSV-G sequences in genomic DNA from treated subjects was used as a screening assay for RCL. RCL has not been detected at any time in any subject as of last follow-up.

Insertional Oncogenesis

Due to their nature, lentiviral vectors integrate into the chromosome of target cells upon transduction and carry a potential risk of insertional oncogenesis. As part of safety evaluation, subjects underwent regular screening. Integration site analysis (ISA) showed polyclonal reconstitution with transduced HSCs for all subjects, based on analysis of DNA from peripheral blood cells. The number of unique mappable integration sites (IS) ranged from 799 to 34857. Insertional sites were most frequently detected in the NF1 (8/22, 36% of subjects), ASH1L (6/22, 27% of subjects) and PACS1 (6/22, 27% of subjects) genes. In individual subjects, the

most frequently represented IS had a relative frequency of 6%. No subject met the criteria for clonal predominance. Please see section 9.2 [Aspect(s) of the Clinical Evaluation Not Previously Covered] where important concerns related to insertional oncogenesis are discussed, in connection to information from and IND for sickle cell disease (SCD) and a BLA for cortical adrenal leukodystrophy (CALD).

Graft vs. Host Disease

There were no reports of Graft vs. Host Disease in HGB-207.

6.1.12.6 Clinical Test Results

Laboratory based AEs were analyzed by the review team using JMP 16 software using shift tables using the laboratory test result analysis (ADLB) dataset, rather than the adverse event analysis (ADAE) dataset, as this is more accurate. Shift tables display the change in the frequency of subjects across specified categories from baseline to post-baseline time points. They depict the shift in the values of laboratory parameters across visits. The table below lists the results.

Table 25 Laboratory Based Abnormalities in $\geq 5\%$ Subjects

Laboratory Based Abnormality	All Grades n (%)	n (%) Gr3 and Gr4
Chemistry		
Serum Alkaline Phosphatase increased	8 (35)	1 (4)
Serum ALT increased	21 (91)	5 (22)
Serum AST increased	17 (74)	1 (4)
Hyperbilirubinemia	8 (35)	4 (17)
Hypocalcemia	20 (87)	1 (4)
Hypercalcemia	7 (30)	1 (4)
Hypoglycemia ^{\$}	1 (5)	0
Hyperglycemia ^{\$}	3 (15)	3 (15)
Hyponatremia	14 (61)	3 (13)
Hypernatremia	4 (17)	0
Hypokalemia	18 (78)	4 (17)
Hyperkalemia	5 (22)	0
Hematology		
Hypophosphatemia	15 (65)	5 (22)
Neutropenia	23 (100)	23 (100)
Thrombocytopenia	23 (100)	23 (100)
Anemia	22 (96)	21 (91)
Erythrocytosis	3 (13)	0
Lymphopenia	22 (96)	16 (70)
Lymphocytosis	3 (13)	0
Leukopenia	23 (100)	23 (100)

Source: Reviewer analysis from adlb.xpt dataset. ^{\$}Denominator is 20, not 23.

Abbreviations: ALT: alanine aminotransferase; AST aspartate aminotransferase

Reviewer Comment

- The ADLB dataset was used to generate incidence of laboratory- based AEs since this is more accurate as opposed to using the adverse event dataset (ADAE dataset).
- A “lab-shift” analysis was carried out wherein baseline laboratory abnormalities that worsened following treatment were recognized i.e., shift of a laboratory result grade from a lower to higher grade.
- During the interval between conditioning and neutrophil engraftment (C to NE period), cytopenias are the expected result of conditioning chemotherapy, after which they tend to resolve as hematopoiesis recovers. Most cytopenia AEs in Study HGB-207 were observed in the peri-transplant (C to <NE period) and improved over time. Some cytopenia AEs were reported in the NE to M24 period (e.g., thrombocytopenia, discussed above). No late bleeding events were documented in association with these cytopenias.
- Hepatic enzyme and bilirubin abnormalities were observed, primarily among the subjects who developed veno-occlusive disease, which is a known complication of busulfan chemotherapy.

Hematology:

Cytopenias:

Recipients of myeloablative conditioning chemotherapy are expected to develop severe cytopenias before reconstitution of hematopoiesis after infusion of HSCs. Many dynamics, including the use of growth factors, can affect time to hematopoietic recovery. Prolonged thrombocytopenia was the most notable cytopenia observed.

Neutrophil engraftment:

Subjects remained hospitalized and underwent daily complete blood count (CBC) monitoring of neutrophil counts. Successful neutrophil engraftment (NE) was defined as 3 consecutive absolute neutrophil count (ANC) laboratory values $\geq 0.5 \times 10^9$ cells/L obtained on different days by Day 43 after a post-transplant value of $< 0.5 \times 10^9$ cells/L, without the need for back-up cells. However, in four of 23 beti-cel treated subjects from HGB-207, the time to neutrophil engraftment was confounded by administration of G-CSF for at least 7 days beyond NE, although all subjects achieved NE before day 42. The median Day of NE was similar between the two cohorts. Neutrophil engraftment results are tabulated below:

Table 26 HGB-207 Neutrophil Engraftment by Cohort

Parameter	Statistic	Cohort 1	Cohort 2	Total
Day of NE (relative day)	N	15	8	23
	Median	24.0	23.0	23.0
	Range	13 - 28	17 - 32	13 - 32

Source: Reviewer calculations from ADSL dataset

Delayed Platelet Engraftment

Reconstitution of the platelet lineage after beti-cel was notably slower than expected and reported in the literature for patients with TDT undergoing AHSCT5. Platelet engraftment (PE) was defined as 3 consecutive platelet values $\geq 20 \times 10^9$ /L obtained on different days after a post-transplant value of $< 20 \times 10^9$ /L, with no platelet transfusions administered for 7 days immediately preceding and during the evaluation period.

Platelet engraftment on \leq Day 30 was observed in 2/23 subjects (8.7%), whereas 21/23 subjects had PE between Days 31 and 94. Data suggest a potential association between beti-cel and delayed PE. As a result, delayed PE has been noted as an important identified risk for beti-cel. Results of platelet engraftment are summarized in the tables below:

Table 27 HGB-207 Day of Platelet Engraftment per Cohort

Parameter	Statistic	Cohort 1	Cohort 2	Total
Day of Platelet Engraftment (relative day)	N	15	8	23
	Median	45	51	46
	Range	32, 84	20, 94	20, 94

Source: Reviewer calculations from ADSL dataset

Platelet engraftment was also evaluated by category based on number of days delayed. A majority of HGB-207 subjects reached PE between Day 3 and $<$ Day 60, but 14% required over 60 days to achieve PE. Please see the table below:

Table 28 HGB-207 Platelet Engraftment Delay Category

Parameter	Statistic	Cohort 1	Cohort 2	Total
\leq Day 30	n (%)	0	2 (25)	2 (9)
Day 30 to \leq Day 60	n (%)	13 (87)	4 (50)	17 (74)
$>$ Day 60 to \leq Day 90	n (%)	2 (13)	1 (13)	3 (13)
$>$ Day 90	n (%)	0	1 (13)	1 (4)

Source: Reviewer calculations from ADSL dataset

Recovery of platelet counts to a sustained level $\geq 50 \times 10^9$ /L and $\geq 100 \times 10^9$ /L HGB-207 subjects treated with beti-cel reached an unsupported platelet count of $\geq 50 \times 10^9$ /L at a median of 58 days (range 20 to 247), and it took them a median of 108.5 days (range 49 to 381) to reach an unsupported platelet count of $\geq 100 \times 10^9$ /L.

55 Ghavamzadeh A, et al. Comparable outcomes of allogeneic peripheral blood versus bone marrow hematopoietic stem cell transplantation in major thalassemia: A multivariate long-term cohort analysis. Biol Blood Marrow Transplant. 2019;25(2):307-12

Reviewer Comment

Compared to allogeneic transplantation for TDT, where patients generally achieve PE around Day 25, beti-cel treatment is associated with delayed platelet engraftment. Splenomegaly might exacerbate this, but TDT subjects in allo-HSCT literature also frequently have splenomegaly, so this cannot adequately explain this observation. Generally, subjects in HGB-207 did not experience excess bleeding events due to the delayed platelet engraftment. However, as discussed previously, one case of grade 3 epistaxis occurred in the context of delayed platelet engraftment on Day 69 (platelet did not engraft until day 80). As described in Section 6.1.12.4, this reviewer attributes the SAE to beti-cel rather than busulfan.

Platelet recovery beyond the point of platelet engraftment, is also slow, for example to thresholds such as $50 \times 10^9/L$ or $100 \times 10^9/L$. Most subjects do not return to their baseline platelet counts during the observation period on the study. This needs additional monitoring, especially since lentiviral transduction during manufacturing is a possible etiology for this delay.

6.1.12.7 Dropouts and/or Discontinuations

One subject discontinued the study upon becoming pregnant after a cycle of mobilization/apheresis and before starting conditioning. The pregnancy led to a live birth at 41 weeks, with a reportedly healthy infant as of 7 weeks of age. The subject was included in the ITT population.

6.1.13 Study Summary and Conclusions

In Study HGB-207, 20 of 22 TI evaluable subjects (90.9%, two-sided 95% CI of 70.8 to 98.9) achieved TI. Among Cohort 1, 14 of 15 TI-evaluable subjects (93.3%, two-sided 95% CI of 68.1 to 99.8) achieved TI at any time, and in Cohort 2, 6/7 TI-evaluable subjects (85.7%, two-sided 95% CI of 42.1 to 99.6) achieved TI at any time. Study HGB-207 safety review confirmed that there were no deaths, 2 beti-cel related SAEs of thrombocytopenia that resolved, and no AEs resulting in discontinuations. Most of the SAEs and AEs were consistent with known effects of myeloablative conditioning. Neutrophil and platelet engraftment have been achieved by all subjects, with no long-term sequelae of serious infection or bleeding events. However, subjects achieved engraftment of platelets at median Day 46, which is notably later than has been reported in allo-HSCT for thalassemia. Subjects reached $50 \times 10^9/L$ platelets at a median of 58 days and required over 108 days to reach and maintain $100 \times 10^9/L$. No evidence of insertional oncogenesis or replication competent lentivirus was found. While the safety profile is largely consistent with the known effects of mobilization/apheresis, myeloablative conditioning, and HSC transplantation, subjects experienced prolonged platelet engraftment.

6.2 Trial #2

Study HGB-212

HGB-212 enrolled only subjects with the most severe TDT phenotype, β^0/β^0 or the functionally equivalent $\beta^0/\text{IVS-I-110}$ and $\text{IVS-I-110}/\text{IVS-I-110}$ genotypes. It excluded candidates with non- β^0/β^0 genotypes who were studied in HGB-207. Otherwise, study HGB-212 is nearly identical in design to Study HGB-207.

6.2.1 Objectives (Primary, Secondary, etc.)

Primary: Evaluate the efficacy of treatment with beti-cel in subjects ≤ 50 years of age with transfusion-dependent β -thalassemia (TDT) who have a β^0/β^0 , $\beta^0/\text{IVS-I-110}$, or $\text{IVS-I-110}/\text{IVS-I-110}$ genotype at the HBB gene

Secondary: Evaluate the safety of treatment with beti-cel in subjects ≤ 50 years of age with TDT who have a β^0/β^0 , $\beta^0/\text{IVS-I-110}$, or $\text{IVS-I-110}/\text{IVS-I-110}$ genotype at the HBB gene.

6.2.2 Design Overview

Interim report of single-arm, Phase 3 study of subjects with TDT who have β^0/β^0 genotype at the hemoglobin beta gene (HBB) found on chromosome 11. This is in contrast to the otherwise similarly designed Phase 3 HGB-207 study in subjects with non- β^0/β^0 genotype. After amendment 5, HGB-212 also permitted subjects with the non- β^0/β^0 genotypes of $\beta^0/\text{IVS-I-110}$, or $\text{IVS-I-110}/\text{IVS-I-110}$ variant at the HBB gene, genotype, because this variant is considered clinically equally severe, with negligible β -globin production. The rationale for designing two parallel phase 3 studies but enrolling subjects differing in genotype is that earlier phase I/II studies suggested that subjects with non β^0/β^0 tended to have better response to beti-cel in terms of the endpoint of transfusion independence.

Reviewer Comment

Transfusion dependent beta thalassemia (TDT) patients can have either the β^0/β^0 genotype, which makes no beta globin, or the non- β^0/β^0 genotype, where some variant beta globin is produced but is insufficient for adequate hemoglobin production without RBC transfusion support. The design of HGB-212 is similar to HGB-207, both studies evaluating efficacy and safety of betibeglogene autotemcel in TDT subjects, but with distinct genotypes. Between Study HGB-207 and HGB-212, beti-cel efficacy and safety will be assessed in these two categories of genotypes. A single study with two cohorts comprised of the β^0/β^0 vs. non β^0/β^0 genotypes, may have been more efficient.

6.2.3 Population

HGB-212 included only candidates with genotypes of β^0/β^0 , $\beta^0/\text{IVS-I-110}$, or $\text{IVS-I-110}/\text{IVS-I-110}$. Otherwise, the populations had identical baseline requirements, including transfusion dependence requirements, as Study HGB-207. Please refer to section 6.1.3 for details.

6.2.4 Study Treatments or Agents Mandated by the Protocol

HGB-212 administered the same investigational beti-cel as Study HGB-207. Please refer to Section 6.1.4 for details.

6.2.5 Directions for Use

HGB-212 beti-cel was administered the same as in Study HGB-207. Please refer to Section 6.1.5 for details.

6.2.6 Sites and Centers

HGB-212 was carried out at 9 study centers in France, Germany, Greece, Italy, UK, and USA.

6.2.7 Surveillance/Monitoring

The applicant ensured appropriate monitoring procedures were performed before, during and after the study. Additionally, HGB-212 had an independent Data Monitoring Committee (DMC) focusing on the safety of subjects. Study candidates underwent a battery of screening procedures within 90 days of mobilization. This included detailed review of records from the prior 2 years on transfusions and hospitalizations. In the pre-conditioning phase of 30 days prior to busulfan administration, subjects were hypertransfused to a hemoglobin level of $\geq 11\text{g/dL}$. Conditioning with busulfan took place between Day -6 to -3, after which subjects had beti-cel infusion on Day 1 and remained in the hospital post infusion until NE. HGB-212 followed the same outline of scheduled activities as Study HGB-207. Please refer to section 6.1.7 for details. After the Month 24 Visit, consenting subjects were enrolled in long-term follow-up Study LTF-303, to be followed for up to an additional 13 years. It included an independent Data Monitoring Committee (DMC).

6.2.8 Endpoints and Criteria for Study Success

The primary efficacy endpoint, as well as secondary, and explorative endpoints were the same in HGB-212 as in HGB-207. Additionally, safety and pharmacodynamic endpoints were identical. Please see section 6.1.8 for details.

6.2.9 Statistical Considerations & Statistical Analysis Plan

Statistical analysis plan (SAP) Version 3.0 (dated 18 December 2020) was used to analyze Study HGB-212 data.

6.2.10 Study Population and Disposition

Nineteen subjects were mobilized (ITT population), 1 subject discontinued after 1 cycle of mobilization, and 18 subjects were conditioned with busulfan and administered beti-cel (EAP). All EAP subjects achieved successful neutrophil engraftment (SEP). Sixteen subjects completed at least the Month 6 visit, 15

subjects completed the Month 12 Visit, and 11 subjects completed the Month 24 visit.

6.2.10.1 Populations Enrolled/Analyzed

As in Study HGB-207, Study HGB-212 was comprised of:

- Intent-to-Treat (ITT) population: All subjects who initiated any study procedures, beginning with mobilization.
- Transplant Population (Efficacy Analysis population): All subjects who received beti-cel
- Successful Engraftment Population (SEP): All subjects who have successfully reached NE after beti-cel infusion

The ITT population was the primary population for the analysis of safety parameters. The EAP was the primary population for efficacy, pharmacodynamic, and transplant-related parameter endpoints. Selected safety analyses were also performed on the EAP. The SEP was to be used to provide supportive data for subjects who engrafted; however, the EAP and SEP were the same for this interim CSR, thus additional analysis for the SEP was not performed.

As the statistical analysis plan was very similar for Study HGB-207, please refer to Section 6.1.8 for details. The main difference was that subjects in HGB-212 were not split prospectively into two cohorts by age like in HGB-207. Thus, the entire subject population's primary efficacy endpoint of TI was analyzed as a point-estimate of the proportion of subjects achieving TI at any time during the study, with a 2-sided 95% CI calculated using the Clopper-Pearson exact binomial method. The success criterion is proposed as a point-estimate of 55.6% (10 out of 18 subjects), which would yield a lower 1-sided 97.5% exact confidence bound of 30.8%, exceeding the 30% minimal criterion. Secondary efficacy endpoints were to be descriptively analyzed in summary tables, where data were available. All efficacy information is presented in data listings

6.2.10.1.1 Demographics

Of 19 subjects in the ITT population, 5 were ≥ 18 years of age (26%), 6 subjects were ≥ 12 to < 18 years of age (32%), and 8 subjects were < 12 years of age (42%) at time of consent/assent. Study HGB 212 complied with the protocol stipulation to enroll at least 10 subjects < 18 years old and ≥ 12 subjects of $\beta 0/\beta 0$ genotype. For their safety, subjects < 12 years old could only enroll after review of initial safety in older subjects. Subjects self-identified as either White (10/19, 52.6%) or Asian (8/19, 42.1%), which is consistent with the epidemiology of TDT; race was not provided for 1 subject. In the ITT population 10 subjects were male (52.6%) and 9 subjects were female (47.4%). Please see table below:

Table 29 HGB-212 Demographics: Age

Parameter	Statistic	$\beta 0/\beta 0$ N=13	Non $\beta 0/\beta 0$ N=6	Total N=19
Age	Median (range)	12 (4, 26)	19 (4,33)	12 (4,33)
≥ 18 years	n(%)	2 (15.4)	3 (50)	5 (26.3)
≥ 12 to <18 years	n(%)	5 (38.5)	1 (16.7)	6 (31.6)
<12 years	n(%)	6 (46.2)	2 (33.3)	8 (42.1)

HGB-212 Sex

Sex	Statistic	$\beta 0/\beta 0$ N=13	Non $\beta 0/\beta 0$ N=6	Total N=19)
Male	n(%)	6 (46.2)	4 (66.7)	10 (52.6)
Female	n(%)	7 (53.8)	2 (33.3)	9 (47.4)

HGB-212 Race/Ethnicity

Race	Statistic	$\beta 0/\beta 0$ N=13	Non $\beta 0/\beta 0$ N=6	Total N=19
Asian	n(%)	8 (61.5)	0	8 (42.1)
White	n(%)	4 (30.8)	6 (100)	10 (52.6)
Not Provided	n(%)	1 (7.7)	0	1 (5.3)

Source: reviewer calculated from ADSL dataset.

Reviewer Comment

The enrolled population ages and genotypes were as stipulated in the protocol. The evaluated subgroups are balanced, although the $\beta 0/\beta 0$ population was younger than the non $\beta 0/\beta 0$. Non $\beta 0/\beta 0$ subjects were all White, due to predominance of this genotype in the Mediterranean region. Subjects were well balanced by sex.

6.2.10.1.2 Medical/Behavioral Characterization of the Enrolled Population

Pathophysiologic features of TDT were reflected in the medical characteristics noted in the studied population. Baseline characteristics included: genotype, $\beta 0/\beta 0$ vs. non- $\beta 0/\beta 0$, spleen size, history of possible splenectomy, age at diagnosis, transfusion onset and duration, use of chelators, baseline transfusion requirements over the 2 years prior to enrollment, and liver iron assessment per MRI at screening. Baseline characteristics are depicted in the table below.

Table 30 HGB-212 Baseline TDT related Medical characteristics

Parameter	Statistic	$\beta 0/\beta 0$ N=13	Non $\beta 0/\beta 0$ N=6	Total N=19
pRBC Transfusion Volume (mL/kg/year)	Median (range)	202.6 (74.6, 289)	185.2 (170.7, 276.1)	198.5 (74.6, 289)
Number of pRBC Transfusions/Year	Median (range)	16.5 (11, 21)	20.3 (12, 40)	17 (11, 40)
Weighted Average Nadir Hb before Transfusion (g/dL)	Median (range)	9.5 (8.2, 11)	9.559 (8.8, 11)	9.521 (8.2, 11)
Screening liver Iron Concentration per MRI (mg/g)	Median (range)	3.8 (1.7, 13.2)	4.5 (1.2, 10.4)	3.8 (1.2, 13.2)
Spleen size (cm ³)	Median (range)	217 (102, 893)	401 (120, 774)	242.7 (102, 893)
Post- splenectomy	n(%)	2 (15)	1 (17)	3 (16)
Intact spleen	n(%)	11 (8)	5 (83)	16 (84)
Age at diagnosis (Months)	Median (range)	6 (0, 72)	7 (0, 19)	6 (0.7)
Transfusion start age (Months)	Median (range)	11 (0, 132)	8 (6, 12)	8 (0, 132)
Chelation start age (Years)	Median (range)	3.5 (0.7, 11)	2.75 (2, 5)	3 (0.7, 11)
Stable RBC regimen age (Months)	Median (range)	42 (2, 264)	12 (6, 48)	36 (2, 264)

Source: Reviewer calculation from ADSL, ADEF1, and ADEF2 datasets.

Reviewer Comment

$\beta 0/\beta 0$ and non- $\beta 0/\beta 0$ genotype status did not differ substantially in baseline clinical parameters, both being diagnosed approximately at six months of age. Transfusion needs continued to be similar, the small number of non $\beta 0/\beta 0$ subjects (n=6) makes inferences difficult. The weighted nadir (pre transfusion) hemoglobin level of both groups was about 9.5 g/dL.

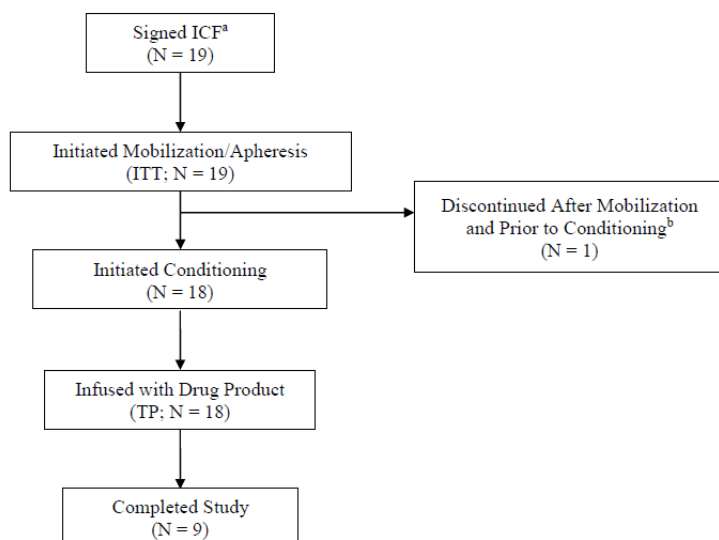
Both groups had significant transfusion requirements, clearly consistent with a severe phenotype. All subjects required at least 100ml/kg/year of pRBCs at baseline, although one 15-year-old subject received 74 ml/kg/year. (She received 11 transfusions per year, over the ≥ 8 transfusions needed per protocol to meet eligibility in those age ≥ 12). Differences in baseline transfusion parameters

between the two genotypes tended to overlap, and variance between the parameters might be due to the small population size as well as variable amount of co-expression of other stable hemoglobins, such as hemoglobin-F HbF). For example, the 15-year-old subject (b) (6) who had < 100ml/kg/year of pRBC at Baseline produced more fetal hemoglobin than any of the other subjects and continued to have the highest HgF level after beti-cel infusion. Non $\beta 0/\beta 0$ subjects tended to have larger spleens (median volume 401 vs. 217 cm³), however this could be an artifact of small sample size and distribution of splenectomy between these subgroups.

6.2.10.1.3 Subject Disposition

A total of 19 subjects signed the informed consent form, of which 18 received beti-cel, and 9 completed the study (9 remaining on study, as it is ongoing). The figure below outlines the disposition:

Figure 2 Disposition



^a As of this interim CSR, 19 subjects have signed informed consent/assent, of which 1 subject (Subject (b) (6)) was initially a screen failure who later re-screened and enrolled with a new subject number (Subject (b) (6)). On 23 January 2018, Subject (b) (6) failed screening because they met the exclusion criteria for advanced liver disease (defined in [Appendix 16.1.1 Protocol HGB-212 Version 3.0](#)), which may be met if liver iron concentration (LIC) is measured at ≥ 15 mg/g by MRI at screening. After receiving iron chelation therapy ([Listing 16.2.6.11](#)), the subject had an LIC of 10.8 mg/g during re-screening ([Listing 16.2.6.8.2](#)) and enrolled with a new subject number (b) (6) on 20 August 2018 ([Listing 16.2.1.1](#)).

Source: Study HGB-212 Study Report Page 96

Subject (b) (6) was initially rejected as a screen failure due to advanced liver disease with elevated liver iron concentration on MRI. However, the subject then received iron chelation and achieved passing iron concentration, and met enrollment criteria, but was enrolled with new subject number: (b) (6). This subject eventually withdrew consent after mobilization and apheresis, leaving N=18 subjects in per protocol set.

Protocol deviations:

Due to COVID-19 restrictions, between 1-4 visits were conducted over the phone and no assessments were done for Subjects (b) (6)

Additional deviations are listed below:

- Subject (b) (6) had the second lot of beti-cel not administered immediately following the first lot as per protocol. There was an approximate 7-hour time gap between infusions.
- Subject (b) (6) had beti-cel Lot 19HT703045 did not meet specifications due to VCN result of 7.0 (specification 0.8 to 6.6) but was administered based on medical need per the Investigator and Medical Monitor, after receiving FDA approval for infusion of the beti-cel, IRB acknowledged being notified. The subject's bone marrow samples were shipped to the central lab and the Sponsor with the subject's name on the labels in a breach of protected health information. A subject status update email inadvertently included the subject's first name in the email in a breach of protected health information.
- Subject (b) (6) did not use contraception during the screening.
- Subject (b) (6) G-CSF was used on Day 18 post-infusion (prior to Day 21) due to a misunderstanding.
- Subject (b) (6) The subject experienced an SAE of Device related infection during preconditioning, Febrile neutropenia, Stomatitis, and Neutropenia that was not reported within 24 hours of awareness. Laboratory assessments for hematology were not done on the first day nor the third day of conditioning. Subject's vital signs were not monitored at 2 hours post-infusion.
- Subject (b) (6) The subject's name was provided in an email correspondence to Sponsor in a breach of protected health information.
- Subject (b) (6) used only one method of contraception during the screening period. The subject's differential blood count was not assessed on 9 scheduled days due to weekend staffing or insufficient blood volume collected.
- Subject (b) (6) The subject's differential blood count was not assessed on 11 days due to weekend staffing or insufficient blood volume collected.

Reviewer Comment

Most of these protocol deviations involve laboratory testing which were missed due to error, or potentially breached subject confidentiality. One involved manufacturing of product with VCN slightly over specification, reported to IRB and regulatory agencies. Overall, the study data are likely not substantially impacted. The prevalence of deviations does not appear excessive.

6.2.11 Efficacy Analyses

As this study is ongoing, the analysis is based on an interim report with last subject visit date of 09 March 2021, and interim data lock date of 13 April 2021.

6.2.11.1 Analyses of Primary Endpoint(s)

The primary efficacy endpoint is the proportion of subjects who meet the definition of TI, defined as a weighted average Hb \geq 9g/dL without any pRBC transfusions for a continuous period of \geq 12 months at any time during the study after beti-cel infusion. The weighted average hemoglobin was pegged at 9 gm/dL, which was to satisfy contemporary guidelines for the hemoglobin target normally achieved with red cell transfusion support. Maintenance of a stable hemoglobin at 9 gm/dL or greater should permit acceptable quality of life while potentially allowing for phlebotomy to treat iron overload that most participants bear as a result of transfusion dependence. The starting point of transfusion independence (T0) occurs once a subject achieves a Hb level of \geq 9 gm/dL at least 60 days post last RBC transfusion. The 60-day lag is necessary to ensure senescence of transfused cells. Of the 18 subjects infused with beti-cel, 14 are TI evaluable. (Defined as having achieved TI, completed 24 month follow up in the study, or have demonstrated that they will not achieve TI). Among these 14 evaluable subjects, 12 (85.7%) have achieved TI, 87.5% of β 0/ β 0 and 83.3% of non β 0/ β 0. Efficacy outcomes are shown in the table below:

Table 31 HGB-212 Primary Efficacy Endpoint (TI) per Genotype Cohort

Parameter	Statistic	β 0/ β 0 N	Non β 0/ β 0 N	Total N
TI evaluable	N	8	6	14
Subjects with TI at any time	n (%) 2-sided 95% CI	7 (88) 47.3, 99.7	5 (83) 35.9, 99.6	12 (86) 57.2, 98.2

Source: reviewer calculation from ADEF2 dataset

Two subjects in the TI evaluable population failed to achieve TI, Subject (b) (6) who was β 0/ β 0 and Subject (b) (6) who was non β 0/ β 0. These two subjects had among the lowest amount of expression of Hb^{A787Q} of all TI evaluable subjects, 0 and 5.1g/dL. In addition, these subjects had among the lowest peripheral blood vector copy number (PB VCN) values of all TI evaluable subjects. (0.179 and 0.393 c/dg at <month 6).

Reviewer Comment

The rate of TI achieved on the study was comparable between subjects with β 0/ β 0 and non β 0/ β 0 genotype. The subjects who failed to achieve TI tended to have some of the lowest HbA787Q and PB-VCN levels at six months.

6.2.11.2 Analyses of Secondary Endpoints

Secondary endpoints focused on characterization of subjects achieving TI. They included the proportion of subjects who meet the definition of TI at the Month 24 visit; duration of TI; time from beti-cel infusion to achievement of TI; and weighted average Hb during TI.

Table 32 HGB-212 Characterization of TI among subjects who achieve TI

Parameter	Statistic	$\beta 0/\beta 0$ N=7	Non $\beta 0/\beta 0$ N=5	Total N=12
Subjects with TI at month 24	N (%) 2 sided 95% CI	6 (86) 42.1, 99.6	3 (75) 19.4, 99.4	9 (82) 48.2, 97.7
TI Duration (months)	Median (range)	20.8 (13.1, 21.7)	18.8 (12.5, 20.5)	19.5 (12.5, 21.7)
Weighted Hgb avg. during TI	Median (range)	10.2 (9.4, 13.5)	10.3 (10, 13)	10.2 (9.4, 13.5)
Time from beti-cel to last pRBC before TI (months)	Median (range)	0.8 (0, 1.5)	1.3 (0.5, 1.9)	0.8 (0, 1.9)
Time from beti-cel to first TI status (months)	Median (range)	15.7 (14.8, 24.5)	15.7 (15.3, 16.1)	15.7 (14.8, 24.5)

Source: review calculations from ADEF2, ADTTE data sets.

Reviewer Comment

Subjects achieving TI appeared to maintain this outcome, and the average nadir Hb level was at least as high or even above that at baseline. Non- $\beta 0/\beta 0$ and $\beta 0/\beta 0$ subgroups had similar outcomes on most secondary TI endpoints, though of the Non $\beta 0/\beta 0$ group, fewer were in TI at Month 24, however, the numbers (N=3) likely are too small to make inferences.

Characterization of transfusion reduction

Other secondary efficacy endpoints evaluated the degree of reduction in transfusion needs. Transfusion reduction was defined as the proportion of subjects a $\geq 60\%$ reduction in the annualized volume of pRBC transfused (in mL/kg) in the post-treatment time period from 12 months post-beti-cel infusion through Month 24 compared to the annualized transfusion requirement during the 2 years prior to study enrollment. The study also looked at the proportion of subjects with a reduction in the annualized mL/kg pRBCs transfused from 12 months post-beti-cel infusion through Month 24 of $\geq 50\%$, 60% , 75% , 90% , or 100% compared to the annualized number and volume of pRBC transfusion requirements during the 2 years prior to enrollment; annualized number and volume of pRBC transfusions from 12 months post-beti-cel infusion through Month 24 compared to annualized number and volume of transfusions during the 2 years prior to enrollment; time from beti-cel infusion to last pRBC transfusion; and time from last pRBC

transfusion to the Month 24 Visit). Results of the 13 subjects with at least 18 months of post beti-cel follow up are listed in the following table:

Table 33 HGB-212 Characterization of Transfusion Reduction

Parameter	Statistic	$\beta 0/\beta 0$ (N=8)	non $\beta 0/\beta 0$ (N=5)	Total (N=13)
<50%	n (%) 2 sided 95% CI	1 (12.5) 0.3, 52.7	0 0, 52.2	1 (7.7) 0.2, 36
$\geq 50\%$	n (%) 2 sided 95% CI	7 (87.5) 47.3, 99.7	5 (100) 47.8, 100	12 (92.3) 64.0, 99.8
$\geq 60\%$	n (%) 2 sided 95% CI	7 (87.5) 47.3, 99.7	5 (100) 47.8, 100	12 (92.3) 64.0, 99.8
$\geq 75\%$	n (%) 2 sided 95% CI	7 (87.5) 47.3, 99.7	5 (100) 47.8, 100	12 (92.3) 64.0, 99.8
$\geq 90\%$	n (%) 2 sided 95% CI	7 (87.5) 47.3, 99.7	5 (100) 47.8, 100	12 (92.3) 64.0, 99.8
100%	n (%) 2 sided 95% CI	7 (87.5) 47.3, 99.7	4 (80) 28.4, 99.5	11 (84.6) 54.6, 98.1

Source: reviewer calculated from ADEF1 dataset

Reviewer Comment

Transfusion independence, i.e., 100% reduction in need of transfusions, was achieved by 11 of 13 treated subjects, but the study protocol also included secondary endpoints revolving on lesser, prespecified degrees of transfusion reduction. From among the pair of subjects who did not achieve TI, one had reduction of < 50% in transfusion needs (15%) remaining dependent on regular transfusions, and the other had a 92% reduction in transfusion requirements while maintaining an average weighted Hgb of 8.0g/dL. These two subjects had the second and third lowest PB VCN values of all TI-evaluable subjects and had among the lowest amounts of HbAT87Q at Month 6.

Weighted Average Nadir Hemoglobin Level

Weighted average nadir Hb during the 2 years prior to enrollment was compared to weighted average nadir Hb from 12 months post-beti-cel infusion through the Month 24 Visit, to give a picture of the degree of anemia the subjects experienced before vs. after therapy. Note that N = 15 are the proportion of subjects from among the N=18 that were beti-cel -treated and had ≥ 12 months of f/u (since the

parameter of interest was the weighted average nadir Hb *over the period from 12 months post DP to last follow up*). Of these 15 subjects, 12 achieved TI). The remaining three subjects evaluable for this analysis from 12 months post-drug product infusion through last follow-up, have not achieved TI at time of data lock: Subject (b) (6) who was not yet TI evaluable for efficacy, and two Subjects (b) (6) who did not achieve TI.

Table 34 HGB-212 Weighted Average Nadir Hemoglobin Levels Compared with Baseline

Parameter	Statistic	Baseline	12 Months post-infusion through Last Follow-up	% change
Total (N=15)	Median g/dL (range)	9.5 (8.2, 10.7)	10 (8.05, 13.6)	8.7 (-17.4, 54.5)
β0/β0 (N=9)	Median g/dL (range)	9.5 (8.2, 10)	10 (9.2, 13.5)	5.8 (-2.3, 39.2)
Non β0/β0 (N=6)	Median g/dL (range)	9.6 (8.8, 10.7)	10.2 (8.05, 13.6)	9.5 (-17.4, 54.5)

Source: reviewer calculations from ADEFF2 data set

Reviewer Comment

This analysis shows that subjects who achieved TI tend to at least maintain the same, or slightly higher hemoglobins levels post beti-cel.

Unsupported total Hb levels over time

This analysis evaluated the hemoglobin levels in subjects achieving TI, without further transfusions. Please see results in table below.

Table 35 Unsupported total Hb levels over time, including Month 6, 9, 12, 18, and 24.

Parameter	Statistic	Month 3	Month 6	Month 9	Month 12	Month 18	Month 24
Un supported Hgb (g/dL)	N Median (range)	13 9.8 (6.2,14.3)	15 10.2 (8.5,13.2)	14 10.7 (7.6, 13.1)	14 10.1 (7.9,14)	11 10.4 (8.5, 13.8)	10 10.6 (7.9, 14)

Source: reviewer calculation ADLB data set

Iron Chelation

Due to regular transfusions, TDT subjects generally need to accept the ongoing burden, expense and potential toxicities from chelator therapy. Of the 18 subjects receiving beti-cel, 12 (75%) stopped their iron chelation after beti-cel infusion, and

the median time for discontinuation of chelation was 7.41 months. This is summarized below:

Table 36 HGB-212 Characterization of Chelation therapy post beti-cel Infusion

Parameter	Statistic	$\beta 0/\beta 0$ N=12	Non- $\beta 0/\beta 0$ N=6	Total N=18
Stopped chelation post beti-cel	n (%)	8 (80%)	4 (67%)	12 (75%)
Time to stopping chelation	N Median (range)	8 7.41 (0.3, 25.5)	4 10.5 (3.9, 22.9)	12 7.41 (0.3, 25.5)
Number remaining off chelation ≥ 6 months post beti-cel	n (%)	7 (70)	3 (50)	10 (62.5)
Time from end chelation to last follow up	N Median (range)	7 8.2 (6.2, 25.5)	3 15 (6, 22.9)	10 11.7 (6, 25.5)

Source: reviewer calculations ADSL data set

Reviewer Comment

Iron overload management was at the discretion of investigators and not protocol specified. This analysis suggests that most subjects were able to stop ongoing chelation therapy.

Assessment of Liver in Vital Organs, Liver, Heart

Liver Iron Content

Liver iron content was measured at baseline and followed at 12 and 24-month visit post beti-cel infusion. Immediately before treatment on study, most subject underwent hypertransfusion and discontinued iron chelation to optimize their stem cell collection. These interventions might have contributed to progression in iron content in the liver, and median liver iron content rose at 12 months, and while it then trended downward at month 24, it remained above baseline levels in all subjects, including those who were able to stop all transfusion (TI population). All subgroups examined except $\beta 0/\beta 0$ subjects who achieved TI, ended up with greater liver iron content at month 24 than at baseline.

Cardiac Iron Burden

The cardiovascular magnetic resonance relaxation parameter $R2^*$ (assessed clinically via its reciprocal $T2^*$) measured in the ventricular septum is used to assess cardiac iron. According to the sponsor: "normal $T2^*$ mean of 40 ms has

been widely used, which equates to 0.50 mg/g of dry weight. Like liver iron content, iron content in the heart was measured at baseline, month 12 and 24. And as in HGB-207, subjects enrolled in HGB-212 generally underwent hyper transfusion and interrupted chelation to optimize HSPC mobilization in anticipation of treatment. The T2 analysis showed decrease in all subjects and subgroups such as genotypes and those achieving TI status (N.B.: indicating rising iron content). Lastly, serum ferritin at baseline, Month 12 and Month 24 Visits was assessed to further observe iron stores. This analysis suggested a modest rise between Baseline and 12-month visit, followed by a decrease. Nevertheless, cardiac iron load was higher even at Month 24 than it had been at Baseline.

Table 37 HGB-212 Ferritin Levels Post Beti-cel Infusion

Parameter	Statistic	Baseline	12 month	24 month	% Change from baseline
Total	N Median (range)	18 3275 1279,8874	14 3878.67 496.6, 6907.9	10 1393 606.7,6780	10 -5 -75.8,76.4)
TI subjects	N Median (range)	12 3275 1279,4831	11 2757.3 497,5894	8 1337 607, 6692	8 -37.7 (-75.8,38.5)
Non-TI subjects	N Median (range)	2 4240.5 3672,4809	2 5564.1 5503.4,5624.7	2 6629.2 6479,6780	2 58.7 (41, 76.4)
Not TI evaluable subjects	N Median (range)	4 2964 2629,8874	1 6907.9	0	0

Source: Reviewer calculation ADLB dataset

Reviewer Comment

LIC and cardiac iron content tended to increase appreciably between Baseline and Month 12, subsequently, there is a trend of decreasing LIC and cardiac iron burden. Most subjects had increased iron overload parameters compared with baseline, even those achieving TI. (The exception were the subgroup with $\beta 0/\beta 0$ genotype, who achieved TI, as they had a slight decrease in LIC at Month 24). There is an expectation that after effective treatment for TDT that abolishes need for RBC transfusion, eventually iron overload will improve or resolve, however, this study clear suggests that such process requires longer than 24 months. However, this time-course corresponds to publications from the allo-HSCT literature, where reports assessed for (and demonstrated) improvement in iron overload between two and seven years after transplant⁶.

⁶ Angelucci E, Muretto P, Lucarelli G, Ripalti M, Baronciani D, Erer B, Galimberti M, Giardini C, Gaziev D, Polchi P. Phlebotomy to reduce iron overload in patients cured of thalassemia by bone

Ferritin has limitations as a marker of iron overload, e.g., it is an acute phase reactant and might increase with the autologous transplant required with beti-cel therapy, thus while it is a less robust marker of iron burden than imaging studies, ferritin may reflect changes more rapidly than the imaging parameters. Thus, subjects' ferritin levels peaked at Month 12 and they generally decreased by Month 24 to less than baseline overall, especially among the responders (TI achieving subjects with a decrease of 37.7% at Month 24).

Health related quality of life

HGB-212 included evaluation of the health-related quality of life (HRQoL), however the amount of HRQoL data was small, for example, (b)(4) tool data were provided at Month 12 only for 5 subjects, and Month 24 outcomes are only available for four. The (b)(4) tool results were provided at Month 24 for seven subjects. These limited amounts of data make drawing of conclusions difficult. In HGB-212, HRQoL was assessed over time using the following validated tools: for pediatrics: (b)(4) parent general core and general core); for adolescents: (b)(4) (parent general core and general core) and (b)(4) (Youth version; (b)(4) ; and for adults: (b)(4)

In general, change from baseline for each subject was variable without a consistent trend across subjects. For the tool called (b)(4) , which looks at subjects' scores as well as their parents' scores (which reflect how parents view their children's QOL), the subjects' scores suggest slightly improved outcomes at Month 24, but the parent derived scores decreased. (i.e., parents of the subjects felt their children had deterioration of quality of life.)

Reviewer Comment

Small numbers of subjects with evaluable QOL data and modest changes make drawing inferences from these outcomes challenging, especially given the limitations of QOL assessments in uncontrolled, open-label trials. As with time-to-event endpoints, interpretation of patient-reported outcomes is challenging in uncontrolled clinical trials, because it is unclear to what extent the outcomes can be attributed to the treatment effect of the regimen vs. to underlying disease and patient characteristics. Furthermore, the Applicant did not seek a labeling claim based on QOL data and these data were not incorporated in the PI. Finally, one of the tools used was generally designed and intended to be used in patients with advanced cancers, and its validity in patients with hemoglobinopathy remains to be demonstrated.

6.2.11.3 Subpopulation Analyses

Age

There were no elderly subjects, as age over 50 was exclusionary. Fourteen of 18 subjects infused with the beti-cel were evaluable for TI, and 12 of those subjects had TI. Four subjects < 12 years of age (80%) achieved TI and had a median (min, max) duration of TI of 18.12 (12.5, 21.2) months. Four subjects ≥ 12 to < 18 years of age (100%) achieved TI with a median (min, max) duration of 20.16 (19.4, 21.7) months and 4 subjects ≥ 18 years (80%) of age reached TI with a median (min, max) duration of TI of 18.99 (13.1, 21.2) months.

Sex

Among the 14 TI evaluable subjects, 6 were female and 8 were male. Seven of 8 (87.5%) evaluable males achieved TI and 5 of the 6 (83.3%) evaluable females achieved TI.

Race

Of 14 evaluable subjects, 10 were white, and 3 Asian, with one with unreported race. Eight of the 10 (80%) White TI evaluable subjects achieved TI. Three of the 3 (100%) Asian TI evaluable subjects reached TI, and the 1 (100%) unreported race TI evaluable subject achieved TI.

Reviewer Comment

The numbers are small, and results might reflect chance. Age analysis suggests that adolescents had the highest rate of achieving TI (100%). Slightly greater fraction of males achieved TI than females. All racial groups tended to have high success rate at reaching efficacy endpoint of TI. Given the small number of subjects, it is difficult to draw any specific conclusions.

6.2.11.4 Dropouts and/or Discontinuations

Only one subject dropped out (before conditioning and beti-cel infusion) and was included appropriately in the safety population.

Reviewer Comment

The most up to date SAP procedures manage withdrawals, dropouts, those subjects lost to follow-up, and COVID-19 pandemic modifications. These procedures appear reasonable.

6.2.11.5 Exploratory and Post Hoc Analyses

None.

6.2.12 Safety Analyses

6.2.12.1 Methods

Key materials used for safety review included:

- The BLA application electronic submission
- Applicant submissions in response to the review team's information requests
- Proposed labeling for beti-cel
- Published literature
- Prior regulatory history

The clinical review of safety was primarily based upon analysis of 19 subjects in Study HGB-212 at the primary data cutoff of 09 March 2021. Analysis datasets (ADaM datasets) were used for the safety analysis. Analyses by the clinical reviewer for safety were performed using JMP 16. All narratives and relevant CRFs were reviewed for all SAEs that occurred in the primary safety population. Adverse events (AEs) were coded using the MedDRA version 23.0, and AE severity was graded using the NCI's CTCAE version 4.03. Concomitant medications were coded using the WHO Drug Dictionary March 2016 or later. Some AEs are presented throughout this review as grouped terms as defined by the review team. The complete list of FDA's grouped terms is presented in APPENDIX. Unless otherwise specified, analyses and tables were generated by FDA clinical reviewer.

The safety analysis set included all subjects treated with any dose of beti-cel. All AEs were collected from the signing of the informed consent form (ICF) until 24 months after beti-cel infusion. SAEs were any AEs that met at least one of the following criteria: fatal, life threatening, required inpatient hospitalization or prolongation of existing hospitalization, resulted in persistent or significant disability, resulted in congenital anomaly or birth defect, or resulted in any other medically important serious event. SAEs were collected from the time of screening. Treatment-emergent adverse events (TEAEs) were defined as all AEs occurring after beti-cel administration through month 24 after beti-cel infusion.

Reviewer Comment

The Applicant's methodology for determining ADRs differs from the one the reviewer team used. The Applicant tended to define ADRs based on assessed by the investigator/ Applicant as to relatedness to beti-cel, based on pharmacology of the beti-cel, temporality, and the consistency of the pattern of symptoms across studies. Such attribution can be subjective. Furthermore, beti-cel therapy is preceded by necessary myeloablative conditioning chemotherapy; therefore, it is often difficult to parse out the causality of AEs. For these reasons and in order to decrease bias in this uncontrolled study, the reviewer considered any AE that occurred after initiating beti-cel treatment as an ADR. Otherwise, the safety reporting methods appear acceptable.

Exposure

The ITT population (N = 19) was the primary population for the analysis of safety. This population included subjects who initiated any study procedure, beginning with mobilization with G-CSF and plerixafor. Plerixafor dose on average was 0.23 to 0.25 mg/kg/day, while G-CSF was dosed at 5 to 5.8 micrograms/Kg/day for splenectomized subjects and 9.3 to 11.8 micrograms /Kg/day for those with intact spleens. G-CSF dose was adjusted for spleen presence.

Eighteen of 19 subjects underwent mobilization/apheresis, conditioning, and treatment with beti-cel (EAP). Subject (b) (6) discontinued after a mobilization/apheresis cycle, before conditioning and beti-cel infusion due to withdrawal of consent. Mobilization took 5-7 days. Fifteen of 19 subjects (78.9%) had one cycle of mobilization, while 3 subjects (15.8%) required 2 cycles, and a single subject had 3 cycles. Busulfan conditioning chemotherapy was administered to 18 subjects with 7 receiving 4 days of daily dosing and 11 receiving every 6 hours for 4 days schedule.

Per protocol, beti-cel dose was $\geq 5.0 \times 10^6$ CD34+ cells/kg, and all subjects received at least this minimum target dose. Multiple drug product lots were required for some subjects to meet dose. Of 18 treated subjects, 14 had single lot to manufacture the dose, and 4 subjects had two lots. Median dose of beti-cel was 10.8×10^6 /Kg CD34+ cells, range 5.9 to 42.1.

Review comment

Subjects received adequate doses of conditioning chemotherapy with busulfan, and all received at least the minimal dose of beti-cel specified by protocol. One subject was an outlier and received more than twice the dose compared to the next highest dose recipient at 42.1×10^6 CD34+ cells/kg.

6.2.12.2 Overview of Adverse Events

Common AEs in the ITT population from time of mobilization <conditioning in 10% or more of subjects were self-resolving and generally Grade 1 and 2. There was one procedural pain AE of Gr 3 and one pain AE of Gr 3. They are listed by SOC below.

Table 38 AEs in the ITT population from time of mobilization to before start of conditioning in 10% or more of subjects

System Organ Class	Preferred Term	All Grades (%)	Grade 3 and 4 (%)
Gastrointestinal disorders	Nausea	2 (10.5)	0 (0)
General disorders and administration site conditions	Catheter site pain	3 (15.8)	0 (0)
Infections and infestations	Nasopharyngitis	2 (10.5)	0 (0)
Injury, poisoning and procedural complications	Procedural pain	6 (31.6)	1 (5.3)
Metabolism and nutrition disorders	Hypocalcemia	7 (36.8)	1 (5.3)
Musculoskeletal and connective tissue disorders	Bone pain	5 (26.3)	0 (0)
Nervous system disorders	Headache	5 (26.3)	0 (0)
Psychiatric disorders	Anxiety	2 (10.5)	0 (0)
Respiratory, thoracic and mediastinal disorders	Epistaxis	2 (10.5)	0 (0)

Source: Reviewer analysis ADAE dataset

Eighteen subjects started conditioning and received beti-cel. Non-laboratory AEs reported by $\geq 10\%$ of subjects starting from Day 1 to Month 24 are presented below:

Table 39 Non-laboratory Adverse Events. N=18

Body System Organ Class/ Group Term/Preferred Term	N All Gr	% All Gr	N Gr3 and Gr4	% Gr 3 and Gr 4
Blood and lymphatic system disorders				
Febrile neutropenia	12	66.7	12	66.7
Gastrointestinal disorders				
Abdominal pain	7	38.9	0	0
Constipation	4	22.2	0	0
Diarrhea	4	22.2	0	0
Mucositis	18	100	10	55.6
Nausea	3	16.7	0	0
Vomiting	8	44.4	0	0
General disorders and administration site conditions				
Fatigue	2	11.1	0	0
Pain	2	11.1	0	0
Puncture site pain	2	11.1	0	0
Pyrexia	7	38.9	1	5.6
Infections and infestations				
Nasopharyngitis	3	16.7	0	0
Oral herpes	3	16.7	0	0
Viral infection	2	11.1	1	5.6
Injury, poisoning and procedural complications				
Procedural pain	6	33.3	0	0
Transfusion reaction	2	11.1	0	0
Metabolism and nutrition disorders				

Decreased appetite	5	27.8	3	16.7
Musculoskeletal and connective tissue disorders				
Muscular weakness	2	11.1	0	0
Musculoskeletal pain	6	33.3	0	0
Nervous system disorders				
Dizziness	2	11.1	0	0
Headache	5	27.8	0	0
Respiratory, thoracic and mediastinal disorders				
Cough	4	22.2	0	0
Dyspnea	2	11.1	0	0
Epistaxis	7	38.9	3	16.7
Hypoxia	3	16.7	1	5.6
Oropharyngeal pain	2	11.1	0	0
Skin and subcutaneous tissue disorders				
Alopecia	11	61.1	0	0
Erythema	2	11.1	0	0
Pigmentation disorder	5	27.8	0	0
Pruritus	3	16.7	0	0
Rash	7	38.9	0	0
Vascular disorders				
Hypertension	3	16.7	0	0

Source: Reviewer analysis ADAE dataset. Abrv. Gr = Grade

Reviewer Comment

The period from study enrollment through mobilization and until just before conditioning has a fairly low rate of AEs, most of which were low grade and self-resolving. These are events that are known to be associated with G-CSF and plerixafor. During the period from conditioning to < Month 24, AEs tended to largely reflect known pattern seen with busulfan conditioning chemotherapy. The AEs in this period include infections, cytopenias, and gastrointestinal AEs such as mucositis and vomiting, as well as alopecia.

AEs related to beti-cel

According to the sponsor, there have been 7 reported beti-cel-related AEs, all of which were considered non-serious.

Two of these AEs (abdominal pain on Day 1 in 2 subjects) were consistent with the presence of DMSO cryoprotectant in the beti-cel and have resolved. An additional subject experienced 4 AEs assessed as possibly related to beti-cel: Grade 2 thrombocytopenia on Day 64 to Day 240, Grade 2 neutropenia on Day 92 to Day 240, and Grade 2 leukopenia on Day 92, Grade 1 thrombocytopenia on Day 281, and Grade 3 autoimmune disorder on Day 817 that were ongoing as of last follow-up (Day 730). The event of autoimmune disorder was attributed to autoimmune thrombocytopenia.

Reviewer Comment

The reviewer agrees with attribution of all these AEs to other treatments than beti-cel. DMSO is known known to be associated with histamine release and can cause infusion reactions that include abdominal pain. However, the autoimmune disorder on Day 817 is more complex. It is described in more detail in section 8.4.8 under immune related events. The reviewer feels this event is likely transplant associated immune thrombocytopenia.

6.2.12.3 Deaths

No deaths occurred.

6.2.12.4 Nonfatal Serious Adverse Events

Five of 19 subjects experienced 12 SAEs. Of these, 2 of 19 (10.5%) occurred between time of informed consent form (ICF) signing to < mobilization (M), two of 19 (10.5%) during M to < conditioning (C), two of 18 (11.1%) C to < neutrophil engraftment (NE), two of 18 (11.1%) during NE to month 24 (M24), and 3 of 18 (16.7%) Day 1 (D1)to M24. Three of 18 treated subjects (16.7%) experienced treatment-emergent SAEs. Two of 18 (11.1%) experienced pyrexia as an SAE after beti-cel infusion.

Five of the 8 treatment emergent SAEs were attributed to the conditioning agent by the investigator, this included one event each of neutropenia, febrile neutropenia, stomatitis, and thrombocytopenia in Subject (b) (6) and 1 event of congestive cardiac failure in Subject (b) (6) . All events resolved and none led to study discontinuation, please see table below:

Table 40 HGB-212 Treatment Emergent Serious Adverse Events by Subject

Subject	Preferred Term	Study Phase	Start Day	End Day	Grade
(b) (6)	Pyrexia	NE to M24	53	58	2
	Febrile neutropenia	C to <NE	12	17	3
	Pyrexia	NE to M24	60	62	1
	Headache	NE to M24	60	62	1
	Neutropenia	C to <NE	11	35	4
	Stomatitis	C to <NE	12	17	3
	Thrombocytopenia	C to <NE	13	31	4
	Congestive heart failure	C to <NE	18	144	3

Source: Reviewer calculations ADAE dataset

Subject (b) (6)

This subject is a 24-year-old female who experienced Grade 3 congestive cardiac failure on Day 18 following beti-cel. Medical history included β -thalassemia, failure to thrive, iron overload, back pain, latent tuberculosis, oligomenorrhoea, asthma,

glucose tolerance impaired, vitamin D deficiency, hypothyroidism, osteoporosis. Concomitant medications within 30 days prior to the congestive cardiac failure event included alendronic acid, calcium carbonate, cholecalciferol, paracetamol, and flucloxacillin. On Day 18, the subject experienced sudden palpitations with shortness of breath at rest. The subject required 60% FiO₂ to maintain adequate oxygen saturation. A chest X-ray showed patchy infiltrates consistent with pulmonary edema. Electrocardiography demonstrated sinus rhythm. On the same day, an echocardiogram demonstrated a dilated left ventricular (LV) cavity with severely impaired systolic function and an LV ejection fraction (LVEF) of 21%. She was given diuretics, beta blockers, angiotensin-converting-enzyme (ACE) inhibitors, and chelation therapy. On Day 21, chelation therapy was held due to neutropenia. Over the next several days, the subject improved somewhat. Repeat echocardiogram showed an ejection fraction of 32%. On Day 32, the subject had an episode of vasovagal syncope. A follow-up transthoracic echocardiogram showed mildly dilated LV size, normal wall thickness, mild to moderately impaired LV systolic function with LVEF of 46%, good longitudinal systolic function, and regional wall motion abnormalities. The subject's general condition was improving, and she was off supplemental oxygen. On Day 38 follow-up cardiac MRI showed LVEF was 49% and cardiac T2* was 15.4 msec. Iron chelation therapy with deferoxamine was continued. On Day 61 the subject was discharged. On Day 138 the Investigator reported the subject was symptomatically improved without dyspnea or clinical signs of heart failure. The event of congestive cardiac failure was considered resolved on Day 144. The Investigator assessed the event met the serious criterion of important medical event and was not related to beti-cel. The event was attributed to conditioning treatment with busulfan.

Reviewer Comment

Congestive heart failure (CHF) in a young adult is an uncommon event. However, this subject's risk factors include chronic iron overload, and chronic anemia with compensatory high cardiac output. The subject did have elevated cardiac iron based on MRI T2 testing of 15.4 sec. (T2 values of less than 20 ms indicated clinically important iron loading, and severe cardiac iron loading if T2 is less than 10 ms⁷) The rigors of the conditioning process compounded potentially by the rarely reported cardiotoxicity of busulfan might have led to the CHF. The investigator favors attribution of this event to antecedent iron cardiotoxicity and busulfan. Mechanistically, infusion of an autologous hematopoietic stem cell product, even with cells transduced by LVV seems less likely as the cause.

Subject (b) (6)

On Day 53, the subject was admitted to the emergency room due to Pyrexia, with WBC count of $2.87 \times 10^9/L$. She had Blood cultures and a chest X-ray which excluded pulmonary consolidation. Empiric antibiotic therapy was initiated and after Day 54, the subject became afebrile. Blood cultures were negative and on

7 He T. Cardiovascular magnetic resonance T2* for tissue iron assessment in the heart. *Quant Imaging Med Surg*. 2014;4(5):407-412. doi:10.3978/j.issn.2223-4292.2014.10.05

Day 58, the event was considered resolved and the subject was discharged from the hospital.

Subject (b) (6)

Neutropenia reported Day 11 and resolved Day 35, febrile neutropenia reported Day 12 and resolved day 17. Stomatitis reported Day 12 and resolved Day 17, thrombocytopenia reported Day 13, and resolved on Day 31, and pyrexia reported Day 60 and resolved on day 62. On Day 60, she was admitted with pyrexia and headache, all cultures were performed and reported negative. She recovered on Day 62 and was discharged. No CBC results were reported.

Reviewer Comment

All these TESAEs, except the pyrexia, were reported early in the expected severe myeloablated state, and attributable to conditioning and resolved within expected time interval. The pyrexia did not have CBC reported, however resolved spontaneously and no evidence of infection was suggested from cultures.

6.2.12.5 Adverse Events of Special Interest (AESI)

As with any lentivirus vector product, detection of vector derived replication competent lentivirus and insertional oncogenicity would constitute an adverse event of special interest (AESI). In addition, infection, human immunodeficiency infection, bleeding, autoimmune disease were considered as AESIs by the sponsor. Autoimmune thrombocytopenia was experienced by Subject (b) (6). The review team also considered cytopenias, including delayed platelet engraftment as an AESI.

Lentiviral Vector Integration Site Analysis/Assessment of Clonal Predominance

Because lentiviral vectors are expected to integrate into host target cell genome, beti-cel therapy may pose a risk of mutagenesis and oncogenesis. ISA data were collected starting at six months post beti-cel infusion, and results are available for 14 subjects at Month 6, and 9 subjects at Month 24 (as study is ongoing). Integration site analyses showed polyclonal reconstitution with transduced HSCs for all subjects. The number of unique mappable integration sites ranged from 2125 to 31844. In individual subjects, ISA analyses showed that the most frequently represented IS had a frequency of 4.233% (ASH1L in Subject (b) (6) at Month 12.) No subjects met the criteria for clonal predominance. No subjects met criterion for oligoclonality (which was introduced as a revision of the integration site analysis (ISA) algorithm after data lock).

Vector-Derived Replication Competent Lentivirus

A concern for LLV gene therapy products is the potential for the generation of replication competent lentivirus (RCL). Consequently, subjects in the study were screened for RCL at Months 3, 6, 12, and 24 following beti-cel infusion. Sixteen subjects had sufficient follow-up to permit RCL assessment, and no vector derived RCL has been detected.

Reviewer Comment

ISA has not revealed predominant clones, and no cases of RCL nor malignancy have been reported.

Cytopenias

Recipients of myeloablative conditioning chemotherapy are expected to develop severe cytopenias before reconstitution of hematopoiesis after infusion of HSCs. Many dynamics, including the use of growth factors, can affect time to hematopoietic recovery. Prolonged thrombocytopenia was the most notable cytopenia observed in this study.

Neutropenia and Neutrophil recovery

Being an autologous HSPC product, beti-cel requires myeloablative chemotherapy prior to its administration. Busulfan infusion leads to ubiquitous cytopenias, and neutrophil engraftment (NE) was stipulated as a safety endpoint while subjects remained hospitalized. NE was defined as 3 consecutive absolute neutrophil count (ANC) laboratory values $\geq 0.5 \times 10^9/L$ obtained on different days after a post-transplant value of $< 0.5 \times 10^9/L$ by Day 43. G-CSF was permitted after day 21. NE was reached by median Day 26 (range 14-39). Delayed NE was not reported in this trial. However, the FDA noted in our ad hoc analysis that three of the 18 beti-cel recipients remained dependent on continued G-CSF for at least 7 days beyond reporting of NE. Although all subjects were reported to have achieved NE before day 42, two remained on G-CSF until Day 44, and Day 77, respectively. This concomitant therapy, though approved in the autologous transplant setting and often used, can confound exact determination of NE. The Agency will mention these findings in the label to inform clinicians and patients.

Platelet Recovery

Time to platelet engraftment was also monitored, although this safety endpoint was not based on meeting engraftment criterion by a specified day. In HGB-212, delayed platelet engraftment was frequent, although generally not associated with significant bleeding complications. Platelet engraftment was defined as 3 consecutive platelet count values $\geq 20 \times 10^9/L$ obtained on different days after post-transplant value $< 20 \times 10^9/L$, with no platelet transfusions in prior 7 days. Among the 18 treated subjects, platelet engraftment was achieved by Day 30 in 3 subjects, between Day 30 and Day 60 in 11 subjects, and between Day 60 and Day 90 for the remaining 4 subjects. The median time to PE was 49.5 days, range of 21- 64 days.

Reviewer Comment

Beti-cel recipients experienced delayed platelet engraftment compared with published outcomes patients with TDT undergoing allo-HSCT, who typically engrafted by Day 25- 30. While delayed PE was not associated with bleeding events among study subject, prolonged thrombocytopenia is a known risk for bleeding complications, and the Agency will include clear warnings about the risk delayed platelet engraftment in the label. Neutrophil engraftment determination was impacted by confounding from G-CSF administration for prolonged periods after neutrophils were reported to have engrafted. In some subjects this confounding was continued beyond Day 42, and information about this potential concern will also be included in the label.

Thrombocytopenia and Slow Platelet Recovery

Platelet recovery beyond the threshold for platelet engraftment is also considered delayed (for example to thresholds such as $50 \times 10^9/L$ or $100 \times 10^9/L$). HGB-212 subjects treated with beti-cel reached an unsupported platelet count of $\geq 50 \times 10^9/L$ at a median of 55 days (range 21 to 142) and required a median of 60 days (range 24 to 424) to reach an unsupported platelet count of $\geq 100 \times 10^9/L$. Most subjects do not return to their baseline platelet counts during the observation period on the study. The subjects' platelet trajectory generally demonstrated slow improvement and plateau, rather than recovery followed by subsequent decrease.

Reviewer Comment

The etiology for these platelet recovery observations is unknown. One hypothesis is that baseline platelet counts in TDT patients are relatively elevated due to the chronic dyserythropoiesis and inflammation, and therapy that ameliorates the TDT phenotype leading to transfusion independence, may also reverse the processes which had been pathologically elevating platelets, thus unmasking the lower platelet values seen after treatment. However, hematopoietic stem cells are transduced with LVV to manufacture beti-cel, and this could lead to changes in gene expression that cause pathologic reductions in platelet counts. The delayed platelet engraftment does not appear to denote increased risk of the product provided adequate warnings and description of this issue are included in the label.

6.2.12.6 Clinical Test Results

Laboratory abnormalities were analyzed with shift tables, which looked at baseline laboratory value grades and compared these with on treatment maximal grades. The table below summarizes treatment-emergent laboratory abnormalities reported in $\geq 10\%$ of subjects. (Safety population N = 18).

Table 41 Treatment Emergent Laboratory Based Abnormalities in $\geq 10\%$ Subjects

Laboratory Based Abnormality	All Grades n(%)	% Gr3 and Gr4
Chemistry		
Hyponatremia	10 (56)	1 (6)
Hypophosphatemia	13 (72)	3 (17)
Hypokalemia	10 (56)	1 (6)
Hyperkalemia	3 (17)	0
Serum Alkaline Phosphatase increased	8 (44)	0
Serum ALT increased	14 (78)	5 (28)
Serum AST increased	13 (72)	0
Bilirubin increased	6 (33)	0
Hypocalcemia	17 (94)	0
Hypercalcemia	3 (17)	0
Serum Creatinine increased	1 (6)	0
Hypoglycemia ^{\$}	2 (13)	0
Hyperglycemia ^{\$}	2 (13)	2 (13)
Hematology		
Anemia	18 (100)	18 (100)
Lymphopenia	17 (94)	9 (50)
Lymphocytosis	2 (11)	1 (6)
Neutropenia	18 (100)	18 (100)
Thrombocytopenia	18 (100)	18 (100)
Leukopenia	18 (100)	18 (100)

Source: Reviewer analysis from adlb.xpt dataset. ^{\$}Denominator 16 instead of 18

Abbreviations: ALT: alanine aminotransferase; AST aspartate aminotransferase

Reviewer Comment

- The ADLB dataset was used to generate incidence of laboratory- based AEs since this is more accurate as opposed to using the adverse event dataset (ADAE dataset).
- A “lab-shift” analysis was carried out wherein baseline laboratory abnormalities that worsened following treatment were recognized i.e., shift of a laboratory grade from a lower to higher grade.
- During the interval between conditioning and neutrophil engraftment (C to NE period), cytopenias are the expected result of conditioning chemotherapy, after which they tend to resolve as hematopoiesis recovers. Most cytopenia AEs were observed in the post myeloablative period and improved over time. No late or serious bleeding or infections events were documented in association with these cytopenias.
- Cytopenias of all grades were the most common laboratory abnormalities as expected and reflect toxicity of the beti-cel regimen, which necessarily mandates myeloablative conditioning chemotherapy.

6.2.12.7 Dropouts and/or Discontinuations

One subject dropped out after start of mobilization, and no subjects dropped out after start of conditioning or beti-cel.

6.2.13 Study Summary and Conclusions

Although HGB-212 specifically enrolled the most serious TDT phenotype, i.e., β^0/β^0 , beti-cel therapy led to transfusion independence in substantial majority 12 of 24 (86%) of TI evaluable subjects. The safety profile of beti-cel included no deaths, and no AEs resulting in study discontinuation, and the main observation was delayed platelet engraftment and prolonged thrombocytopenia. Beti-cel-related AEs include abdominal pain on the day of infusion in two subjects and mild chronic cytopenias in 1 subject. The SAEs and AEs are generally consistent with the known effects of mobilization/ conditioning, myeloablative conditioning, and HSC transplant. Neutrophil engraftment has been successful in all subjects, with no long-term sequelae of serious infection. While there were no excess late bleeding events, despite the delayed platelet engraftment, this is considered a special risk of the product. Platelet engraftment delay will be included in the label warnings and precautions section. There was no evidence of gene therapy-related insertional oncogenesis or replication competent lentivirus.

6.3 Trial #3

HGB-204. Phase 1/2, Open Label Study Evaluating the Safety and Efficacy of Gene Therapy in Subjects with β -Thalassemia Major by Transplantation of Autologous CD34+ Stem Cells Transduced Ex Vivo with a Lentiviral βA^{T87Q} -Globin Vector.

6.3.1 Objectives (Primary, Secondary, etc.)

Primary objective: To evaluate the safety of treatment with beti-cel in subjects with TDT

Secondary objective: To evaluate the efficacy of treatment with beti-cel in subjects with TDT

6.3.2 Design Overview

HGB-204 was a single-arm, multi-site, single dose, Phase 1/2 study of subjects with TDT. Initial subject enrollment was staggered with second subject beginning myeloablative conditioning only after the first subject achieved engraftment of neutrophils without SAEs (attributed to beti-cel). HGB-204 included adult and adolescent subjects, who were treated with a single dose of beti-cel; subjects had non- β^0/β^0 genotype or β^0/β^0 genotype. HGB-204 was composed of 4 stages:

1. Screening
2. Autologous stem cell mobilization with GCSF +/- plerixafor and apheresis, myeloablative conditioning with busulfan over 4-5 days followed by 72 h washout.
3. Infusion of vector dose $\geq 3 \times 10^6$ CD34+ cells/kg
4. Follow-up through engraftment in the hospital, and as an outpatient for 24 months after beti-cel infusion.

After the Month 24 Visit, consenting subjects were enrolled in long-term follow-up Study LTF-303, to be followed for up to an additional 13 years. It included an independent Data Monitoring Committee (DMC).

6.3.3 Population

Eligibility Criteria

- Age ≥ 12 to ≤ 35 years at the time of consent, including 3 adolescents < 18 years of age
- History of ≥ 100 mL/kg/year of RBCs or ≥ 8 transfusions of RBCs per year in each of the 2 years prior to enrollment

Of 19 subjects meeting the eligibility criteria, one was withdrawn by the investigator due to inadequate stem cells for beti-cel manufacture. This left 18 eligible subjects, including 3 adolescents (12, 16, and 16 years of age).

6.3.4 Study Treatments or Agents Mandated by the Protocol

Beti-cel was manufactured with a Generation 1 process of autologous CD34+ cell-enriched hematopoietic stem cells transduced with lentiviral vector encoding the β^{A-T87Q} -globin gene. Each beti-cel lot was prepared individually for each subject using their autologous cells, and each subject received a single dose of $\geq 3.0 \times 10^6$ CD34+ cells/kg on Day 1.

6.3.5 Directions for Use

Beti-cel(s) was thawed and administered via IV infusion at a dose of $\geq 3.0 \times 10^6$ CD34+ cells/kg. In cases where ≥ 1 beti-cel lot was administered, the second lot was administered immediately after the first.

Reviewer Comment

HGB-204 enrolled subjects with different age range than the Phase 3 studies, and included any TDT genotype, in addition, the dose of beti-cel administered was lower, than the case in Phase 3 studies. The beti-cel version administered was of substantially lower transduction percentage and beti-cel VCN than the generation of beti-cel used in Phase 3 studies. In consideration of these differences, safety data were used as supportive to those from Phase 3. Efficacy was not reviewed.

6.3.6 Sites and Centers

Ramathibodi Hospital, Mahidol University, Bangkok, Thailand, Children's Hospital of Philadelphia, Philadelphia, PA, Cell & Molecular Therapies and Institute of Haematology, Royal Prince Alfred Hospital, Camperdown, Australia, Ronald Reagan UCLA Medical Center, Los Angeles, CA, Children's Hospital of Chicago, Northwestern University, Chicago, IL, UCSF Benioff Children's Hospital, Oakland, CA.

6.3.7 Surveillance/Monitoring

The applicant ensured appropriate monitoring procedures were performed before during and after the study. Screening procedures to ensure eligibility of candidates was performed with review of records and baseline laboratory and imaging tests. An independent Data Monitoring Committee (DMC) comprised of members with appropriate scientific and medical expertise monitored the safety of the study. Detailed schedule of events from HGB-204 is in the appendix.

6.3.8 Endpoints and Criteria for Study Success

For this early phase study, the primary endpoint was safety. The following endpoints were assessed:

- Success and kinetics of HSC engraftment
- Incidence of transplant-related mortality through 100- and 365-days post beti-cel infusion, and overall survival
- Detection of vector-derived replication competent lentivirus (RCL) in any subject
- Integration Site Analysis (ISA) to determine the presence of clonal dominance
- Descriptive analysis of adverse events
- Changes in laboratory parameters and frequency and severity of clinical AEs.

Primary Efficacy Endpoint

The proportion of subjects with a sustained production of ≥ 2.0 g/dL of hemoglobin A (HbA) containing β^{A-T87Q} -globin for the 6 months between Month 18 and Month 24 post- beti-cel infusion

Reviewer Comment

Due to substantial differences between the version of beti-cel administered in HGB-204 and the later beti-cel administered in Phase 3 studies, efficacy data from HGB-204 were not reviewed. The primary focus for review of Study HGB-204 was safety.

6.3.9 Statistical Considerations & Statistical Analysis Plan

Analyses followed the Statistical Analysis Plan (SAP) for Study HGB-204 (v2.0, dated 23 August 2017). The following subject populations were evaluated and used for presentation and analysis of the data:

- Intent-to-Treat Population (ITT): All subjects who initiated any study procedures, beginning with mobilization by G-CSF with or without plerixafor.
- Transplant Population (Efficacy Analysis Population [EAP]): All subjects in ITT population receiving beti-cel infusion.
- Successful Engraftment Population (SEP): All subjects who had successful NE after beti-cel infusion.

- The primary population for analysis of safety was ITT. The primary population for efficacy analyses was the EAP. SEP was the same as the EAP in this study, and so no separate SEP analyses were performed.

Reviewer Comment

The study design, and statistical methods appear adequate for an early phase exploratory study. The regulatory efficacy endpoint of transfusion independence, unlike the protocol efficacy endpoint, should be informative of clinical benefit to subjects. Please refer to Statistical review for detailed information.

6.3.10 Study Population and Disposition

6.3.10.1 Populations Enrolled/Analyzed

HGB-204 enrolled 19 eligible subjects but one was withdrawn from study due to “inadequate stem cell mobilization” after 1 mobilization cycle. The remaining 18 were infused with beti-cel, successfully engrafted, and completed the study.

6.3.10.1.1 Demographics

Please see table above. The 13 (72.2%) female subjects outnumbered the 5 (27.8%) males. A total of 77.8% of participants were Asian, and 22.2% White. A total of 83.3% of subjects were aged 18 and above, and 16.7% were 12 to < 18 years of age. The participants met inclusion criteria based on genotype and RBC transfusion dependence criteria. Please see table below.

Table 42 Demographic and Medical History Parameters, by Genotype

Parameter	Statistic	Non- β^0/β^0 (N=10)	β^0/β^0 (N=8)	Overall (N=18)
Sex				
Male	n (%)	3 (30.0)	2 (25.0)	5 (27.8)
Female	n (%)	7 (70.0)	6 (75.0)	13 (72.2)
Race				
Asian	n (%)	8 (80.0)	6 (75.0)	14 (77.8)
White	n (%)	2 (20.0)	2 (25.0)	4 (22.2)
Age at Informed Consent or Assent (category)^a				
≥18 years	n (%)	8 (80.0)	7 (87.5)	15 (83.3)
≥12 to <18 years	n (%)	2 (20.0)	1 (12.5)	3 (16.7)
<12 years	n (%)	0	0	0
Spleen Present				
Yes	n (%)	7 (70.0)	5 (62.5)	12 (66.7)
No	n (%)	3 (30.0)	3 (37.5)	6 (33.3)
Medical History in 2 Years Prior to Study Enrollment				
pRBC (mL/kg/year)	Median (min, max)	151.28 (140.0, 234.5)	182.59 (124.4, 273.2)	169.05 (124.4, 273.2)
pRBC (number transfusions/year)	Median (min, max)	13.75 (10.0, 16.5)	13.75 (12.5, 17.5)	13.75 (10.0, 17.5)
Weighted mean Hb nadir (g/dL)	Median (min, max)	9.1 (7.0, 9.8)	9.4 (8.7, 10.1)	9.31 (7.0, 10.1)
Hb trigger (g/dL)	Median (min, max)	7.0 (7.0, 9.0) N=5 ^b	9.0 (7.0, 9.0) N=3 ^b	8.0 (7.0, 9.0) N=8 ^b
Hospitalization, 2 years prior to enrollment (days/year)	n (%)	0	0	0
Hospitalization, 2 years prior to enrollment (visits/year)	n (%)	0	0	0

Parameter	Statistic	Non- β^0/β^0 (N=10)	β^0/β^0 (N=8)	Overall (N=18)
Age at:				
Consent/assent (years) ^a	Median (min, max)	19.5 (16, 34)	23.0 (12, 35)	20.0 (12, 35)
β -Thalassemia Major Diagnosis (months)	Median (min, max)	69.5 (0, 315)	4.5 (0, 92)	7.5 (0, 315)
1 st pRBC transfusion (months)	Median (min, max)	30.0 (0, 132)	6.0 (2, 12)	10.0 (0, 132)
Estab. regular pRBC transfusion regimen (months)	Median (min, max)	72.0 (8, 312)	7.0 (2, 84)	42.0 (2, 312)
Start iron chelation (years)	Median (min, max)	7.5 (2, 26)	3.5 (2, 18)	6.5 (2, 26)

Source: Original sBLA 125717/02: Clinical Study Report HGB-204. Page 83-84.

Reviewer Comment

Demographics correlate with the regions of the World where TDT is prevalent. However, there were more female subjects than males. The study only included adults and adolescents who were at least 12 years old.

6.3.10.1.2 Medical/Behavioral Characterization of the Enrolled Population

Please see table above for baseline age at diagnosis, severity of disease (transfusion requirements) and use of chelators.

6.3.10.1.3 Subject Disposition

HGB 204 enrolled ITT population of N =19, and subjects in the ITT population who underwent beti-cel infusion (N=18) were considered the transplant population (EAP). One subject was withdrawn due to “inadequate mobilization of stem cells” after starting mobilization/apheresis. HGB-204 study disposition is summarized in the table below:

Table 43 Disposition by Genotype (ITT)

Parameter	Statistic	Non- β^0/β^0 (N=11)	β^0/β^0 (N=8)	Overall (N=19)
Subjects Mobilized	n (%)	11 (100.0)	8 (100.0)	19 (100.0)
Subjects Infused with Drug Product	n (%)	10 (90.9)	8 (100.0)	18 (94.7)
Subjects with Successful Engraftment ^a	n (%)	10 (90.9)	8 (100.0)	18 (94.7)
Subjects Completing the Study through their Month 24 Visit	n (%)	10 (90.9)	8 (100.0)	18 (94.7)
Subjects who Discontinued from Study	n (%)	1 (9.1)	0	1 (5.3)
Reasons for Study Discontinuation:				
Adverse Event	n (%)	0	0	0
Investigator Decision	n (%)	1 (9.1) ^b	0	1 (5.3) ^b

Source: Table 14.1.1.2; Listing 16.2.1.1

^a Defined as ANC $\geq 0.5 \times 10^9/L$ for 3 consecutive days post-LentiGlobin BB305 Drug Product infusion or ANC $\geq 0.5 \times 10^9/L$ for 3 consecutive measurements done on separate days.

^b Discontinued from the study due to inadequate stem cell mobilization (Listing 16.2.1.1)

Source: Original sBLA 125717/02: Clinical Study Report HGB-204. Page 81.

6.3.11 Efficacy Analyses

HGB-204 Data were not analyzed for efficacy, only for supportive safety as the product version was less potent compared to the commercial product.

6.3.12 Safety Analyses

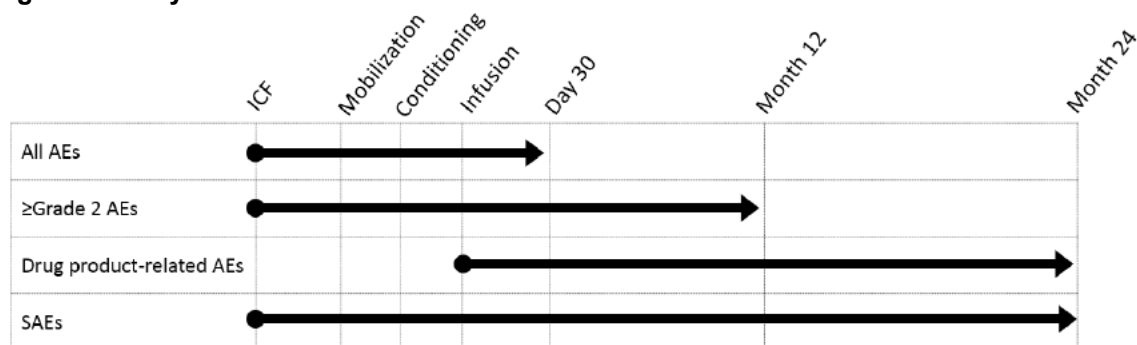
6.3.12.1 Methods

Key materials used for the safety review included:

- The BLA application electronic submission
- Applicant submissions in response to the review team’s information requests
- Proposed labeling for beti-cel
- Published literature
- Prior regulatory history

The clinical review of safety was primarily based upon analysis of 19 subjects in Study HGB-204, which was completed on 21 February 2018. Analysis datasets (ADaM datasets) were used for the safety analysis. Analyses by the clinical reviewer for safety were performed using JMP 16. All narratives and relevant CRFs were reviewed for all SAEs that occurred in the primary safety population. AEs were coded using the MedDRA version 19.0, and AE severity was graded using the National Cancer Institute's NCI's CTCAE version 4.03. Concomitant medications were coded using the WHO Drug Dictionary. Some AEs are presented throughout this review as grouped terms as defined by the review team. The complete list of FDA's grouped terms is presented in APPENDIX. The safety analysis set included all subjects treated with any dose of beti-cel. All AEs were collected from the signing of ICF until Day 30 after beti-cel infusion, those \geq Gr 2 were collected until month 12. SAEs were collected until Month 24 and were defined as any AEs that met at least one of the following criteria: fatal, life threatening, required inpatient hospitalization or prolongation of existing hospitalization, resulted in persistent or significant disability, resulted in congenital anomaly or birth defect, or resulted in any other medically important serious event. TEAEs were defined as all AEs occurring after initiation of beti-cel administration through the month 24 visit. Please see figure below:

Figure 3 Safety outline in HGB-204



Source: From HGB-204 Study Report V 1.0, Page 61

The ITT population was the primary population for the analysis of safety parameters (N=19). This population is defined as all subjects who initiated any study procedure, beginning with mobilization. Nineteen subjects underwent mobilization and apheresis (ITT), but one was withdrawn before conditioning. Exposure to busulfan is summarized below:

Table 44 HGB-204 Conditioning Chemotherapy Exposure

Parameter	Statistic	Overall (N=18)
Average Daily Dose (mg/kg/day) ^a	Mean (SD)	3.29 (0.517)
	Median	3.18
	Min, Max	2.7, 4.3
Estimated average AUC (µM*min) ^b	Mean (SD)	3972.7 (485.44)
	Median	4092.5
	Min, Max	3030, 4714

Source: Table 14.1.7

Note: The weight closest but prior to the first date of conditioning is used in calculation for dose (mg/kg).

^a According to protocol-allowed dosing as per regional standard of care, Subjects (b) (6) both in Australia, received half the recommended dose on the first 2 days of dosing, followed by 3 days of dosing at the recommended dose.

^b Average AUC by subject using observed and derived AUC (where derived AUC is defined using the average of the observed AUC per dose, multiplied by observed dose when AUC is missing).

Source: Original BLA 125717 HGB 204 CSR Page 163

Beti-cel Dose

The Protocol stipulated $\geq 3 \times 10^9$ /L CD34+ cells/Kg. The median dose delivered was 8.1×10^9 /L CD34+ cells/Kg; range 5.2 to 18.1.

Reviewer Comment

Subjects received adequate myeloablation and received at least the recommended protocol dose of beti-cel.

6.3.12.2 Overview of Adverse Events

The non-laboratory treatment emergent adverse events (TEAEs) in the N=18 beti-cel recipients were reported below:

Table 45 AEs reported in $\geq 10\%$ of subjects, per Body System Organ Class and Preferred Term, D1 to Month 24

Body system organ class	Group-Term	N with All Grade AEs (%)	N with Grade ≥ 3 AEs (%)
Blood and lymphatic system disorders	Febrile neutropenia	11(61)	10 (56)
Eye disorders	Conjunctivitis	2 (11)	0
Gastrointestinal disorders	Abdominal pain	6 (33)	1 (6)
	Constipation	8 (44)	0
	Diarrhea	7 (39)	1 (6)
	Dyspepsia	6 (33)	1 (6)

	Gastrointestinal inflammation	2 (11)	1 (6)
	Mucositis	16 (89)	14 (78)
	Nausea	2 (11)	0
	Proctalgia	2 (11)	0
	Toothache	2 (11)	0
	Vomiting	9 (50)	0
General disorders and administration site conditions	Catheter site pain	2 (11)	0
	Fatigue	4 (22)	0
	Pyrexia	5 (28)	1 (6)
Hepatobiliary disorders	Veno occlusive liver disease	3 (17)	2 (11)
Immune system disorders	Drug hypersensitivity	2 (11)	0
Infections and infestations	Anal abscess	2 (11)	0
	Bacteremia	2 (11)	1 (6)
	Cellulitis	2 (11)	1 (6)
	Upper respiratory tract infection	4 (22)	0
Injury, poisoning and procedural complications	Contusion	2 (11)	0
	Infusion related reaction	2 (11)	0
	Procedural pain	2 (11)	0
	Skin abrasion	3 (17)	0
	Skin laceration	2 (11)	1 (6)
	Transfusion reaction	8 (44)	1 (6)
Metabolism and nutrition disorders	Decreased appetite	3 (17)	0
Musculoskeletal and connective tissue disorders	Musculoskeletal pain	7 (39)	0
Nervous system disorders	Dizziness	2 (11)	0
	Headache	2 (11)	1 (6)
Psychiatric disorders	Insomnia	4 (22)	0

Reproductive system and breast disorders	Menstruation irregular	4 (22)	3 (17)
	Ovarian failure	2 (11)	0
	Vaginal hemorrhage	6 (33)	0
Respiratory, thoracic and mediastinal disorders	Cough	4 (22)	0
	Epistaxis	9 (50)	2 (11)
Skin and subcutaneous tissue disorders	Alopecia	17 (94)	0
	Ecchymosis	2 (11)	0
	Pigmentation disorder	5 (28)	0
	Pruritus	4 (22)	0
	Rash	5 (28)	0
	Urticaria	3 (17)	0
	Xerosis	2 (11)	0

Source: Reviewer analysis ADAE dataset.

Reviewer Comment

Non-laboratory adverse events were mostly mild, with no Grade 4 events. These toxicities are common with myeloablative cytotoxic chemotherapy with busulfan. No specific safety signals related to beti-cel treatment were identified.

6.3.12.3 Deaths

No deaths occurred

6.3.12.4 Nonfatal Serious Adverse Events

SAEs reported within 6 months of beti-cel infusion generally reflected autologous HSCT. Other SAEs included wild type HIV infection with a number of complications, and there were also thrombotic SAEs, which were likely attributable to iatrogenic (concomitant) medications. The SAEs reported in HGB-204 are listed in tabular form below

Table SAEs experienced in HGB-204 Day 1 to Month 24

Body System Organ Class	Preferred Term	Start day AE	End day, or not resolved status	AE number
Blood and lymphatic system disorders	Anemia	624	656	1
Blood and lymphatic system disorders	Neutropenia	829	848	1
Cardiac disorders	Intracardiac thrombus	357	458	1

Hepatobiliary disorders	Gallbladder enlargement	1294	1338	1
Hepatobiliary disorders	Gallbladder polyp	1294	1338	1
Hepatobiliary disorders	VOD	27	48	1
Hepatobiliary disorders	VOD	29	50	Event #2
Infections and infestations	Appendicitis	9	21	1
Infections and infestations	Asymptomatic HIV infection	693	ONGOING	1
Infections and infestations	Bacillus bacteremia	829	849	1
Infections and infestations	Cat scratch disease	624	684	1
Infections and infestations	Cellulitis	366	377	1
Infections and infestations	Diarrhea infectious	396	399	1
Infections and infestations	Gastroenteritis	432	437	1
Infections and infestations	Salmonella bacteremia	718	732	1
Metabolism and nutrition disorders	Diabetic ketoacidosis	1960	1961	1
Metabolism and nutrition disorders	Hyperglycemia	629	630	1
Pregnancy, puerperium and perinatal conditions	Ectopic pregnancy	1534	1590	1
Pregnancy, puerperium and perinatal conditions	Fetal death	2116	2138	1
Psychiatric disorders	Major depression	803	805	1
Respiratory, thoracic and mediastinal disorders	Pulmonary embolism	1960	ongoing	1
Vascular disorders	Brachiocephalic vein thrombosis	58	60	1

Source: Reviewer Calculation from ADAE dataset. Abbreviation VOD = veno occlusive disease

Reviewer Comment

Select, pertinent serious adverse events are further outlined in narrative form below. The SAEs were selected for further discussion due to factors including being related to infections, related to thrombosis, or veno occlusive disease. Infections are of interest because of cytopenias noted after conditioning

chemotherapy as required by the protocol, and thrombosis may be of concern given the novel nature of beti-cel and its impact on hemoglobin levels which could cause rheostatic changes and possibly increase risk of thrombosis. Veno occlusive disease is a known toxicity of busulfan, and subject in HGB-204 were not protocol stipulated to take prophylaxis before treatment (unlike later Phase 3 subjects), the higher rate of VOD reported led to modification of the Phase 3 protocols to include prophylaxis.

Subject (b) (6), a 21-year-old female, experienced an SAE of vena cava thrombosis (Day 58). Her history included splenomegaly, therapeutic embolization, patent ductus arteriosus, iron overload. Pertinent concomitant medication was medroxyprogesterone. On Day 51 the subject developed intermittent sharp chest and back pain that worsened with deep inspiration or cough and was not relieved with acetaminophen. On Day 58 she was diagnosed with innominate vein thrombosis (Grade 2) and treated with tissue plasminogen activator (t-PA), followed on Day 59 by removal of CVC. On Day 60, the subject was discharged from the hospital and the event was reported as resolved. The investigator assessed the event of vena cava thrombosis as unrelated to beti-cel and attributed the event to the central venous catheter placement.

Reviewer Comment

This SAE is likely related to the central venous catheter and medroxyprogesterone use, with background of splenomegaly (all factors associated with increased thrombotic risk) and resolved with treatment. The reviewer concludes that beti-cel was unlikely related to this event.

(b) (6)

A 27-year-old Asian female experienced an SAE of cellulitis on Day 366.

Reviewer Comment

Localized cellulitis one year after autologous SCT therapy is likely not related to the study drug, SAE resolved with antibiotics.

(b) (6)

A 21-year-old Asian female experienced SAEs of veno-occlusive liver disease (VOD) (Day 27). On Day 48 the subject improved and having completed defibrotide treatment, was discharged. The event of VOD was reported as resolved. The investigator attributed VOD to conditioning.

Intracardiac Thrombus (Day 357): At Month 12 follow-up visit, MRI of the heart showed a mass at the apex of the right ventricle. An echocardiogram confirmed the mass. She was admitted and anticoagulated. Day 357 platelet count was 341. Norethindrone and ethinyl estradiol was discontinued, IV heparin started. She was

discharged on rivaroxaban. The Day 377 echocardiogram found no evidence of thrombus. On Day 458, rivaroxaban was stopped, and the event was considered resolved. The investigator assessed the event of intracardiac thrombus (Grade 3) as not related to beti-cel.

Reviewer Comment

First SAE of VOD is a known toxicity of busulfan and would mechanistically not be expected from autologous stem cells; this event is likely due to conditioning procedure not beti-cel. The second SAE of Intracardiac thrombus occurred about a year post beti-cel, in the setting of high normal platelet count and a subject on a contraceptive known to be associated with thrombosis. It resolved with appropriate therapy. This reviewer agrees this SAE is unlikely related to beti-cel.

(b) (6)

A 21-year-old Asian male in Thailand experienced an SAE of appendicitis (Day 9), an SAE of diarrhea infectious (Day 396), and an SAE of asymptomatic HIV infection (Day 693).

Appendicitis (Day 9): On Day 9, while the subject was in the hospital for conditioning, he experienced appendicitis secondary to Klebsiella pneumoniae (Grade 3). D10, he had surgery after RBC and platelet transfusion, and then he improved and on Day 21 the event was considered resolved. He was discharged from the hospital on Day 44 once NE was achieved. The investigator assessed the event of appendicitis secondary to Klebsiella unrelated to beti-cel.

HIV: Subject was negative for HIV at Month 12, but positive at Month 24, (D693) reporting high risk sexual exposure. He was referred to infectious disease MD and started therapy.

Reviewer Comment

The SAE of appendicitis resolved with antibiotics and surgery and is likely not related to beti-cel. His HIV at Month 24 is likely from sexual activities, and not from beti-cel. After myeloablation and transplant, neutropenia is ubiquitous and infectious complications are common.

(b) (6)

A 17-year-old Asian female experienced an SAE of veno-occlusive liver disease (Day 29) and was hospitalized for therapy with defibrotide. On Day 50, the event of VOD was considered resolved and the subject was discharged from the hospital. The investigator considered the event of VOD not related to beti-cel but to busulfan.

Reviewer Comment

VOD is a known toxicity of busulfan and would mechanistically not be expected from autologous stem cells; this event is likely due to conditioning and not beti-cel. The product label will recommend prophylaxis against VOD before conditioning.

(b) (6)

A 12-year-old Asian male experienced an SAE of anemia (Day 624) and an SAE of cat scratch disease (Day 624). Presenting with 12-day history of intermittent fevers up to 104°F (40°C), persistent non-productive cough, fatigue, constipation for 3 days, and a 3 kg weight loss over the past month, subject had erythrocyte sedimentation rate (ESR) >130 mm/h (NR: 0-20), WBC of $7.1 \times 10^9/L$ (NR: $3.6-9.1 \times 10^9/L$), Hb of 76 g/L (NR: 128-160 g/L), platelet count $235 \times 10^9/L$. The subject's anemia was treated with 1 unit of pRBCs. On Day 632, a white punctate subretinal lesion in the right eye was observed. Infectious evaluation revealed Bartonella henselae IgG titer elevated at 1:1024. In addition, Epstein Barr virus (EBV) DNA was detected in a quantity of 3.93 log IU/mL (NR: no EBV DNA detected), treated with doxycycline. The constellation of prolonged fevers, weight loss and subretinal lesions was thought to be most consistent with cat scratch disease. The anemia was considered resolved on Day 656, and the cat scratch disease was considered resolved on Day 684. Investigator considered events of anemia (Grade 3) and cat scratch disease (Grade 3) unrelated to beti-cel.

Reviewer Comment

The subject suffered a very highly inflammatory systemic infection with Bartonella henselae, and this likely caused his anemia and infectious symptoms, resolving with appropriate antibiotics. This was not related to beti-cel.

6.3.12.5 Adverse Events of Special Interest (AESI)

Integration site analysis for clonal predominance was performed on subjects in the study and no subject reached definition of clonal predominance (no ISA relative frequency was >30%). Although no subject met the definition of clonal predominance, 2 subjects had unusual distributions of IS amongst the top10 IS:

- Subject (b) (6) showed a restricted, repetitive pattern of IS, with the same 2 IS (CBFB and XPO7) detected most frequently from Month 6 through Month 60. This pattern of expression was shown to be due to the presence of a progenitor having 2 IS in one cell, by analyses of IS in single erythroid colonies isolated from marrow sample. Further analyses using qPCR showed that clonal contribution was never higher than approximately 10% (at Month 24) and was approximately 5% (4.2% estimated by XPO7 and 4.6% by CBFB qPCR results) at Month 60. Integration sites that may be within the same clone include two proto-oncogenes, XPO7 and CBFB, and the following additional genes: DNAJC13, LINC00430, and ZMYM4. The IS-specific VCN in whole blood at the last reported visit on October 29, 2020, was 0425 c/dg in XPO7 and 0461 c/dg in CBFB.

- Subject (b) (6). The top 10 most frequent integration sites were primarily composed of the same IS between Month 24 and Month 60. This pattern is consistent with a major contribution by a progenitor having multiple IS in one cell, or with continued high relative contributions by a restricted number of progenitors. qPCR data estimated the clonal contribution of the 9 most frequent IS as between 7% to 15% at Month 60. Their similar clonal contributions make it possible that all these IS may be present in a single clone; however, this would have to be verified through single colony analysis or (b)(4). Nevertheless, none of these IS show a clonal contribution that would satisfy the criteria of a predominant clone (i.e., > 50% clonal contribution). Integration sites that may be within the same clone include a proto-oncogene, BCR, and the following additional genes: ASH1L-AS1, FNBP1, BTBD7, SACM1L, SFSWAP, PIP5K1A, SELP, TTBK2, and ZFAND3. For BCR, the integration site-specific vector copy number in whole blood at the last reported visit on November 18, 2020, was 0.1245 c/dg.

Reviewer Comment

These subjects had uncommon IS patterns but did not meet clonal predominance criteria. Of additional concern was the observation that one of these had the longest time to platelet engraftment of 191 days, and both had IS into known oncogenes. Section 9.2 contains discussion based on amended ISA algorithm submitted by the applicant after the 90-day safety update, further analyzing these two subjects.

Replication competent lentivirus

Replication competent lentivirus (RCV) screening was performed at Months 3,6, 12 and 24, and was negative.

Cytopenias

Initial safety concerns following ablation of bone marrow will be related to speed of engraftment of the infused stem cells to reconstitute the hematopoietic and immune systems. Neutrophil engraftment and platelet engraftment definitions were the same as in the Phase 3 studies discussed earlier.

Neutrophil and platelet engraftment:

- Median time to engraftment of neutrophils was 18.5 (range 14, 30) days.
- Median time to engraftment of platelets was 39.5 (range 19, 191) days.

Reviewer Comment

Platelet engraftment was delayed (relative to allo HSCT for TDT), which was also a concern reported in Phase 3 studies.

Bone marrow Morphology Abnormality

A non-serious event was assessed by the Investigator as possibly related to beti-cel; the Sponsor concluded the event was unlikely related to beti-cel:

Subject (b) (6), a 16-year-old female, experienced non-serious Grade 1 Dysplasia that was identified at the Month 24 Visit (Day 723). A Month 24 bone marrow biopsy revealed mild dysplastic changes in the erythroid series which were considered more prominent than in the previous bone marrow aspirate at Month 12 (N.B., the official pathology report of the Month 12 sample does not report dysplasia). The Month 24 pathology report stated that no significant neoplastic blast population was identified. Normal cytology was observed for lymphocytes, monocytes, and neutrophils. White blood cell and platelet counts were normal. Integration site analysis (ISA) from peripheral blood (PB) performed at Month 24 did not indicate emergence of a predominant clone. A follow-up bone marrow biopsy performed at Month 30 in this subject showed active trilineage hematopoiesis with maturation, no atypia and no blasts, and no dysplasia noted. ISA results up to Month 48 (Day 1441) showed no clonal predominance. Per Agency's request, bone marrow samples from Month 24 and Month 30 visit were compared by an independent pathologist on 07-Jan-2022. This independent pathologist revised the report to indicate "mild dysplastic-like changes in the erythroid series in bone marrow aspirates collected are now noted in both the Month 24 and Month 30 report". The revised Month 30 report also states when compared to previous Month 24 bone marrow biopsy, the current material (Month 30) is overall more cellular than the previous and the occasional dysplastic-like cells persist.

Reviewer Comment

Subject (b) (6) had findings of mild dysplastic-like changes reported on month 16, 24 and 30 marrow aspirate reports. These might be associated with her ongoing transfusion dependent thalassemia, especially since beti-cel therapy failed to induce transfusion independence. Unfortunately, she did not have a baseline marrow sample thus it is unknown if the changes preceded conditioning chemotherapy and beti-cel (baseline bone marrows were not mandated in Phase 1/2 studies). However, according to literature, such findings are noted in thalassemia patients, and occasionally in untreated, healthy marrow donors. She has continued follow up for approximately 6 years with regular RBC transfusions, and WBC and platelet counts remain unremarkable. Insertional site analysis (ISA) from month 48 visit in December 2019 was polyclonal. She missed her month 60 visit, but was tested at Year 6, results are pending. This subject's dysplastic-like changes are more likely related to persistent TDT, rather than MDS.

6.3.12.6 Clinical Test Results

Laboratory abnormalities which were adverse events were analyzed via shift tables comparing baseline abnormal grade to maximal treatment emergent grade. All subjects experienced AEs of anemia, thrombocytopenia, neutropenia and

leukopenia between D1 and month 24, which is anticipated following myeloablative conditioning chemotherapy. Other common laboratory AEs included liver enzyme and bilirubin abnormalities, which are common with busulfan chemotherapy. These are in the table below:

Table 46 Laboratory-Based Abnormalities in $\geq 10\%$ subjects

Laboratory Based Abnormality	All Grades n (%)	% Gr3 and Gr4
Chemistry		
Serum Alkaline Phosphatase increased	5 (27.8)	0 (0)
Serum ALT increased	16 (88.9)	2 (11.1)
Serum AST increased	13 (72.2)	1 (5.6)
Bilirubin increased	8 (44.4)	7 (11.1)
Hypocalcemia	14 (77.8)	0 (0)
Hypercalcemia	4 (22.2)	0 (0)
Hyperglycemia	1 (5.6)	1 (5.6)
Hypokalemia	16 (88.9)	2 (11.1)
Hyperkalemia	2 (11.1)	0 (0)
Hyponatremia	13 (72.2)	2 (11.1)
Hypernatremia	1 (5.6)	0 (0)
Hypophosphatemia ^{\$}	7 (43.8)	2 (12.5)
Hematology		
Anemia	18 (100)	17 (94.4)
Lymphopenia	18 (100)	11 (61.1)
Neutropenia	18 (100)	18 (100)
Thrombocytopenia	18 (100)	18 (100)
Leukopenia	18 (100)	18 (100)

Source: Reviewer analysis from adlb.xpt dataset. ^{\$}Denominator 16 instead of 18

Abbreviations: ALT: alanine aminotransferase; AST: aspartate aminotransferase

6.3.12.7 Dropouts and/or Discontinuations

One subject discontinued from the study after starting mobilization, at the investigator's decision, due to insufficient stem cell mobilization.

6.3.13 Study Summary and Conclusions

Due to overall differences in LVV product used in HGB-204 and the two Phase 3 studies, safety data from HGB-204 may lend support to the primary safety data from the phase 3 studies. Overall, the safety signal is similar to Phase 3 studies, where events potentially attributable to myeloablation dominate, along with notable delay of platelet engraftment.

6.4 Trial #4

LTF-303

6.4.1 Objectives (Primary, Secondary, etc.)

LTF-303 is a non-interventional study intended to monitor for long-term safety and efficacy of the gene therapy product used in bluebird bio-sponsored clinical studies in treated subjects with hemoglobinopathies.

6.4.2 Design Overview

This is a multi-center, long-term safety and efficacy study which accepts subjects completing the parent studies. During Study LTF-303, subjects are followed every 6 months through 5 years post-beti-cel infusion and then annually from 5 years through 15 years post-beti-cel infusion.

6.4.3 Population

All consenting subjects that received a bluebird LVV treatment for hemoglobinopathy and are able to comply with study requirements are eligible (No exclusion criteria). Although subjects with both TDT (treated with beti-cel or LentiGlobin HPV569 beti-cel) and subjects with SCD (treated with bb1111) are enrolled in Study LTF-303, as of 09 March 2021, the objectives of this interim CSR are to discuss the long-term safety and efficacy of beti-cel (Studies HGB-204, HGB-205, HGB-207, or HGB-212) or LentiGlobin HPV569 beti-cel (used only in the proof-of-concept Study LG001 and discontinued) to support regulatory submissions, including a marketing application for beti-cel.

6.4.4 Study Treatments or Agents Mandated by the Protocol

No investigational treatments are to be administered in this study

6.4.5 Directions for Use

Insert text here

6.4.6 Sites and Centers

Subjects are to be monitored at 13 study centers in Australia, England, France, Germany, Greece, Italy, Thailand, and USA

6.4.7 Surveillance/Monitoring

Subjects are followed every 6 months through 5 years post-beti-cel infusion and then annually from year 5 to 15 post-beti-cel infusion.

6.4.8 Endpoints and Criteria for Study Success

Safety Endpoints:

- Overall survival (OS)
- All drug-product related adverse events (AEs) through Year 15 post beti-cel infusion
- All serious adverse events (SAEs) through Year 15 post beti-cel infusion
- Immune-related AEs (e.g., autoimmune disorders, GVHD, opportunistic infections, HIV)

- New or worsening hematologic disorders or neurologic disorders
- Incidence of vector-derived replication competent lentivirus (RCL), assessed as clinically indicated
- Malignancies; subjects with insertional oncogenesis (myelodysplasia, leukemia, lymphoma, etc.)

Exploratory Endpoints:

- The number of subjects with clonal predominance

Pharmacodynamic Endpoints:

- Vector copy number (VCN) in peripheral blood over time post-beti-cel infusion to Year 5, Year 10, and Year 15
- β^{A-T87Q} -globin expression in peripheral blood to Year 5, Year 10, and Year 15
- Hemoglobin (Hb) fractions (including Hb^{AT87Q}, HbA, HbA2, HbF, HbE) to Year 5, Year 10, and Year 15.

Efficacy Endpoints:

- Proportion of subjects who meet the definition of transfusion independence (TI), defined as a weighted average Hb \geq 9 g/dL without any packed red blood cell (pRBC) transfusions for a continuous period of \geq 12 months at any time after beti-cel infusion in parent study and/or Study LTF-303
- Proportion of subjects with TI at timepoints including Year 5, 10, and 15 post-beti-cel, and at last follow-up
- Characterization of TI
- Characterization of transfusion reduction (TR)

Exploratory Endpoints:

- Annualized measures of health resource utilization from 6 months post-beti-cel infusion through last Follow up with Number of hospitalizations
- Events of interest related to complications of TDT
- Additional measures of dyserythropoiesis over time as compared to parent study baseline, assessed with Serum transferrin receptor, Erythropoietin, Hepcidin, and Bone marrow morphology

6.4.9 Statistical Considerations & Statistical Analysis Plan

Analyses were conducted per LTF-303 SAP, in which an interim analysis of subjects with TDT was conducted to provide supportive data for this marketing application for beti-cel. Subjects from the Efficacy Analysis population, defined as all subjects who were enrolled in Study LTF-303 and treated in a parent study with beti-cel, were evaluated and used for presentation and analysis of the data. Unless otherwise noted, analyses include data collected during the parent study pooled with the long-term follow-up data collected during Study LTF-303. Tabulations were produced for appropriate demographic, baseline, efficacy, pharmacodynamic, and safety parameters by study phase, within study phase by genotype, and by parent study. For categorical variables, summary tabulations of the number and percentage of subjects within each category (with a category for missing data) of the parameter are presented. For continuous variables, the

number of subjects, mean, standard deviation (SD), median, minimum, and maximum values are presented. Descriptive summary statistics as well as 2-sided, 95% confidence intervals, as appropriate, are presented on selected parameters. Longitudinal data were presented by appropriate time intervals, depending on the nature of the data. Baseline is defined by data collected in the original parent study for each subject prior to beti-cel infusion. Averaged data from the 2 years of retrospective pre-parent study enrollment (date of informed consent/assent in the parent study) (e.g., annualized pRBC transfusion requirements [volume and frequency], weighted Hb nadirs) were used as baseline. For other efficacy parameters as well as for pharmacodynamic parameters, baseline was defined as the most recent measurement prior to conditioning in the parent study. The conditioning start date was defined as the first date of busulfan administration. For safety parameters, including key laboratory (e.g., hematology and chemistry) parameters, the most recent value prior to the beginning of the autologous cell harvesting procedure in the parent study was used as the baseline assessment. All subjects enrolled in this study were evaluated for safety. Safety evaluations include the incidence of beti-cel-related AEs, SAEs, serious or non-serious immune-related AEs (e.g., autoimmune disorders, GVHD, opportunistic infections, HIV), and new or worsening hematologic or neurologic disorders or malignancies, as collected per the protocol. Changes in laboratory parameters were also evaluated. Any additional safety data are presented in data listings. Summarization focuses on incidence of beti-cel related AEs, treatment-emergent SAEs, immune-related AEs, new or worsening hematologic disorders, new or worsening neurologic disorders, and malignancies using the following mutually exclusive time periods post beti-cel infusion: Day 1 to Month 24, > Month 24 to Month 36, > Month 36 to Month 48, > Month 48 to Month 60, > Month 60 to Year 7, > Year 7 to Year 10, and > Year 10 to Year 15, based on calendar time. Please refer to Statistical review for detailed information.

6.4.10 Study Population and Disposition

A total of 47 subjects with TDT treated with beti-cel (18 from Study HGB-204, 19 from Study HGB-207, and 10 from Study HGB-212) consented to enroll in Study LTF-303, and all of these subjects continue to participate in Study LTF-303 as of the interim database lock for this CSR. No subjects have yet completed Study LTF-303.

6.4.10.1 Populations Enrolled/Analyzed

In this observational study, subjects from the Transplant Populations of the parent studies, were included and analyzed in LTF-303. The demographics and degree of representation to the broader population were discussed earlier in this review within respective sections of each parent study.

6.4.10.1.1 Demographics

Of the 29 Phase 3 study subjects enrolled, 13 were female (44.8%) and 16 were male (55.1%). Median (min, max) age at consent or assent in parent study was 17.0 (7, 34) years; 15 (51.7%) subjects were ≥ 18 years of age, 6 (20.6%) subjects

were ≥ 12 and < 18 years of age, and 8 (27.6%) subjects were < 12 years of age. Eleven (37.9%) subjects identified as Asian, and 16 (55.2%) subjects identified as White. Twenty-three (79.3%) subjects were of non- β^0/β^0 genotype, and 6 (20.1%) subjects were of β^0/β^0 genotype.

6.4.10.1.3 Subject Disposition

Twenty-nine subjects had a median of 29.5 months (range 22.9 - 48.2) of follow up. The 19 subjects from HGB-207 had a median of 30.6 months of follow up (range 24.4-48.2), while 10 subjects from HGB-212 had a median follow up of 25.2 months (range 23-36). There were no discontinuations.

6.4.11 Efficacy Analyses

There are 29 Phase 3 study subjects evaluable for TI. The following sections provide detailed analyses.

6.4.11.1 Analyses of Primary Endpoint(s)

A total of 25 of 29 Phase 3 study subjects achieved TI (86%). These subjects maintained a weighted average Hb level of 11.8 during IT (range 9.4, 13.7).

6.4.11.2 Analyses of Secondary Endpoints

Duration of TI. All subjects who achieved TI at any time were TI at their latest visit in Study LTF-303.

- The median (min, max) duration of TI was 26.3 (13.1, 39.4) months (N = 25).
- Median (min, max) weighted average Hb during TI was 11.8 (9.4, 13.7) g/dL at time of the interim Analysis, suggesting that most subjects who achieved TI were able to maintain normal or near normal level of total Hb.
- Subjects achieved a median (min, max) -100 (-100 , -3.4) % change in transfusion volume from 6 months after beti-cel infusion through last follow-up.

Hepatic and Cardiac Iron Burden

As was indicated in specific sections for Studies HGB-207 and HGB-212, most subjects had increasing LIC and cardiac iron burden between Baseline and month 12, then tended to revert towards baseline levels by the end of the parent study, though the levels remained above baseline. These parameters were further monitored in LTF-303, though very few subjects so far have had LIC assessments past Month 24 in Study LTF-303 (N= 5 at Month 36; N = 0 at Month 48). Similarly, only nine subjects have available cardiac T2* assessments for Month 36 in Study LTF-303.

Liver Iron Content

Only a total of 5 subjects had LIC data past Month 24 in Study LTF-303. Among these, the median liver iron was 0.4 (range) -1.52 to 0.81) higher at Month 36 (last date with available data) than at baseline.

Cardiac Iron Burden by Cardiac T2*

Cardiac T2* values were obtained in 9 subjects on LTF-303 through Month 36. The median change in cardiac T2* was -3.11 (-9.04-6.13). Note that decreasing cardiac T2* indicates increasing cardiac iron. Thus, among these participants, cardiac iron load was higher even at Month 36 than it had been at Baseline.

Ferritin Level

Ferritin levels at Month 36 were available in 10 Phase 3 subjects. Compared to baseline values, ferritin levels dropped by median (range) -35.7% (-79.8, 5.35), remaining at a median of 2096.5 pmol/L (range 283.1 to 17666)

Reviewer Comment

Hepatic and cardiac iron burden continued to modestly worsen after subjects completed 24 months of post beti-cel follow up in the parent studies and entered Study LTF-303. These findings suggest that as in the allogeneic HSCT setting, iron levels are slow to change following effective therapy for TDT. Ferritin, which indicated an early increase in the parent studies followed by a return to Baseline before Month 24, continued to decrease over the course of LTF-303. However, very small numbers of subjects with available data (N=5 for LIC, N=9 for cardiac T2), limits interpretation. Ferritin is known to be less specific and predictive than the other iron burden parameters, although it also does appear to respond faster than the other tools used to assess iron content.

Health Related Quality of Life

The Applicant followed subject Health-Related Quality of Life (HRQoL), assessed by questionnaires as a secondary endpoint. Quality of life assessments were to be collected at Month 24, 36, 48 and 60. As the studies are continuing, the overall data are limited beyond Month 24 (which was discussed above in the respective sections for Studies HGB-207 and HGB-212). However, for tools including (b)(4), the scores remained generally stable throughout long-term follow up. For (b)(4), scores generally remained stable or improved beyond Month 24 compared with Baseline scores. For the (b)(4) tool and the (b)(4), data were limited.

Reviewer Comment

Small numbers of subjects with evaluable QOL data and modest changes make drawing inferences from these outcomes challenging, especially given the limitations of QOL assessments in uncontrolled, open-label trials. As with time-to-event endpoints, interpretation of patient-reported outcomes is challenging in uncontrolled clinical trials, because it is unclear to what extent the outcomes can be attributed to the treatment effect of the regimen vs. to underlying disease and patient characteristics. Furthermore, the Applicant did not seek a labeling claim based on QOL data and these data were not incorporated in the PI. Finally, one of the tools used was generally designed and intended to be used in patients with

advanced cancers, and its validity in patients with hemoglobinopathy remains to be demonstrated.

6.4.11.3 Subpopulation Analyses

Refer to prior sections for the parent studies

6.4.11.4 Dropouts and/or Discontinuations

No subjects have dropped out or discontinued from the study.

6.4.11.5 Exploratory and Post Hoc Analyses

The study is ongoing and no exploratory or post hoc analyses were conducted.

6.4.12 Safety Analyses

6.4.12.1 Methods

All consenting subjects from parent studies were observed and analyzed for safety in Study LTF-303.

6.4.12.2 Overview of Adverse Events

There were no deaths or AEs resulting in study discontinuation. One subject experience an SAE of cholelithiasis on LTF-303 starting Day 837, which is discussed in the SAE section below, and another subject experienced one non-serious AE of Autoimmune disorder, discussed in section 8.4.8 in detail.

6.4.12.3 Deaths

No deaths were reported.

6.4.12.4 Nonfatal Serious Adverse Events

In analyzing Phase 3 subjects in LTF-303 who were reported to have SAEs after Month 24 (i.e., those reported after end of the Phase 3 study, and while followed in LFT-303), one SAE experienced an event of cholelithiasis.

6.4.12.5 Adverse Events of Special Interest (AESI)

No malignancies were reported. There was no evidence of insertional oncogenesis or LVV-derived RCL. Subject (b) (6) experienced one non-serious AE of Autoimmune disorder, attributed to autoimmune thrombocytopenia, which was assessed as possibly related to beti-cel by the investigator. This event was reported at the Month 24 Visit of parent Study HGB-212 and it is ongoing as of the database lock date. This is discussed in section 8.4.8 in greater detail.

6.4.12.6 Clinical Test Results

One subject experienced Grade 3 hyperbilirubinemia.

6.4.12.7 Dropouts and/or Discontinuations

There were no dropouts or discontinuations.

6.4.13 Study Summary and Conclusions

LTF-303 provided safety data beyond the 24 month follow up duration of the parent studies. Most subjects enrolled in Study LTF-303 achieved TI (25/29 subjects; 20/23 subjects of non- β^0/β^0 genotype and 5/6 subjects of β^0/β^0 genotype) and maintained TI through to their last follow-up, with the longest duration of TI of 39.4 months. For subjects who achieved TI, median weighted average Hb during TI was 11.8 g/dL. While the sample size of Phase 3 study subjects with iron burden data is small, liver and cardiac iron content generally remain above what it had been at baseline, whereas serum ferritin, indicates a trend for subjects who achieved TI having increased benefit.

No deaths, no beti-cel-related SAEs, and no AEs leading to study discontinuation have been reported. The one reported SAE was unrelated to beti-cel (cholelithiasis). There were no reports of new or worsening rheumatologic, autoimmune, neurologic or hematologic disorders, or malignancies with onset during Study LTF-303. In addition, there was no evidence of insertional oncogenesis or replication competent lentivirus. Interim data from Study LTF-303 demonstrate benefit of beti-cel without identification of any new safety signals.

7. INTEGRATED OVERVIEW OF EFFICACY

7.1 Indication #1

Treatment of subjects with β thalassemia who require regular red blood cell (RBC) transfusions

7.1.1 Methods of Integration

Efficacy data from the Phase 3 studies HGB-207 and HGB-212 were pooled, as the study designs were nearly identical with the exception of the genotype of the study population and subjects administered the same dose of beti-cel made using the same manufacturing process. Upon completing the final, Month 24 visit, subjects from all primary studies were invited to enroll in the long-term observational study, Study LTF-303. While this study focused on safety, it also collected additional efficacy data informing of long-term outcomes and durability. Data from Phase 1/2 studies were not pooled as they used different beti-cel manufacturing processes and differing doses of beti-cel were administered. For the integrated assessment of efficacy, data from the efficacy analysis population (EAP) were analyzed.

7.1.2 Demographics and Baseline Characteristics

Demographics of EAP in the Phase 3 studies HGB-207 and HGB-212 are summarized below:

Table 47 Demographics of Phase 3 study subjects; Age

Parameter	Statistic	HGB-207 N=23	HGB-212 N=18	Total N=41
<12	n (%)	8 (34.8)	8 (44.4)	16 (39)
≥12 to <18	n (%)	6 (26.1)	5 (27.8)	11 (26.8)
≥18	n (%)	9 (39.1)	5 (27.8)	14 (34.1)

Sex

Parameter	Statistic	HGB-207 N=23	HGB-212 N=18	Phase 3 Total N=41
Male	n (%)	11 (47.8)	10 (55.6)	21 (51.2)
Female	n (%)	12 (52.2)	8 (44.4)	20 (48.8)

Race

Parameter	Statistic	HGB-207 N=23	HGB-212 N=18	Phase 3 Total N=41
Asian	n (%)	13 (56.5)	7 (38.9)	20 (48.8)
White	n (%)	8 (34.8)	10 (55.6)	18 (43.9)
Other	n (%)	2 (8.7)	0	2 (4.9)
Not reported	n (%)	0	1 (5.6)	1 (2.4)

Ethnicity

Parameter	Statistic	HGB-207 N=23	HGB-212 N=18	Phase 3 Total N=41
Latino	n (%)	1 (4.3)	0	1 (2.4)
Non-Latino	n (%)	21 (91.3)	17 (94.4)	38 (92.7)
Not reported	n (%)	1 (4.3)	1 (5.6)	2 (4.9)

Source: Reviewer calculations, ADSL dataset from ISE

7.1.3 Subject Disposition

Of the N=41 subjects in the Phase 3 studies, one subject from each study discontinued before conditioning but after starting mobilization. HGB-212 had no discontinuations due to screening failure; HGB-207 had five screen failures and 3 subjects who withdrew consent before mobilization. The screening failures were due to excessive organ disease or insufficient TDT severity. These data are presented below:

Table 48 Disposition of phase 3 Study Subjects

Parameter	Statistic	HGB-207	HGB-212	Phase 3 Total
Number of subjects	N	32	19	51
Signed ICF	n (%)	8 (25.0)	0	8 (15.7)
Screening failed	n (%)	5 (15.6)	0	5 (9.8)
Withdrew consent before mobilization	n (%)	3 (9.4)	0	3 (5.9)
Received Mobilization (ITT)	n (%)	24 (75)	19 (100)	43 (84.3)

Source: Calculated by reviewer ISE ADSL

Reviewer Comment

Screen failure did occur for excessive organ disease and insufficient TDT severity. This should not significantly impact applicability to the general population. Due to the known toxicity of autologous transplant/myeloablation, patients should be considered beti-cel candidates provided adequate organ function (especially liver and heart).

7.1.4 Analysis of Primary Endpoint(s)

Phase 3 study efficacy endpoints were the proportion of subjects who met the definition of “transfusion independence” (TI) at any time after beti-cel infusion. As the phase 3 studies are ongoing, not all subjects treated have had sufficient follow up, therefore, 36 of 41 beti-cel recipients (88%) are evaluable for assessment of transfusion independence (TI evaluable). Of the 36 subjects in the TI evaluable population, 32 (89%) of the subjects reached TI at any time. The 2 sided 95% CI is 73.9 to 96.9. Four (11%) of the 36 subjects in the efficacy analysis population did not meet TI in the Phase 3 studies, two from HGB 207 and two from HGB 212. In addition, five subjects are not yet evaluable for TI. The TI-evaluable subjects who did not achieve TI had the lowest PB VCN values of TI-evaluable subjects and were producing the lowest amounts of Hb^{AT87Q} at Month 6 in their respective studies. The subjects’ beti-cel parameters (i.e., beti-cel VCN and %LVV+ Cells) were below the median for the Phase 3 subjects. TI outcomes are summarized below:

Table 49 Phase 3 study Transfusion Independence Outcomes

Parameter	Statistic	HGB-207	HGB-212	Phase 3 total (N=41)
TI-evaluable	N (%)	23	14	36
Subjects with TI at any time	n (%) 2-sided 95% CI	20 (91) 55.4, 97.2	12 (86) 57.2, 98.2	32 (89) 73.9, 96.9

Source: Reviewer analysis, from ISE ADEFF2 dataset

Reviewer Comment

In the pooled analysis for the Phase 3 studies, 89% of the TI evaluable subjects achieved TI at any time. As these studies are ongoing at time of data lock, and not all subjects are TI evaluable, analysis of TI evaluable subjects is acceptable. It should be pointed out that per protocol, the “primary endpoint will be analyzed as a point-estimate of the proportion of subjects achieving TI, with a 2-sided 95% confidence interval calculated using the exact binomial method. The EAP will be used for primary conclusions of gene-therapy efficacy.” Thus, beti-cel efficacy was also analyzed in the entire population treated with beti-cel (not just those currently TI evaluable). In Study HGB-207 20/24 = 83% (95% C.I. 62.6, 95.6) and in Study

HGB-212 12/19 = 63% (95% C.I. 38.4, 83.7) of treated subjects achieved TI. Of the N=41 total Phase 3 subjects, 32 (78%; 95% C.I. 62.4, 89.4) achieved TI.

7.1.5 Analysis of Secondary Endpoint(s)

Secondary efficacy endpoints were designed to further evaluate and characterize the impact of beti-cel on TI, and these are presented in the table below:

Table 50 Characterization of Transfusion Independence (TI subjects only)

Parameter	Statistic	Phase 3 total (N=32)
Maintaining TI at		
Month 24	n/N (%)	27/31 (87)
Month 36	n/N (%)	9/10 (90)
Month 48	n/N (%)	0/1
Observed duration of TI (months)	Median (range)	25.7 12.5, 39.4
Time from beti-cel to last transfusion prior to TI (months)	Median (range)	0.84 0, 2.4
Time from beti-cel to achievement of TI (months)	Median (range)	15.7 14.8, 24.5
Weighted avg. Hb during TI (g/dL)	Median (range)	11.5 9.3, 13.7

Source: Reviewer calculations from ISE ADEF2 and ATTE dataset

Summary of Transfusion Independence

Out of 36 TI evaluable subjects in the Phase 3 studies, 32 (89%) achieved TI, and those who achieved TI, have maintained it for a median 25.7 (12.5, 39.4) months. Median time from beti-cel infusion to last pRBC transfusion among TI achievers was median 0.84 (range 0-2.4) months. Median weighted avg. Hb was 11.5 gm/dL during TI (range 9.3-13.7).

Reviewer Comment

Subjects achieving TI tended to remain independent from transfusions to last follow up, and they were capable of maintaining a weighted average Hb during TI, which equaled or exceeded their pretreatment baselines. Median duration of TI was 25.7 months, suggesting durability of effect.

In addition to being evaluated for the achievement of transfusion independence, transfusion reduction (TR) compared to Baseline, was also analyzed in the studies. Results are in the table below:

Table 51 Proportion of Subjects with % Reduction in Transfusion Volume; Month 6 through Last Follow-up Compared to Baseline

Parameter	Statistic	<50%	≥60%	≥75%	≥90%	100%
Phase 3 Overall (N=39)	n (%) 2 sided 95% CI	3 (7.7) (1.6, 20.9)	36 (92.3) (79.1, 98.4)	36 (92.3) (79.1, 98.4)	36 (92.3) (79.1, 98.4)	35 (89.7) (75.8, 97.1)
TI (N=32)	n (%) 2 sided 95% CI	0 (0, 10.9)	32 (100) (89.1, 100)	32 (100) (89.1, 100)	32 (100) (89.1, 100)	32 (100) (89.1, 100)
Non-TI (N=4)	n (%) 2 sided 95% CI	3 (75.0) (19.4, 99.4)	1 (25.0) (0.6, 80.6)	1 (25.0) (0.6, 80.6)	1 (25.0) (0.6, 80.6)	0 (0, 60.2)
Not TI evaluable (N=3)	n (%) 2 sided 95% CI	0 (0, 70.8)	3 (100) (29.2, 100)	3 (100) (29.2, 100)	3 (100) (29.2, 100)	3 (100) (29.2, 100)

Source: reviewer analysis, ISE ADEF1 dataset

7.1.6 Other Endpoints

7.1.7 Subpopulations

Subpopulation analyses by age, sex, race, and genotype subgroup were performed for TI, and are addressed in the following sections:

Transfusion Independence by Genotype

Analysis of TI at any time for subjects by genotype severity is presented in the Table below. Subjects with either genotype ($\beta 0/\beta 0$ vs. Non $\beta 0/\beta 0$) achieved similar percentages of TI. Of the subjects evaluable for TI in Phase 3 studies, 12/13 subjects with non- $\beta 0/\beta 0$ genotype and 7/8 subjects with a $\beta 0/\beta 0$ genotype achieved TI. Please see table below:

Table 52 TI Achievement by Genotype

Parameter	Statistic	Non- $\beta 0/\beta 0$ (N=29)	$\beta 0/\beta 0$ (N=12)
TI at any time	N evaluable subjects n (%)	28 25 (89)	8 7 (88)

Source: Reviewer calculations from ISE ADEF2 dataset

Reviewer Comment

Genotype had little impact on TI outcome in the Phase 3 studies, in contrast to Phase 1/2 studies with earlier, lower transduction % version of beti-cel given at lower doses.

Transfusion Reduction by Genotype

The median % change in annualized transfusion volume was 100% reduction in all genotypes in the Phase 3 studies, ranging from 31.3% to 100% reduction in subjects with non- $\beta 0/\beta 0$ genotype, and 3.4% to 100% reduction for subjects with

a $\beta 0/\beta 0$ genotype. Additionally, results of median change from Baseline in weighted average nadir Hb shows that most subjects' nadir Hb is higher post-treatment compared to Baseline. Transfusion reduction is not impacted by genotype in Phase 3 study subjects, as shown below

Table 53 Analysis of Transfusion Reduction From 6 Months through Last Follow-up by Genotype (Efficacy analysis population)

Parameter	Statistic	Non $\beta 0/\beta 0$	$\beta 0/\beta 0$
Change from baseline annualized RBC transfusion volume (%)	N	29	10
	Median	-100	-100
	(range)	-100, -31.3	-100, -3.4
	N		
Change from baseline in Weighted Average Nadir Hb(g/dL)	Median	1.4	1.2
	(range)	-1.8, 5.5	-0.4, 3.9

Source: Reviewer Calculations ISE ADEFF1 dataset

Non- $\beta 0/\beta 0$ genotype subjects had slightly higher median total unsupported Hb compared to subjects with a $\beta 0/\beta 0$ genotype, but ranges overlap, suggesting no meaningful difference. This is depicted below:

Table 54 Unsupported Total Hemoglobin by Genotype (EAP)

Parameter	Statistic	Non $\beta 0/\beta 0$	$\beta 0/\beta 0$
Unsupported total Hb at Month 6 (g/dL)	N	28	9
	Median	11.5	10.1
	(Range)	(8.4, 13.3)	(8.8, 13.2)

Source: Reviewer calculations, ADEFF2 dataset

Subpopulations based on Subject Age

Impact of age on achievement of TI

The adolescent subgroup achieved a higher % of TI than children under 12 years or adults over 18 years old. Similar TI outcomes are observed in subjects ≥ 18 years of age and in subjects < 18 years of age. The following table demonstrates TI by age subgroups.

Table 55 Age impact on Transfusion Independence (EAP population)

Parameter	Statistic	<12 Years	≥ 12 to < 18 Years	<18 years	≥ 18 years
TI-evaluable TI at anytime	N	12	10	22	14
	n (%)	10 (83)	10 (100)	20 (91)	12 (86)

Source: Reviewer Calculations, ISE ADEF2 dataset

Transfusion Reduction; effect of age.

As with transfusion independence, transfusion reduction was comparable between age subgroups. Please see the table below:

Table 56 Impact of Age on Transfusion Reduction and Change in Avg. Weighted Nadir Hg relative to Baseline

Parameter	Statistic	<12 Years	≥ 12 to < 18 Years	≥ 18 years
% Change from Baseline	N	14	11	14
Annualized	Median	-100	-100	-100
Volume	(Range)	-100, -3.4	-100, -100	-100, -32.4
Change from Baseline in Weighted Avg Nadir Hb (g/dL)	Median	0.94	2.15	2.46
	(Range)	-1.2, 1.7	-0.3, 4.6	-1.8, 5.5

Source: Reviewer Calculations, ISE ADEF1 and ADEF2 dataset

All age groups have a median percent reduction in annualized transfusion volume of 100% and maintained a median weighted average nadir Hb above baseline Hb levels. Young pediatric subjects <12 years appeared to have a smaller improvement in weighted average nadir Hb levels compared to older groups. This might be partly explained by the lower PB VCNs in this subgroup (median at Month 6 of 0.797 c/dg for subjects < 12 years of age [N = 12] compared to 1.399 c/dg for subjects ≥18 years of age [N = 14] and 1.910 c/dg for subjects ≥12 to < 18 years of age, which can lead to lower Hb^{AT87Q} levels.

Unsupported Total Hemoglobin, effect of age

Impact of age on the level of unsupported Total Hemoglobin at Month 6 is summarized below. Youngest subjects < 12 years old had lower unsupported hemoglobin levels compared with adolescents, and these were lower than adults. Similar trends are observed among the healthy population. I.e., differences observed between children and adults, and adult males and females, and children aged 2-12 years.

Table 57 Unsupported Total Hemoglobin at Month 6 by Age (EAP)

Parameter	Statistic	≥18 years	< 18 years	≥12 to < 18 years	<12 years
Unsupported total Hb at Month 6 (g/dL)	N	12	20	10	10
	Median	12.0	10.2	11.6	10.1
	(Range)	8.8, 13.3	9.3, 12.3	9.7, 12.3	9.3, 12.1

Source: Reviewer Calculations, ISE ADEF2 dataset

Transfusion Independence at Any Time by Sex

Rates of TI appear higher for males treated in Phase 3 studies compared to females, although the differences are small. Differences in total Hb values are anticipated considering that in healthy adults the total Hb differs between the sexes, males maintaining higher hemoglobin expression than females.

Table 58 Transfusion Independence at Any Time by Sex (EAP)

Parameter	Statistic	Male	Female
TI evaluable	N	19	17
TI at any time	n (%)	18 (95)	14 (82)

Source: Reviewer Calculations, ISE ADEF1 and ADEF2 dataset

Reviewer Comment

Differences in total Hb between male and female subjects as well as adult and pediatric subjects are expected based on differences in normal Hb levels between these subgroups. Subgroup analyses suggest that males, and adolescents may have slightly higher likelihood of TI achievement than other studies subgroups, although the differences appear small and not clinically significant. Moreover, these subpopulation analyses are on a limited sample size and not prospectively powered to make conclusions. Male subjects appeared to have better attainment of TI than females. Perhaps defining the weighted hemoglobin calculation on a single point Hb value (≥ 9 gm/dL) favors adult males achieving TI, because of impact of androgens on hemoglobin production (and expectation that more healthy males will have a Hb above 9 gm/dl than females).

Analysis of TI by Racial Subgroups

Proportion of subjects of different racial subgroups achieving transfusion independence is shown below:

Table 59 Transfusion Independence at Any Time by Race (EAP)

Parameter	Statistic	Asian	White	Other
TI Evaluable	N	15	18	2
TI at any time	n (%)	14 (93)	16 (89)	1 (50)

Source: Reviewer Calculations, ISE ADEF2 dataset

Reviewer Comment

The proportion of Asian subjects achieving TI appears slightly greater than Whites, but this difference does not appear clinically meaningful, likely reflecting chance variations, given the small numbers.

7.1.8 Persistence of Efficacy

Among subjects achieving TI, the time to onset of treatment effect may be inferred from time from beti-cel infusion to last RBC transfusion before TI onset. In general, the transfused RBCs would be expected to dissipate after 60-90 days. After that point, hemoglobin levels would be attributed to beti-cel as well as other endogenous Hb production. In the Phase 3 studies, the median time from beti-cel infusion to last RBC transfusion was 0.84 months, range 0 - 2.4 months.

Of the 36 TI evaluable subjects, 32 achieved TI (89%), and all maintained it to the Month 24 visit in the parent study, and then to the last follow up (longest duration

of TI in the Phase 3 studies was 39.4 months). Additionally, surrogate markers of efficacy such as presence of peripheral blood VCN and Hb^{AT87Q} support the clinical endpoint of TI. PB VCN levels increase in the first few months after beti-cel infusion, reaching a stable plateau maintained over time, suggesting durable persistence of the transduced long-term HSCs over time. Hb^{AT87Q} levels stabilized by approximately 6 months after beti-cel infusion and were maintained through last follow-up, with a median follow-up time of 27.2 months (range 4.1 - 48.2) for subjects treated in Phase 3 studies.

7.1.9 Product-Product Interactions

Not Applicable.

7.1.10 Additional Efficacy Issues/Analyses

None

7.1.11 Efficacy Conclusions

Beti-cel treatment resulted in a high proportion of treated subjects reaching TI. In Study HGB-207 Cohort 1, 14 of 15 TI-evaluable subjects (93%, 2-sided 95% CI of 68.1 to 99.8) achieved TI at any time post infusion; whereas six of seven TI-evaluable subjects (86%, 2-sided 95% CI of 42.1 to 99.6) achieved TI at any time post infusion in Cohort 2. Therefore, the success criterion for Cohort 1 has been met (cohort 2 had no determined success criterion). In Study HGB-212, 12 out of 14 TI-evaluable subjects (86%; 2-sided 95% CI of 57.2 to 98.2) achieved TI at any time post infusion. Therefore, the success criterion of study HGB-212 has also been met. Overall, 32/36 (89%) TI evaluable subjects achieved TI at any time. Once achieved, TI remained durable, with all subjects achieving TI at any time maintaining it to their last visit, the longest being 39.4 months.

Secondary endpoints provide supportive evidence of beti-cel effectiveness. Subjects who achieved TI in Phase 3 studies had median time from beti-cel infusion to last RBC transfusion of 0.84 (range 0-2.4) months. Subjects who achieved TI were able to maintain median weighted average Hb during TI of 11.5 g/dL, which equaled or exceeded baseline Hb levels.

In subjects having at least 6 months of follow-up, 35/39 (90%) achieved 100% reduction in annualized transfusion volume and frequency with similar or higher weighted average nadir Hb during the period from 6 months post beti-cel infusion through last follow-up compared with their annualized baseline pre-treatment transfusion requirements (32 of these had achieved TI and three subjects were not yet TI evaluable). Another four subjects with at least 6 months of follow-up after treatment had transfusion reduction < 100%: One subject had a ≥ 60% reduction in transfusions but with a weighted average nadir Hb 1.8 g/dL lower than the baseline, and 3 subjects had a <60% reduction in transfusions all with weighted average nadir Hb similar to or below baseline. Unsupported total Hb (without any RBC transfusions within prior 60 days) remained stable over time with median values between 10.9 and 12.7 g/dL from Month 6 to last visit among subjects of

Phase 3 studies. Primary and secondary efficacy analyses demonstrate the efficacy of beti-cel in the treatment of TDT across all subpopulations.

8. INTEGRATED OVERVIEW OF SAFETY

8.2 Safety Database

8.2.1 Studies/Clinical Trials Used to Evaluate Safety

The main safety data in support of beti-cel originated from the Phase 3 studies HGB-207 and HGB-212, as only these subjects received a product version comparable to the commercial version of the product. All subjects subsequently enrolled in the long term follow up study, Study LTF-303, to undergo observation for a total of 15 years following beti-cel infusion, therefore, pooled data from these studies were integrated to evaluate beti-cel safety. Data from Phase 1/2 study HGB-204 were reviewed as supportive.

This BLA also contained submission of safety data pertaining to insertional oncogenesis from IND 15905 evaluating lovo-cel for sickle cell disease (SCD) and BLA 125755, evaluating eli-cel for cerebral adrenoleukodystrophy (CALD). The same lentiviral vector and gene payload is used to manufacture beti-cel and lovo-cel, whereas eli-cel uses a related LVV. As such, data pertinent to insertional oncogenesis from these regulatory submissions were reviewed in support of the evaluation of this BLA for TDT.

8.2.2 Overall Exposure, Demographics of Pooled Safety Populations

Safety analysis was performed on data obtained from ongoing Studies HGB-207 and HGB-212, which were integrated along with data from long-term follow-up Study LTF-303 for subjects who enrolled in LTF-303 after completion of the parent studies. The TDT pool consisted of 43 subjects who initiated mobilization (ITT), 41 of whom received beti-cel (EAP). Two subjects in the TDT pool discontinued after 1 cycle of mobilization and were not treated with beti-cel: one due to pregnancy from HGB-207, and one due to withdrawal of consent in Study HGB-212. Of 41 subjects in the TDT pool who were treated with beti-cel, 29 completed the parent studies and enrolled in Study LTF-303.

Table 60 Disposition of the ITT population for the TDT pool is summarized

Parameter	Statistic	Phase 3 (N=43)
Subjects Mobilized	n (%)	43 (100)
Subjects infused with beti-cel	n (%)	41 (95.3)
Discontinued due to Withdrawn Consent	n (%)	1(2.4)
Discontinued due to Investigator Decision	n (%)	1(2.4)
Subjects enrolled into LFT 303	n (%)	29 (70.7)

Source: Reviewer calculations from ADSL ISS dataset

Exposure to beti-cel

In the 41 subjects who received beti-cel, the median dose was 9.4×10^6 CD34+ cells (range 5-42.1).

Duration of observation

Of the 41 beti-cel recipients, the median duration of follow up was 27.2 months (range 4.1 to 48.2) after infusion.

8.2.3 Categorization of Adverse Events

Adverse events (AEs) and serious adverse events (SAEs) were evaluated during clinic visits, hospitalizations, and follow-up visits per protocol-defined guidelines. Safety data are available for a total of 41 subjects who received beti-cel before the data cutoff of 09 March 2021. Adverse events were also assessed for the period from enrollment to the planned time of infusion to assess risks to subjects related to CD34+ cell collection or manufacturing issues.

Forty-three subjects started mobilization and apheresis. For the safety review, "Day 1" refers to the day of beti-cel infusion, and some AEs are presented as grouped terms. The applicant used preferred terms and grouped certain terms to present adverse reactions, but the grouping used was limited. For a more comprehensive evaluation of safety, the clinical reviewer's analysis included grouped AEs that represented the same or similar clinical conditions. Please refer to APPENDIX for the full list of FDA's grouped terms.

8.3 Caveats Introduced by Pooling of Data Across Studies/Clinical Trials

Both Phase 3 studies were designed with a parallel design and were single arm. The main caveat of grouping data from the Phase 3 studies is that they differed in the TDT genotype. Theoretically, $\beta 0/\beta 0$ subjects may have a more severe phenotype, but this is not expected to mechanistically affect safety.

8.4 Safety Results

8.4.1 Deaths

No deaths were experienced

8.4.2 Nonfatal Serious Adverse Events

In the TDT pool, 18 (42%) of the 43 subjects in the ITT population experienced 38 SAEs from start of mobilization to Month 24. Five (12%) experienced 6 SAEs prior to beti-cel administration. These events were attributed to either study procedure, apheresis, or other. Forty-one subjects started conditioning chemotherapy and underwent beti-cel infusion on D1, these were considered the efficacy analysis population. Fifteen subjects (37%) experienced 32 treatment-emergent SAEs through Month 24, shown in the table below:

Table 61 Treatment Emergent Serious Adverse Events in $\geq 3\%$, from Day 1 to Month 24. N=41.

Body System Organ Class / Preferred Term	% All Grade AEs	% Grade 3 and 4
Blood and lymphatic system disorders		
Febrile neutropenia	2 (4.9)	2 (4.9)
Neutropenia	2 (4.9)	2 (4.9)
Thrombocytopenia	3 (7.3)	3 (7.3)
Cardiac disorders		
Atrial fibrillation	1 (2.4)	1 (2.4)
Congestive heart failure	1 (2.4)	1 (2.4)
Gastrointestinal disorders		
Stomatitis	2 (4.9)	2 (4.9)
General disorders and administration site conditions		
Pyrexia	4 (9.8)	0
Hepatobiliary disorders		
Veno-occlusive liver disease	3 (7.3)	3 (7.3)

Source: Reviewer calculations from ISS ADAE dataset.

SAEs were mostly attributed by the investigator and Applicant to study interventions other than beti-cel. The investigator ascribed the SAE of grade 3 thrombocytopenia from Day 114 to Day 163 as possibly due to beti-cel due to the delayed nature of platelet recovery (Subject (b) (6)). This reviewer agrees with the attribution of this SAE to beti-cel.

However, this reviewer determined that the SAE of epistaxis experienced by Subject (b) (6) on Day 69 resolving on Day 70 is more appropriately considered as a primary event of severe thrombocytopenia which was complicated by the secondary event of epistaxis on D69 that required hospitalization. This reviewer attributes this SAE to beti-cel rather than busulfan because it occurred in context of delayed platelet engraftment, a known risk of beti-cel. It is further discussed in section below on beti-cel related AEs.

Two subjects experienced two SAEs of bleeding: one was the epistaxis described above in Subject (b) (6), and another was in Subject (b) (6), an 18-year-old who experienced a grade 1 contusion on Day 505 to 522 after getting into an altercation (with unremarkable platelet counts). In addition, Subject (b) (6) a 20-year-old experienced an SAE of hypotension on Day 11 to Day 12, secondary to a bleeding event of epistaxis in context of severe thrombocytopenia on Day 9 to Day 23 (considered by investigator as not serious). These SAEs were ascribed to busulfan conditioning, which is reasonable.

SAEs reported in more than two subjects included the following:

- Three subjects experienced 3 SAEs of Grade 3 or Grade 4 VOD that were considered related to busulfan conditioning.
- Four subjects experienced 4 SAEs of Grade 1 or Grade 2 Pyrexia, all requiring hospitalization: one from Day 37 to Day 44 (attributed to

- conditioning), one from Day 44 to 46 (attributed to viral syndrome), one from Day 53 to Day 58 (unknown attribution), and one from Day 60 - 62 (attributed to viral rhino-pharyngitis), and all resolved.
- Two subjects experienced 2 SAEs of Grade 4 Neutropenia prior to NE and were attributed to busulfan conditioning.
 - Three subjects experienced 3 SAEs of Grade 3 or Grade 4 Thrombocytopenia. Two occurred prior to neutrophil engraftment and were attributed to busulfan conditioning. One was reported as related to treatment with beti-cel by the Investigator (Subject (b) (6)).

Reviewer Comment

Serious adverse events following beti-cel in the integrated analysis suggest a similar pattern of events as noted in the individual phase 3 studies, specifically that most serious events are related to myeloablative chemotherapy. This includes the hepatic veno occlusive disease, which is a known complication of busulfan, and did occur despite prophylaxis introduced with Phase 3 study protocols (2 SAEs of VOD in HGB-207, none in HGB-212). Events such as pyrexia and neutropenia are also frequent events after myeloablative conditioning chemotherapy. Two SAEs of thrombocytopenia in the context of slow platelet recovery were noted. Both recovered, and one was not associated with bleeding (did have bruising reported).

8.4.3 Study Dropouts/Discontinuations

No subjects with TDT dropped out after being dosed with beti-cel. Two subjects discontinued after starting mobilization, one (HGB-207) due to pregnancy, and one (HGB-212) due to withdrawal of consent.

8.4.4 Common Adverse Events

Adverse Events between start of Mobilization to Before Start of Conditioning

The studies' safety populations were comprised of the ITT population who started any of the planned study interventions starting with mobilization. Events reported during period of mobilization- apheresis (M to <C) are typical of the known safety profiles of those procedures and products. The following table demonstrates AEs experienced after start of mobilization and apheresis, but before conditioning chemotherapy, and reflect toxicities related to plerixafor, G-CSF, and line insertion.

Table 62 AEs Reported among $\geq 10\%$ of subjects from Mobilization to <Conditioning. (N= 43)

Body System Organ Class	Preferred Term	Subjects with AE n (%)
Blood and lymphatic system disorders	Thrombocytopenia	9 (21)
Gastrointestinal disorders	Nausea	7 (16)
	Vomiting	6 (14)
Injury, poisoning and procedural complications	Procedural pain	13 (30)
Metabolism and nutrition disorders	Hypocalcemia	12 (28)
Musculoskeletal and connective tissue disorders	Bone pain	9 (21)
Nervous system disorders	Headache	7 (16)

Source: Reviewer calculation ISS ADAE dataset

Adverse Events between Day 1 and Month 24

The bulk of AEs experienced by subjects occurred after start of conditioning, and include gastrointestinal toxicities such as emesis and mucositis, hepatobiliary (including veno-occlusive disease) and myelosuppression (cytopenias). While the majority of these events are expected complications of myeloablative conditioning with busulfan, it is impossible to completely extricate contribution of beti-cel from the AEs reported, as beti-cel administration follows 48h after completion of conditioning chemotherapy, moreover, beti-cel therapy requires that conditioning chemotherapy be administered.

The observed types and rates of AEs seen from conditioning to before neutrophil engraftment (C to <NE), are reasonably expected from busulfan conditioning. While febrile neutropenia was common, severe grade infections were not. There were 4 AEs of sepsis (6.8%), one of which being grade 3 and one grade 4. The table below summarizes non-laboratory AEs reported from D1 to Month 24.

Table 63 Non-Laboratory Treatment Emergent Adverse Events in $\geq 10\%$ subjects between D1 and Month 24

Body System Organ Class AE	% Any Grade	% Grade 3 or Higher
Blood and lymphatic system disorders		
Febrile neutropenia	51	51
Gastrointestinal disorders		
Mucositis#	95	63
Vomiting	49	0
Abdominal pain	39	2
Diarrhea	27	0
Nausea	25	2
Constipation	24	0
Dyspepsia	10	5
Gingival bleeding	10	2
General disorders & administration site conditions		
Pyrexia	48	12
Fatigue	12	0
Hepatobiliary disorders		
Veno occlusive liver disease	10	7
Infections and infestations		
Viral infection	17	2
Upper respiratory tract infections#	15	0
Nasopharyngitis	12	0
Sepsis	10	10
Injury, poisoning and procedural complications		
Procedural pain	15	0
Transfusion reaction	15	0
Metabolism and nutrition disorders		
Decreased appetite	24	15
Musculoskeletal and connective tissue disorders		
Musculoskeletal pain#	37	0
Nervous system disorders		
Headache	29	0
Respiratory, thoracic and mediastinal disorders		
Epistaxis	42	20
Cough	34	0
Oropharyngeal pain#	15	0
Dyspnea	12	0
Hypoxia	12	7
Rhinitis	12	0
Skin and subcutaneous tissue disorders		
Alopecia	44	0
Rash	27	0
Pigmentation disorder	24	0
Pruritus	22	0
Vascular disorders		
Hypertension	10	0

Source: FDA Analysis ISS ADAE3.xpt AE: Adverse event, SOC: System organ class, PT: preferred term. * Includes grouped terms as detailed in APPENDIX; # Encompasses more than one system organ class

The integrated summary of safety includes data collected from the long term follow up study LTF 303, which enrolled subjects from the parent studies. Subjects experienced very few additional AEs beyond 24 months following beti-cel treatment. In fact, analysis of non-laboratory TEAEs occurring in $\geq 10\%$ of Subjects between Day 1 to last follow up identified only one additional subject with an event of procedural pain.

Most AEs resolved before the last follow up. The most common AEs which did not resolve reported in $\geq 5\%$ of subjects include alopecia, anemia, thrombocytopenia, and ALT increase, neutropenia, procedural pain, and skin hypopigmentation.

Clinical Test Results

Laboratory based AEs were evaluated separately by using shift tables, utilizing ISS ADLB dataset. As mentioned above, conditioning chemotherapy with busulfan is expected to lead to frequent laboratory toxicities but is an integral and necessary step prior to beti-cel administration. Therefore, laboratory AEs were analyzed from start of cytoablation, and compared with baseline rates of AEs. Like non-laboratory AEs, virtually all laboratory AEs occurred during the 24-month observation period of the parent studies. The following table summarizes laboratory-based abnormalities in $\geq 5\%$ subjects from D1 to month 24.

Table 64 Laboratory based abnormalities in $\geq 10\%$ subjects Laboratory Based Abnormalities. D1 to Month 24. N= 41

Laboratory Based Abnormality	All grades n (%)	Grade 3-4 n (%)
neutropenia	41 (100)	41 (100)
thrombocytopenia	41 (100)	41 (100)
leukopenia	41 (100)	41 (100)
anemia	40 (98)	39 (95)
lymphopenia	39 (95)	25 (61)
hypocalcemia	37 (90)	1 (2)
ALT Increased	35 (85)	10 (24)
AST Increased	30 (73)	1 (2)
hypokalemia	28 (68)	5 (12)
hypophosphatemia	28 (68)	8 (20)
hyponatremia	24 (59)	4 (10)
ALP Increased	16 (39)	0
hyperbilirubinemia	14 (34)	4 (10)
hypercalcemia	10 (24)	1 (2)
hyperkalemia	8 (20)	0
hyperglycemia	5 (14)	5 (14)
lymphocytosis	5 (12)	1 (2)

Abbreviations: ALP: alkaline phosphatase; ALT: alanine aminotransferase; AST aspartate aminotransferase. Note that denominator was 36 for Glucose due to missing data

Reviewer Comment

- *Laboratory data were used to generate incidence of laboratory- based AEs since this is more accurate as opposed to using the adverse event dataset.*
- *A “lab-shift” analysis was carried out wherein baseline laboratory abnormalities that worsened following treatment were recognized i.e., shift of a laboratory grade from a lower to higher grade.*
- *Cytopenias of all grades were the most common laboratory abnormalities as expected and reflect toxicity of the entire investigational protocol including myeloablative conditioning chemotherapy.*

The overall pattern of reported laboratory AEs are consistent with expected types and rates of events from mobilization and myeloablative conditioning chemotherapy. Most AEs resolved, except infrequent AEs including mild cytopenias, alopecia, liver enzyme abnormality and skin hypopigmentation. The cytopenias are further discussed below in 8.4.8, Adverse Events of Special Interest section.

8.4.6 Systemic Adverse Events

Please refer to prior discussions of AEs above

8.4.7 Local Reactogenicity

Not applicable

8.4.8 Adverse Events of Special Interest

Due to the mechanism of action of beti-cel, and the potential for LVV-related insertional oncogenesis, the potential for alteration of HPSC gene expression, as well as necessary myeloablative conditioning, the FDA considered the following as adverse events as of special interest: cytopenias and hematopoietic cell recovery, bleeding events, immune-related events, infection events, and insertional oncogenesis.

Cytopenias and Hematopoietic Cell Recovery

Thrombocytopenia

Platelet engraftment (PE) was delayed (relative to published outcomes from allogeneic HSCT for TDT), with a median time to PE of 46 days (range 20-94). Beyond the threshold of $20 \times 10^9/\text{L}$ platelets required to meet PE definition; platelet recovery was slow. Of the Phase 3 subjects, Six (15%) did not achieve an unsupported platelet count $\geq 50 \times 10^9/\text{L}$ until \geq Day 100, that is, they continued to experience grade ≥ 3 thrombocytopenia on or after Day 100.

Neutropenia

Two (5%) subjects remained dependent on G-CSF beyond Day 42, one of them through Day 77. G-CSF discontinuation was accompanied by transient decreases in neutrophil count to < 500 cells/μL after Day 42 in six (15%) subjects.

Bleeding

One event of interest was identified as it was a serious bleeding event in the setting of delayed PE, an identified risk of beti-cel: Subject (b) (6), a 5-year-old female, was hospitalized on Day 69 due to Grade 3 Epistaxis in the setting of delayed PE. The SAE resolved the next day following transfusions with pRBCs and platelets. She achieved PE on day 80). Following PE, she has not experienced any further bleeding events even though subsequent recovery of platelets remained slow. This reviewer attributed the SAE to beti-cel due to the delayed time course.

Immune related events

Subject (b) (6), a 21-year-old female described above due to thrombocytopenia, experienced a nonserious Grade 3 Autoimmune Disorder (updated to Immune Thrombocytopenia after data cut-off) approximately 2 years after beti-cel treatment, in the setting of ongoing Grade 1 Thrombocytopenia.) A platelet antibody work-up was performed and was positive for cell-bound anti-glycoprotein IIb/IIIa antibodies. While this event was assessed by the investigator as possibly related to beti-cel; the Applicant concluded the event was unlikely related to beti-cel and instead concluded this finding was likely reflective of the phenomenon of transplant- associated immune thrombocytopenia, rather than beti-cel related immunogenicity.

Reviewer Comment

This subject developed persistent thrombocytopenia which might be immune mediated, as supported by presence of glycoprotein IIb/IIIa antibodies. The event was not associated with symptoms and did not require pharmacologic intervention. This reviewer believes that this likely represents transplant-associated immune thrombocytopenia, rather than immunogenicity due to beti-cel. Transplant associated immune thrombocytopenia is more commonly reported with allogeneic transplant but has been described with autologous transplant.

Infections

Thirty-three subjects with TDT experienced 92 events of infections. Five subjects had 7 treatment emergent serious adverse events (TESAEs) in the Infections and infestations system organ class (SOC). These SAEs predominantly included typical viral and bacterial infections observed in the peri-transplant period. Two of three SAEs of sepsis, occurred within 14 days of beti-cel during the typical nadir of neutrophil levels post myeloablation, whereas the third SAE of sepsis was related to a defective central line starting on Day 149. Of the sepsis SAEs occurring early, one was reported Day 9 and resolved Day 10. The last sepsis SAE was reported on Day 13 and resolved Day 16. Both resolved with antibiotics. The

one associated with central line resolved on Day 162 with removal of line and antibiotics.

Reviewer Comment

The incidence and pattern of infection related AEs are typical of myeloablation required for beti-cel administration and do not raise safety concerns for beti-cel.

8.5 Additional Safety Evaluations

Reviewer Comment

The review team conducted an analysis by of safety by age, and concludes that overall, there are no clinically meaningful differences between subjects < 18 years old vs. ≥ 18 with respect to safety signal attributed to beti-cel. Because younger subjects tended to have later engraftment, risks of infections (due to neutropenia) and bleeding (due to thrombocytopenia), might be expected to differ. Analysis was performed using SMQ analysis and found that despite longer times to platelet and neutrophil engraftment in younger vs. older subgroups, rates of bleeding and infectious events was comparable.

Supportive Safety Findings from HGB-204 (Phase 1/2 study)

Reviewer Comment

Although HGB-204 administered a lower dose on an earlier generation beti-cel with substantially lower % transduction and beti-cel VCN, the Agency felt that review of safety data from this study would be supportive and thus analysis would be useful. The review team pooled the safety data from the two Phase 3 studies and Phase 1/2 HGB-204 study. Overall, analysis of the safety data from HGB-204 suggests a similar safety signal to what was observed with the Phase 3 studies, with the majority of the adverse events likely related to conditioning chemotherapy. HGB-204 data also demonstrated two cases of oligoclonal LVV integrations on ISA. These are further described in sections 6.3 and 9.2.

8.5.1 Dose Dependency for Adverse Events

Not applicable

8.5.2 Time Dependency for Adverse Events

Not applicable

8.5.3 Product-Demographic Interactions

Not applicable

8.5.4 Product-Disease Interactions

Not applicable

8.5.5 Product-Product Interactions

Not applicable

8.5.6 Human Carcinogenicity

Not applicable

8.5.7 Overdose, Drug Abuse Potential, Withdrawal, and Rebound

Not applicable

8.5.8 Immunogenicity (Safety)

Not applicable

8.5.9 Person-to-Person Transmission, Shedding

Not applicable

8.6 Safety Conclusions

Integration of safety data from the Phase 3 studies does not suggest any new safety signals compared to the analysis of the individual study datasets. There have been no deaths reported during the clinical development of beti-cel. Two beti-cel-related SAE of Grade 3 Thrombocytopenias were reported. The adverse reactions of beti-cel therapy included largely hematologic and gastrointestinal events related to the necessary to myeloablative conditioning and resolved. All subjects successfully reached neutrophil engraftment and platelet engraftment (PE), although PE was delayed as compared to the allo-HSCT literature. Delayed PE is an important identified risk of treatment with beti-cel, though not associated with a clinically meaningful increase in bleeding events. After achievement of PE, most subjects did not return to their baseline platelet counts. However, most returned into the normal range and all subjects' platelet counts settled into a stable trajectory, without evidence of progressive decrease in platelet counts. No cases of LVV-derived RCL or insertional oncogenesis (potential risks of LVV-base therapy) were reported.

9. ADDITIONAL CLINICAL ISSUES

9.1 Special Populations

9.1.1 Human Reproduction and Pregnancy Data

No animal studies of reproduction or developmental toxicity have been performed, and beti-cel has not been studied in pregnant women.

Reviewer Comment

Effective contraception was required for clinical trial participation of beti-cel, and this will be advised in the label.

9.1.2 Use During Lactation

N/A

9.2 Aspect(s) of the Clinical Evaluation Not Previously Covered

Bone Marrow Morphology Abnormalities in subjects with TDT

Dyserythropoiesis and features related to abnormal iron metabolism in marrow morphology, are known complications of TDT. Consequently, subjects underwent serial (baseline, Month 12 and Month 24) marrow sampling to assess the impact of beti-cel on dyserythropoiesis of thalassemia. Unfortunately, the bone marrow data are limited, as the studies were not designed to evaluate cytopenias, but to evaluate the evolution of dyserythropoiesis changes between baseline and Month 24. Pathologists were not free to order ancillary molecular, cytogenetic or other studies on the marrow samples. Nonetheless, the bone marrow assessments that were conducted did identify several subjects with abnormalities such as ring sideroblasts or dysplastic megakaryocytes.

In Study HGB-207, one subject had emergent abnormal (monolobated) megakaryocytes at Month 12 in concert with borderline thrombocytopenia ($158 \times 10^9/L$ platelets), which were reported as concerning for MDS by an independent pathologist. However, the subject declined subsequent biopsies, so there is no follow up regarding this patient. The subject's platelet counts slowly increased. Four other post-beti-cel marrow samples contained post beti-cel ring sideroblasts, but baseline samples lacked iron stains, making it challenging to attribute the finding to thalassemia or beti-cel treatment. Similarly, in Study HGB-212, although ring sideroblasts and dysmegakaryopoietic changes were observed in a small percentage of subjects, it is challenging to attribute this emergence of ring sideroblasts to beti-cel treatment.

Of note, dysmegakaryopoietic changes were noted in a few subjects following beti-cel administration. The marrow data were limited such that it was not feasible to determine whether some subjects experience emerging or worsening dysplastic features of megakaryocytes. Bone marrow samples were reviewed by outside pathologists who concluded there was no morphologic evidence of developing MDS in the post-beti-cel samples; rather, the abnormalities observed were ascribed to chronic stress erythropoiesis due to underlying thalassemia.

Reviewer Comment

While the FDA cannot conclude if the morphologic changes are a result of chronic stress erythropoiesis, the abnormalities observed might confound evaluation of bone marrow morphology for emergent pathology.

Risk of Insertional Oncogenesis

While no subjects treated with beti-cel were reported to have clonal predominance or oligoclonality prior to time of data lock nor in the 3 months safety update period, the BLA included submission of safety data from IND 015905 where the Applicant's LVV-based beti-cel manufactured with the same LVV as beti-cel, was evaluated in subjects with sickle cell disease (SCD), and also in BLA 125755, where the applicant treated subjects with cerebral adrenal leukodystrophy with a related LVV-based product.

LVV integration into target cell genes during transduction is associated with potential risk of disrupting expression of nearby genes. Such insertional mutagenesis can lead to oncogenesis after the development of a predominant cell clone. Beti-cel recipients are monitored regularly for this complication with integration site analysis (ISA) of blood cells.

While the ISA algorithm used in the studies is described in the Appendix, the Applicant proposed a new ISA algorithm after the time of data lock and the 3-month safety follow up. Please refer to the Appendix for explanation of the ISA algorithms. The proposed new algorithm introduces the concept of oligoclonality, defined as a clone reaching 10% relative frequency. Persistent oligoclonality, which remains positive 3 months later, would be reported to regulatory agencies.

Three subjects meet the oligoclonality definition based on the revised ISA. Of these, two subjects from Study HGB-204 have demonstrated stable oligoclonality for several years without development of a predominant clone or malignancy, but had cytopenias and LVV integration into proto-oncogenes (please also see section 6.3):

- Subject (b) (6) continues to have mild thrombocytopenia and has not reached platelet count of $\geq 100 \times 10^9/L$ as of Day 737 after beti-cel infusion. This subject appears to have integration sites that may be within the same clone, including the proto-oncogenes XPO7 and CBFB. The most recent IS-specific VCN was 0425 c/dg in XPO7 and 0461 c/dg in CBFB
- Subject (b) (6) remains mildly thrombocytopenic and did not reach platelet count $\geq 100 \times 10^9/L$ until after Day 501; this subject has integration sites that may be within the same clone including proto-oncogene BCR. For BCR, most recently measured integration site-specific VCN was 0.1245 c/dg.

- Subject (b) (6) had oligoclonality identified with a single IS of $\geq 10\%$ RelFreq in gene *MAP4K2* but achieved unremarkable platelet reconstitution. (Data for this subject were submitted after 90-day safety follow up, in a submission explaining the newly proposed ISA algorithm).

Reviewer Comment

One subject with TDT showed a restricted pattern of IS in the CBFB and XPO7 clones between Month 6 through Month 60 and on further evaluation with qPCR, the clonal contribution reached as high as approximately 10% (at Month 24) and dropped to $< 5\%$ at month 60. Another TDT subject had an IS which persisted in the Top 10 most frequent IS list between Month 24 and Month 60. This pattern is unusual, and the IS similar clonal contributions made it possible that all these IS may be present in a single clone. The Applicant confirmed that based on (b)(4) none of these IS represented a predominant clone.

After time of data lock and after 3-month safety follow up, the Applicant submitted to this BLA a revised integration site analysis (ISA) algorithm. This would reclassify both of the above HGB-204 subjects as having met the persistent oligoclonality criterion of $\geq 10\%$ relative frequency of integrations in a clone determined on two occasions at least three months apart.

Furthermore, XPO7, CBFB and BCR genes are protooncogenes and integration into these sites is considered potentially more worrisome than integrations into less sensitive loci. However, under prolonged ISA monitoring, these two subjects' IS patterns appear stable, and even improved over time (i.e., decrease in IS relative frequency). Moreover, the two subjects' hematologic parameters also remain stable, arguing against any pathologic process related to these ISA results. These Upon evaluation of these findings, the review team concludes that the safety implications of these findings can be addressed by a postmarketing safety study, and product label.

Hematologic malignancies following treatment with similar or related LVV-based products for other diseases

Sickle Cell Disease (SCD)

The BLA contained data from IND 15905, which administered a LVV-based product called lovo-cel to subjects with SCD. Lovo-cel is an autologous CD34+ product that contains HSCs transduced with BB305, a LVV encoding the β^{A-T87Q} globin gene which has anti-sickling properties while conserving the function of β -globin. Lovo-cel and beti-cel share the same LVV, gene cassette including target gene and promoter, and only have modest manufacturing differences. Myeloablative conditioning is also required before lovo-cel infusion. As of the data cut off, 49 subjects have received lovo-cel in clinical studies for SCD, and cases of MDS and AML have been reported after administration of lovo-cel, though a

causal role of LVV has not been conclusively demonstrated in the development of these malignancies.

Two subjects with SCD were diagnosed with AML, and one of these was found to have a predominant clone in the *VAMP4* gene, which encodes vesicle-associated membrane protein 4. The causal role of this integration is not clear, as the subject's leukemic blasts also contained known driver mutations for AML.

Notably, integrations into *VAMP4* have been detected in 31 of 55 subjects with TDT (56%), with 59 unique *VAMP4* IS detected in beti-cel treated subjects. The highest maximum frequency detected was 0.217%.

Reviewer Comment

The presence of a predominant clone with VAMP4 gene integration in one of the 49 lovo-cel recipients is of concern, although its role in the development of that subject's AML is unknown. While low relative frequency integrations into VAMP4 gene are common in beti-cel recipients, they do not appear associated with delayed platelet engraftment, and the role of VAMP4 integrations in TDT subjects is unclear.

Other hematologic malignancy concerns following lovo-cel treatment for SCD

Besides the case of AML with predominant clone containing *VAMP4* gene integrations in SCD subjects, there were two subjects with bone marrow findings concerning for MDS.

- A 14-year-old male subject with 10%-20% erythroid dysplasia in the bone marrow and persistent Trisomy 8 and Tetrasomy 8 on (b)(4) at months 12 and 14 following lovo-cel infusion. Dysplastic bone marrow findings included binucleations, nuclear budding, vacuolated pro-normoblasts, basophilic stippling, and nuclear-cytoplasmic asynchrony. Findings confounded by B12 deficiency, but the trisomy 8 and bone marrow erythroid dysplasia persisted despite B12 supplementation at 2 years following lovo-cel infusion.
- 20-year-old female with SCD with transfusion-dependent anemia (previously with only periodic transfusions and normal bone marrow at baseline), 10%-20% erythroid dysplasia on bone marrow, and trisomy- and tetrasomy 8 (by (b)(4) 8 months following lovo-cel infusion. Repeat (b)(4) studies negative although erythroid dysplasia and need for ongoing transfusions for severe anemia persists. Subject also noted to have ATM germline mutation.

Reviewer Comment

1. *Although the applicant suggested the etiology of anemia in these two subjects is due to "stress erythropoiesis and 2-gene alpha deletion*

worsening non-alpha to alpha globin gene imbalance, the review team disagrees for the following reasons:

- i) Both subjects were expected to have much higher Hb's based on their VCN post-GT. 20-year-old subject only became newly transfusion dependent after gene therapy.
 - ii) The hypothesis that 2-gene-alpha globin deletion leads to increased hemolysis remains unproven. Coinheritance of 2-gene alpha thalassemia is asymptomatic in patients with SCD. Since expression of β A-T87Q -globin and sickle hemoglobin are competitive, ratio of non-alpha to alpha globin post gene therapy is not expected to change. A report from Applicant of expert pathology review concluded that the theory of stress erythropoiesis causing anemia is "admittedly speculative."
 - iii) 20-year-old subject also carries the ATM germline mutation predisposing her to malignancy. Her marrow was normal at baseline, developing persistent dysplasia only post lovo-cel.
2. Malignancy is a multi-step process, underpinned by germline, somatic, and environmental drivers, and differences between SCD and TDT at each step may cause differing outcomes:
- i. SCD and TDT are caused by different germline mutations, and the subsequent inflammatory milieu of the bone marrow microenvironment is likely different between SCD and TDT patients. This might be the reason behind the uniquely elevated cancer risk in patients with SCD suggested by registry data⁸.
 - ii. Patients with SCD and those with TDT are treated differently prior to autologous transplant (e.g., most SCD patients may have been treated with hydroxyurea prior to transplant, unlike TDT patients)
 - iii. Manufacturing differences between beti-cel and lovo-cel may affect risk of malignancy

Considering the above differences between subjects with SCD vs. TDT, the review team concludes that these differences may contribute to any differences in risk level of hematologic abnormalities observed. However, considering the insertional oncogenesis risk inherent to lentiviral vectors, a PMR safety study will be required.

Cerebral Adrenoleukodystrophy (CALD)

The BLA also contains data from BLA 125755 related to safety and insertional oncogenesis following treatment of subjects with eli-cel for CALD. Eli-cel is a related LVV-based product for CALD, a rare, X-linked neurodegenerative disease caused by mutations in the ATP-binding cassette (ABC), subfamily D, member 1

8 Brunson A, Keegan THM, Bang H, Mahajan A, Paulukonis S, Wun T. Increased risk of leukemia among sickle cell disease patients in California. *Blood*. 2017 Sep 28;130(13):1597-1599. doi: 10.1182/blood-2017-05-783233. Epub 2017 Aug 22. PMID: 28830890; PMCID: PMC5620417.

(*ABCD1*) gene that encodes the transporter protein adrenoleukodystrophy protein (ALDP). Deficiency of ALDP causes accumulation of very long-chain fatty acids in the central nervous system, Leydig cell of the testes, and the adrenal cortex. Boys with early CALD are treated with allogeneic hematopoietic stem cell transplant. An unmet medical need exists for therapeutic options because of insufficient suitable donors. Eli-cel is comprised of autologous CD34+ HSCs transduced with a LVV containing the *ABCD1* gene.

Of 67 subjects administered eli-cel in clinical studies, three have been diagnosed with MDS. Two of these were diagnosed within ≤ 24 months of eli-cel treatment and contain a dominant clone with LVV integration into the MDS1 and EVI1 complex locus (*MECOM*) oncogene with overexpression of *EVI1*.

Given the findings of delayed thrombocytopenia after beti-cel therapy and the oligoclonality reported in a couple subjects, as well as the complexity of lentiviral integrations and potential impact or contribution to the development of malignancy, the above findings were presented to an advisory committee of pertinent experts in order to gain further insight and to assist in determination of benefit-risk of beti-cel. The Advisory committee hearing was held June 9-10, 2022, and the members voted overwhelmingly in favor of benefit exceeding the risk.

Reviewer comment

The review team carefully considered the safety results for the clinical trials with beti-cel, the findings in regulatory submissions for similar/related products for SCD and CALD, and the Advisory Committee's conclusions and recommendations regarding risk of insertional oncogenesis with beti-cel.

Similar to the AC conclusions, the clinical review team does not believe that the hematologic malignancies observed in SCD and CALD subjects who received a similar or related LVV-based product are significantly informative regarding the risk of hematologic malignancy in patients with β -thalassemia who received ZYNTEGLO as:

- the pathophysiologic characteristics of SCD may predispose patients to increased risk of hematologic malignancy
- the Applicant's product for the treatment of CALD is considered to be distinct from ZYNTEGLO.

Nonetheless, the review team is concerned regarding potential risk of insertional oncogenesis following treatment with beti-cel, and is recommending a safety PMR study.

10. CONCLUSIONS

EFFICACY

Efficacy of beti-cel is based on prevalence of transfusion independence demonstrated in multicenter, open label, single arm clinical trials in adults with TDT who required regular red cell transfusion. A total of 41 subjects were infused with beti-cel in Phase 3 trials, and 32 of 36 (89%) TI evaluable subjects achieved TI at any time. TI remained durable, with all subjects who achieved TI maintaining it to their last visit, the longest being 39 months. These efficacy outcomes were generally consistent across subgroups with respect to genotype and age. The basis of FDA's conclusion of substantial evidence of effectiveness is the magnitude of benefit driven primarily by the rate of durable transfusion independence.

In summary, Studies HGB-207 and HGB-212 represent adequate and well-controlled trials that provide substantial evidence of effectiveness of beti-cel in adult and pediatric patients with transfusion-dependent thalassemia. The results support a traditional approval for beti-cel.

SAFETY

The safety profile of beti-cel therapy entails the rigors of myeloablation with frequent cytopenias and gastrointestinal symptoms, which resolved. Delayed platelet engraftment with thrombocytopenia is a notable safety finding, although with limited clinical sequelae. Due to insertional oncogenesis risk inherent to lentiviral vectors, a PMR safety study will be required.

In summary, Studies HGB-207 and HGB-212 represent two adequate and well controlled studies that provided substantial evidence of effectiveness and safety.

11. RISK-BENEFIT CONSIDERATIONS AND RECOMMENDATIONS

11.1 Risk-Benefit Considerations

The following table summarizes the risk/benefit considerations for beti-cel for the treatment of children and adults with β thalassemia who require regular red blood cell transfusions.

Table 65 Benefit Risk

Decision Factor	Evidence and Uncertainties	Conclusions and Reasons
Analysis of Condition	<ul style="list-style-type: none"> Transfusion dependent β-thalassemia (TDT) is a hereditary hemoglobinopathy characterized by absent or reduced β-globin expression, causing α β-globin chain imbalance with unpaired α-globin build-up leading to premature death of RBCs precursors in the marrow. Nearly 90 million people (approximately 1.5% of the global population) are carriers of β-thalassemia mutations, leading to the birth of approximately 60,000 symptomatic patients annually. The ineffective erythropoiesis in β thalassemia leads to anemia and subsequent complications such as hemolysis, hypercoagulability, transfusion-related iron toxicity including, heart disease, endocrinopathies and cirrhosis. 	<ul style="list-style-type: none"> Originating in southeast Asia and Mediterranean basin, due to immigration, β thalassemia impacts the US population and is associated with substantial morbidity and mortality. TDT leads to shortened survival largely due to iron overload in major organs, involving the heart, liver, and endocrine glands.
Unmet Medical Need	<ul style="list-style-type: none"> The sole FDA approved drug to treat anemia secondary to transfusion dependent β thalassemia has modest clinical benefit and carries risks. Patients with TDT require red blood cell transfusions to maintain an acceptable hemoglobin range and iron chelation therapy due to transfusion-related iron overload. Blood transfusions are associated with transmission of blood-borne pathogens, transfusion reactions, alloimmunization, and transfusion related iron overload. Hematopoietic stem cell transplantation from a matched family donor is a potentially curative treatment option for pediatric patients but this modality is limited by lack of appropriate donors and potential risks of stem cell transplantation, including graft vs. host disease. 	<ul style="list-style-type: none"> Only a small minority of TDT patients has an appropriate HSPC transplant donor. Consequently, an unmet medical need exists for therapeutic options because of insufficient suitable donors.
Clinical Benefit	<ul style="list-style-type: none"> Beti-cel treatment demonstrated clinically meaningful transfusion independence in most subjects on Studies HGB-207 and HGB-212, and reductions in RBC transfusion burden. These results were durable and robust as demonstrated by consistency across all subgroups, including $\beta 0/\beta 0$ genotypes 	<ul style="list-style-type: none"> Beti-cel treatment resulted in transfusion independence and decreased RBC transfusion requirements in subjects with TDT
Risk	<ul style="list-style-type: none"> The most common adverse reactions in subjects with TDT treated with beti-cel were: mucositis, febrile neutropenia, vomiting, pyrexia, alopecia, epistaxis, abdominal pain, musculoskeletal pain, cough, headache, diarrhea, rash, constipation, nausea, decreased appetite, pigmentation disorder, and pruritus. Serious adverse reactions were reported 37% of patients. The most common serious adverse reactions (> 3%) were fever, thrombocytopenia, liver veno-occlusive disease, febrile neutropenia, neutropenia, stomatitis. 	<ul style="list-style-type: none"> The overall risk for the proposed population appears acceptable. Long-term safety will be an important consideration and will be part of a post-marketing requirement/commitment. While no evidence of insertional oncogenesis or myelodysplastic syndrome (MDS) was noted with beti-cel, the concern exists because of reports of these hematologic malignancies in studies of other LVV products. To minimize risks, labeling will include warnings and precautions for delayed platelet engraftment and prolonged thrombocytopenia. There were no death or discontinuation due to an adverse reaction reported.
Risk Management	<ul style="list-style-type: none"> Warnings and instructions in the package insert, the PMR study, and the pharmacovigilance plan would be adequate to manage the risks 	<ul style="list-style-type: none"> If beti-cel were approved for patients with TDT, the PMR study and routine measures, such as the package insert and pharmacovigilance plan, would be adequate to manage the risks

11.2 Risk-Benefit Summary and Assessment

TDT is a severe disease with life-long dependence on regular RBC transfusions for survival, marked by cardiac, liver, and endocrine iron toxicity and shortened lifespan. The only potential curative treatment option is allogeneic HSCT but this can be complicated by GVHD and most patients lack appropriate HSC donors. Among 41 phase 3 study subjects treated with beti-cel, 36 are TI evaluable and of these, 32 (89%) have achieved TI, which represents a substantial clinical benefit. This clinically relevant endpoint is further supported by secondary endpoints and pharmacodynamic parameters.

Submitted data provide evidence of benefit with beti-cel treatment. The safety profile indicates delayed platelet engraftment, and prolonged thrombocytopenia. Although there exists a potential risk of insertional oncogenesis, there are no identified cases thus far.

The benefit risk of beti-cel for the treatment of transfusion dependent beta-thalassemia is favorable.

11.3 Discussion of Regulatory Options

The applicant has provided substantial evidence of effectiveness and safety from two adequate well controlled trials for beti-cell treatment of TDT. The provided data have demonstrated evidence of effectiveness of bet-cel, while indicating that safety is largely consistent with the prerequisite myeloablation, and delayed platelet engraftment followed by slow resolution of thrombocytopenia. On this basis, beti-cel will be granted regular approval.

The safety signal of this LVV-based therapy, along with concerns for potential insertional oncogenesis in related products, warrants close surveillance for secondary malignancy in the long term follow up study and the PMR safety study.

11.4 Recommendations on Regulatory Actions

The review team recommends regular approval for beti-cel for the treatment of children and adults with β thalassemia who require regular red cell transfusions.

11.5 Labeling Review and Recommendations

The review team negotiated with the applicant on several sections of the label, which included the Highlights section, the Adverse Reactions section, and sections 1, 2.2, 2.3, 3, 5.2, 5.6, 6, 6.1, 8.1, 8.4, 11, 12, 12.2, 14, 17, and the Patient Information Section, which were successfully resolved the time of BLA approval.

Reviewer Comment

Labeling negotiations with the Applicant have been completed.

11.6 Recommendations on Postmarketing Actions

The sponsor will conduct a PMR safety study. Upon consideration of the risks of LVV-based therapy, the previously considered registry-based analysis of spontaneous postmarketing adverse events reported under section 505(k)(1) of the Federal Food, Drug, and Cosmetic Act (FDCA) will not suffice. The pharmacovigilance system under section 505(k)(3) of the FDCA is also considered insufficient to assess this serious risk. Therefore, the Applicant will be required to conduct a postmarketing, prospective, multi-center, observational study to assess the long-term safety of betibeglogene autotemcel and the risk of secondary malignancies post ZYNTGLO. This PMR study under Section 505(o) of FDCA will enroll at least 150 patients TDT, to be followed for 15 years after product administration. The study design will specify regular monitoring for clonal expansion with adequate testing methods.

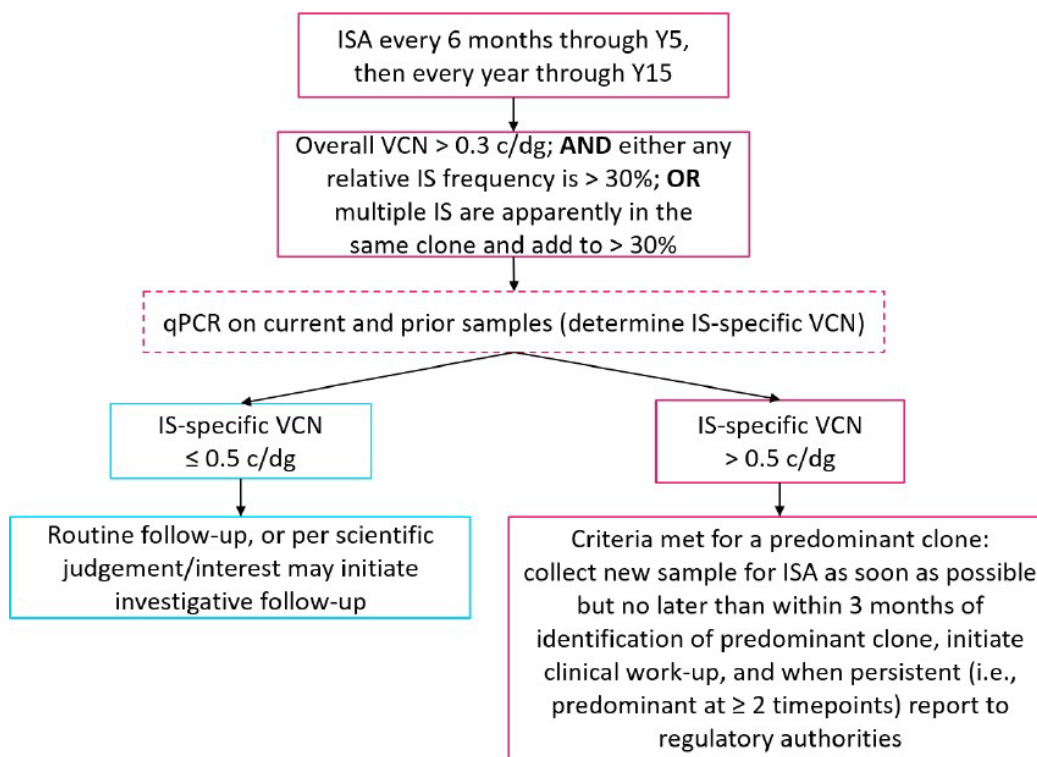
APPENDIX

Integration Site Analysis

Lentiviral vectors have the potential to alter the host genome at undesirable locations, thus portending a theoretical risk of malignancy. While insertional oncogenesis has not been observed with beti-cel to date, hematologic malignancy has been reported after treatment with LVV-based products in other diseases.

To evaluate for clonal predominance, beti-cel recipients underwent surveillance with an insertion site analysis (ISA) algorithm. ISA was performed using high-throughput, semi-quantitative methods which identify integration sites (IS) based on vector sequence primers. Identified insertion sites are considered of interest when the overall peripheral blood VCN is > 0.3 c/dg AND either any relative IS frequency is $> 30\%$ OR multiple IS are apparently in the same clone and add up to $> 30\%$. Multiple insertion sites apparently in the same clone is defined as more than one relative frequency where values are within 20% of each other (e.g., $5\% \pm 1\%$, $10\% \pm 2\%$, $15\% \pm 3\%$, etc.), as well as any additional cases identified through the Applicant's internal review of ISA reports. When multiple IS appeared in the same clone, a confirmatory bone marrow or peripheral blood colony-forming unit assay was performed. IS of interest would be interrogated, using a quantitative assay (e.g., qPCR) designed to detect the specific IS and determine an IS-specific VCN that will help to estimate clonal contribution. If results of the quantitative, IS-specific follow-up assay reveal an IS-specific VCN > 0.5 c/dg, estimating $> 50\%$ clonal contribution, criteria will be met to consider the subject as having a predominant clone. This threshold also applies to individual lineage evaluations (myeloid, lymphoid, etc.) when performed. Clinical work-up would be recommended for a predominant clone, and a repeat sample was to be collected within 3 months after identification of a predominant clone, with retrospective testing by IS-specific qPCR done on sample(s) previously collected, as available. A predominant clone identified at 2 or more time points is considered persistent. The algorithm is presented in the figure below:

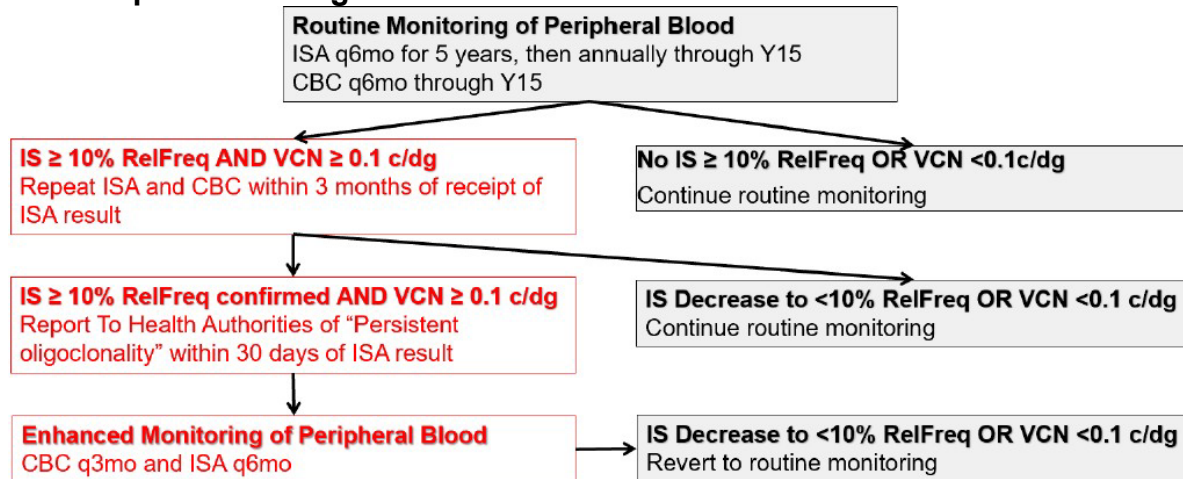
Figure 4 Original ISA Algorithm



Proposed Revision of IS screening strategy

To increase the sensitivity of the IS screening algorithm to detect persistent oligoclonality, a known risk factor for malignancy, the Applicant proposed a new ISA algorithm, lowering the minimum threshold Relative Frequency (RelFreq) to $\geq 10\%$ associated with a vector copy number (VCN) of ≥ 0.1 c/dg and used this as a trigger for increased clinical hematologic monitoring. Persistent oligoclonality ($\geq 10\%$ Relative Frequency at two consecutive ISA timepoints) would be reported to Health Authorities. The figure below illustrates the proposed algorithm:

Figure 5 Proposed ISA Algorithm



Note: If a VCN result is not available for the same visit at which the RelFreq exceeds 10% for the first time, the VCN will be done at the same time that the ISA is repeated. During enhanced monitoring, VCN will be performed on same samples as ISA if not already scheduled as per SOE.

Of note: The same ISA algorithm is used across trials for the related product (lovo-cel) to treat sickle cell disease (SCD), and for a different product (eli-cel) used to treat childhood cerebral adrenoleukodystrophy (CALD).

Study HGB-207 schedule of events

	Follow-Up: Day (D), Month (M) (Visit Window, days) Post-Drug Product Infusion																		
	D30 (±7)	D60 (±7)	D90 (±7)	D120 (±14)	D150 (±14)	D180 (±14)	D210 (±14)	D240 (±14)	D270 (±14)	D300 (±14)	D330 (±14)	D360 (±30)	D420 (±30)	D450 (±30)	D480 (±30)	D540 (±30)	D600 (±30)	D660 (±30)	D720 (±30)
Physical examination ¹	X	X	X	X	X	X			X			X		X		X			X
Vital signs	X	X	X	X	X	X			X			X		X		X			X
Blood for serum β-human chorionic gonadotropin for women of child-bearing potential (serum pregnancy test) ²			X			X													
Blood for clinical laboratory tests ³	X	X	X	X	X	X			X			X		X		X			X
Blood for CBC only							X	X		X	X		X		X		X	X	X
Blood for fasting Glucose/Insulin levels ⁴						X						X				X			X
Estimated GFR			X			X						X							X
Blood for iron studies ⁵			X			X						X		X		X			X
Blood for immunology ⁶			X			X						X							X
Blood for hormonal and dyserythropoiesis testing ⁷												X							X
Blood for globin in autologous cells												X							X
Blood for globin HPLC & VCN		X	X			X			X			X				X			X
Blood for RCL analyses ⁸			X			X						X							X ⁹
Blood for ISA						X						X				X			X
Blood for storage, potential biomarker analysis (optional)						X						X							X
Bone marrow ¹⁰												X							X
Liver and spleen SOC MRI												X							X

Procedure	Follow-Up: Day (D), Month (M) (Visit Window, days) Post-Drug Product Infusion																		
	D30	D60	D90	D120	D150	D180	D210	D240	D270	D300	D330	D360	D420	D450	D480	D540	D600	D660	D720
	M1 (±7)	M2 (±7)	M3 (±7)	M4 (±14)	M5 (±14)	M6 (±14)	M7 (±14)	M8 (±14)	M9 (±14)	M10 (±14)	M11 (±14)	M12 (±30)	M13 (±30)	M14 (±30)	M15 (±30)	M16 (±30)	M17 (±30)	M18 (±30)	M19 (±30)
Pulmonary function tests ¹¹												X							X
Cardiac and liver T2* MRI & echocardiology ¹²												X							X
12-lead ECG												X							X
Bone imaging (X-ray and/or DEXA Scan) ¹³																			X
HRQoL Assessment			X ¹⁴			X						X				X			X
Record transfusions	Continuous from ICF signing																		
Record hospitalizations	Continuous from post-drug product infusion discharge																		
Adverse event collection	Continuous from ICF signing																		
Concomitant medication (incl. iron chelators & phlebotomy)	Continuous from ICF signing																		

Abbrev: AE, adverse event; CBC, complete blood count; ECG, electrocardiogram; EPO, erythropoietin; EQ5D, EuroQol-5D; FACT-BMT, Functional Assessment of Cancer Therapy Bone Marrow Transplant; FSH, follicle stimulating hormone; GFR, glomerular filtration rate; HPLC, high performance liquid chromatography; HRQoL, health-related quality of life; ICF, informed consent form; IGF, insulin-like growth factor; IGFBP, insulin-like growth factor binding protein; Ig, immunoglobulin; ISA, integration site analysis; LH, luteinizing hormone; MRI, magnetic resonance imaging; PBL, peripheral blood leukocytes; PTH, parathyroid hormone; RBC, red blood cell; RCL, replication competent lentivirus; SOE, Schedule of Events; SOS, sinusoidal obstruction syndrome; T4, thyroxine; TSH, thyrotropin; VCN, vector copy number

¹ Includes weight at every visit, height and performance status every 6 months after drug product infusion. Tanner staging should be performed every 6 months during puberty, if relevant. For subjects <18 years of age, neurocognitive development will be evaluated every 6 months.

² Should be confirmed prior to mobilization

³ Hematology (CBC, platelets, reticulocytes, nucleated RBCs); serum chemistry and liver function tests, and additional clinical laboratory tests as clinically indicated. Clinical laboratory tests should be obtained on a more frequent basis if clinically indicated (e.g. monitoring & evaluation of AEs). If the results from blood tests are not as expected, additional testing may need to be performed and may include a physical exam, blood tests, imaging tests, or a bone marrow biopsy to allow for further investigation of stem cells.

⁴ Fasting glucose and insulin levels (HOMA index) at least every 6 months. An oral glucose tolerance test should be performed for an abnormal fasting glucose.

⁵ Iron studies (iron, ferritin, serum transferrin receptor, transferrin) should also be performed prior to restarting iron chelation/phlebotomy.

⁶ T cell subsets (CD4, CD8), B cells (CD19), and natural killer cells (CD16 and/or CD56); immunoglobulins (IgG, IgM, and IgA).

⁷ Hormonal testing includes thyroid function (free T4, TSH); PTH. For subjects after puberty, also includes LH, FSH and estradiol (females only); testosterone (males only). For subjects <18 years of age, includes growth hormone (IGF-1 and IGFBP-3). Dyserythropoiesis testing includes EPO and hepcidin.

⁸ Two samples required, one for RCL screening test, another for potential coculture of PBLs if RCL screening test is positive.

⁹ If a subject's previous RCL tests were all negative, the 24 Month sample will be archived.

¹⁰ Bone marrow for dyserythropoiesis studies (reticulocytes, nucleated RBC, serum transferrin receptor, hepcidin, and erythropoietin), as well as morphology, cellularity, cell count, and iron content; other research tests (e.g., VCN, ISA, HPLC) may be performed if sufficient sample is available.

¹¹ Including oxygen saturation; corrected % predicted FVC; % predicted FEV1; % predicted RV; and % predicted DLco (corrected for Hb and/or alveolar volume, as clinically indicated). If subject cannot perform these pulmonary function tests due to age or cognition-related restrictions, then respiratory exam, chest radiograph, and pulse oximetry will substitute for these assessments. If subject becomes able to perform spirometry and lung diffusion capacity test, these pulmonary function tests should be performed per SOE and/or at an unscheduled timepoint.

¹² Annual echocardiography is only required if clinically significant abnormality is observed on the Screening echocardiogram, or any subsequent echocardiogram, or if there is evidence of iron overload (cardiac T2* ≤20 ms) or other clinically significant abnormality on cardiac T2* MRI.

¹³ Age appropriate bone imaging (bone age/ mineral density) to be done based on investigator judgment.

¹⁴ At the Day 90 Visit, only the EQ-5D and FACT-BMT tools are to be completed.

Source: Page 59 HGB-207

Study HGB-204 Schedule of Activities

Schedule of events during the HGB-204 study

	Follow-Up: Day (D), Month (M) (Visit Window, days)																			
	D30 (±7)	D60 (±7)	D90 (±7)	D135 (±14)	D180 (±14)	D210 (±14)	D240 (±14)	D270 (±14)	D300 (±14)	D330 (±14)	D360 (±30)	D420 (±30)	D450 (±30)	D480 (±30)	D540 (±30)	D600 (±30)	D630 (±30)	D660 (±30)	D720 (±30)	
Procedure	M1 (±7)	M2 (±7)	M3 (±7)	M4.5 (±14)	M6 (±14)	M7 (±14)	M8 (±14)	M9 (±14)	M10 (±14)	M11 (±14)	M12 (±30)	M14 (±30)	M15 (±30)	M16 (±30)	M18 (±30)	M20 (±30)	M21 (±30)	M22 (±30)	M24 (±30)	
Physical examination ¹	X	X	X	X	X			X			X		X		X		X		X	
Vital signs	X	X	X	X	X			X			X		X		X		X		X	
Local lab: Blood for clinical laboratory tests ²	X	X	X	X	X			X			X		X		X		X		X	
Local lab: Blood for CBC only						X	X		X	X		X		X		X		X	X	
Local lab: hormonal testing ³											X								X	
Blood for iron studies ⁴			X								X								X	
Local lab: Blood for immunology ⁵			X		X			X			X									
Central lab: Blood for globin, VCN	X	X	X		X			X			X		X		X		X		X	
Central lab: Blood for RCL			X		X						X								X	
Central lab: Blood for ISA					X						X				X				X	
Blood for storage: potential biomarker analysis (optional)					X						X								X	
Bone marrow for globin, VCN ⁶											X								(X)	
Bone marrow for dyserythropoiesis testing ⁷											X								(X)	
Liver MRI/SQUID (iron assessment)											X								X	
Pulmonary function tests ⁸											X								X	
Cardiac MRI & echocardiology											X								X	
HRQoL Assessment					X						X				X				X	
Record transfusions	Continuous from ICF signing																			
Record hospitalizations	Continuous from ICF signing																			
Adverse event collection	Continuous from ICF signing																			
Concomitant medication (incl. iron chelators & phlebotomy)	Continuous from ICF signing																			

Procedure	Follow-Up: Day (D), Month (M) (Visit Window, days)																			
	D30 (±7)	D60 (±7)	D90 (±7)	D135 (±14)	D180 (±14)	D210 (±14)	D240 (±14)	D270 (±14)	D300 (±14)	D330 (±14)	D360 (±30)	D420 (±30)	D450 (±30)	D480 (±30)	D540 (±30)	D600 (±30)	D630 (±30)	D660 (±30)	D720 (±30)	
Included weight and spleen size at every visit, height and performance status every 6 months after Leni/Globin BB305 Drug Product infusion. Tanner staging should be performed every 6 months during puberty, if relevant.																				
¹ Hematology (CBC, platelets, reticulocytes, unclashed RBCs); iron studies (iron, ferritin, transferrin, soluble transferrin receptor), serum chemistry and liver function tests.																				
² Includes TSH, T3, T4, FSH, LH, estrogen or testosterone level, as applicable. For adolescents, include also growth hormone (IGF-1) and IGF BP-3).																				
³ Iron studies should also be performed prior to restarting iron chelation/phlebotomy, if needed.																				
⁴ T cell subsets [CD4, CD8], B cells (CD19), and natural killer cells (CD16 and/or CD56); immunoglobulins (IgG, IgM, and IgA)																				
⁵ VCN in bone marrow to be performed at any time if there is evidence of clonal skewing in peripheral blood leukocytes (PBLs); at D360 if sufficient sample available when collecting for dyserythropoiesis testing; or at the investigator's discretion; globin analysis of erythroid burst forming units may be done at the investigator's discretion																				
⁶ At D360 visit, and at D720 at investigator's discretion																				
⁷ Including FVC; FEV1; TLC; RV; VC; and DLCO: % predicted FVC, % predicted FEV1, % predicted RV, % and predicted DLCO (corrected for hemoglobin)																				

Procedure	Follow-Up: Day (D), Month (M) (Visit Window, days)																			
	D30	D60	D90	D135	D180	D210	D240	D270	D300	D330	D360	D420	D450	D480	D540	D600	D630	D660	D720	
	M1 (±7)	M2 (±7)	M3 (±7)	M4.5 (±14)	M6 (±14)	M7 (±14)	M8 (±14)	M9 (±14)	M10 (±14)	M11 (±14)	M12 (±30)	M14 (±30)	M15 (±30)	M16 (±30)	M18 (±30)	M20 (±30)	M21 (±30)	M22 (±30)	M24 (±30)	
¹ Include weight and spleen size at every visit, height and performance status every 6 months after LentiGlobin BB305 Drug Product infusion. Tanner staging should be performed every 6 months during puberty, if relevant.																				
² Hematology (CBC, platelets, reticulocytes, nucleated RBCs); iron studies (iron, ferritin, transferrin, soluble transferrin receptor), serum chemistry and liver function tests.																				
³ Includes TSH, T3, T4, FSH, LH, estrogen or testosterone level, as applicable. For adolescents, include also growth hormone (IGF-1 and IGF BP-3).																				
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⁷ At D360 visit, and at D720 at investigator's discretion																				
⁸ Including FVC; FEV ₁ ; TLC; RV; VC; and DLco; % predicted FVC; % predicted FEV ₁ ; % predicted RV; and % predicted DLco (corrected for hemoglobin)																				

Source: Copied from Page 51 and 52 of HGB-204 Study Report

FDA Group Terms:

Abdominal pain: abdominal pain upper, abdominal pain lower, abdominal pain, abdominal discomfort

Anemia: Anaemia postoperative, Autoimmune haemolytic anaemia, Haemolytic anaemia

Cough: Cough and Productive cough, Upper-airway cough syndrome

Headache: Migraine, Headache

Mucositis: mucosal inflammation, oral mucosal exfoliation, oral mucosal roughening, stomatitis, anal inflammation, pharyngeal inflammation

Musculoskeletal pain: musculoskeletal discomfort, musculoskeletal chest pain, bone pain, musculoskeletal discomfort, chest pain, myalgia, Neck pain, Non

cardiac chest pain, Pain in extremity, spinal pain, tendon pain, back pain

Oropharyngeal pain: Jaw pain, Oral pain, Oropharyngeal discomfort

Pharyngitis: Pharyngeal inflammation, Pharyngitis streptococcal

Pigmentation disorder: Skin hyperpigmentation, Skin hypopigmentation, Pigmentation disorder, Oral pigmentation

Rash: Rash follicular, Macular rash, Pruritic rash, Vesicular rash, Pustular rash,

Acne dermatitis, Acneiform, Macule, Petechiae, Dermatitis atopic

Rhinitis: Rhinorrhea, Rhinitis allergic,
Sepsis: Fungal sepsis, Bacterial sepsis, Neutropenic sepsis, Sepsis
Thrombosis: atrial thrombosis, Deep vein thrombosis, Deep Vein Thrombosis,
Device related thrombosis, Infective thrombosis, Jugular vein thrombosis, venous
thrombosis, Embolism, Pulmonary Embolism
Tachycardia: Sinus tachycardia, Tachycardia, Supraventricular tachycardia
Transaminases increased, Transaminases increased, and Transaminasaemia
URTI: Upper airway cough syndrome, Upper respiratory tract infection, Viral
upper respiratory tract infection Nasopharyngitis, pharyngitis, pharyngitis
streptococcal,
Viral infection: influenza like illness, Influenza, Parainfluenzae virus infection, BK
virus infection, Human rhinovirus test positive, rhinovirus infection, Human
rhinovirus test positive, SARS-CoV-2 test positive
Xerosis: dry eye, dry skin, dry mouth, Lip dry

Do Not Change Anything Below This Line