Study Results Synopsis for Public Disclosure

Name of product: OTQ923 (ADPT03) infusion

Protocol identification number: CADPT03A12101 (EUDRACT number 2019-003489-41, ClinicalTrials.gov number NCT04443907)

Title of study: A first-in-patient Phase I/II clinical study to investigate the safety and efficacy of genome-edited hematopoietic stem and progenitor cells in subjects with severe complications of sickle cell disease

Study centers: 3 centers in USA

Publication (reference): Sharma A, Boelens J-J, Cancio M et al (2023) CRISPR-Cas9 editing of the HBG1 and HBG2 promoters to treat sickle cell disease. N Engl J Med. 31;389(9):820-832. doi: 10.1056/NEJMoa2215643.

Sharma A, Hankins JS, Boelens J-J et al (2025) Normalization of cerebral hemodynamics after gene therapy in adults with sickle cell disease. American Journal of Hematology. 0:1–4. https://doi.org/10.1002/ajh.27757.

Study period:

Study initiation date: 25-Aug-2020 (first subject first visit)

Early termination date: 06-Jan-2025 (last subject last visit: the study was terminated due to business reasons)

Phase of development (phase of this clinical study): I/II

Objectives: The primary and secondary objectives and associated endpoints are presented below:

Table 0-1 Primary objectives and related endpoints

Objectives	Endpoints
Primary objectives	**Endpoints for primary objectives**
• To assess the safety and tolerability of genome-edited hematopoietic stem cell transplant (HSCT) in subjects with severe complications of sickle cell disease.	• Monitoring of adverse events, vital signs, electrocardiogram (ECG) and laboratory results.
• To assess time to engraftment	• Time to reach absolute neutrophil count (ANC) ≥ 500/µL for 3 consecutive days.
• To assess fetal hemoglobin (HbF) expression	• Quantify HbF expression 6 months after HSCT

Table 0-2 Secondary objectives and related endpoints

Secondary objectives	Endpoints for secondary objectives
• To assess the durability of hematologic engraftment, persistence of chimerism	• Engraftment durability/persistence by measuring the proportion of alleles with on-target CRISPR modification

kinetics and HbF expression	in peripheral blood (total white blood cells (WBC)) and bone marrow over time up to 24 months • Proportion of subjects to achieve 30% of total HbF at 12 months • Time to achieve 30% total HbF • Time to peak total HbF • Percentage of edited WBC and bone marrow cells by time points, derivation of kinetic parameters as appropriate and summary statistics for the extent and duration of engraftment
• To evaluate presence of pre-existing or treatment induced anti-Cas9 humoral and cellular immunogenicity	• Summary of incidence and prevalence of humoral (anti-cas9 antibodies) and cellular immunogenicity (Cas9 reactive T cells) including pre-existing and treatment induced over time up to 24 months, if appropriate as supported by data • Impact of pre-existing and treatment-induced immunogenicity (humoral and cellular) on efficacy, safety, and chimerism kinetics*
• Overall survival	• Overall Survival • Transplant-related mortality • Change from baseline annualized vaso-occlusive crises (VOC) rate by 65% by 12 months • Change from baseline annualized sickle cell disease (SCD) complications (aggregate of VOC, acute chest syndrome (ACS), priapism and stroke) and, if relevant, rate of transfusions, by 65% by 12 months
• To evaluate changes in patient-reported outcomes	• Changes from baseline pre-HSCT and post-HSCT over time up to 24 months in the Adult Sickle Cell Quality of Life Measurement Information System (ASCQ-ME), Patient-Reported Outcomes Measurement Information System (PROMIS) Fatigue and PROMIS Physical Functioning.

*The analysis of this secondary objective was not performed as no meaningful data was available for the analysis.

Study design and methodology: This multicenter, open-label, first-in-human, proof-of-concept study in SCD subjects involved mobilized hematopoietic stem and progenitor cells (HSPCs) apheresis, *ex vivo* CRISPR/Cas9 genome editing, myeloablative conditioning, and autologous HSCT, with follow-up for 1-2 years. Genome editing was done via HSPC electroporation with guide RNA and Cas9 protein.

The study had 2 parts: Part A enrolled adult subjects dosed with OTQ923, with the first 2 subjects monitored as sentinels for 28 days before treating others. Part B, intended for pediatric subjects, was planned but not opened for enrollment.

To optimize the quality and quantity of mobilized HSPCs, subjects were to stop hydroxyurea (HU) (if applicable) and receive red blood cell transfusions for at least 2 months prior to

mobilization with plerixafor. In addition to collecting sufficient CD34+ cells for gene editing, an additional back up unit of unedited HSPCs was also collected for graft failure contingencies.

After meeting release criteria, subjects received busulfan based conditioning to minimize SCD toxicity, followed by genome-edited HSPCs infusion. Subjects remained under inpatient care until engrafted and deemed safe for outpatient care. Study visits occurred weekly until 2 months after HSCT, then monthly until 6 months after HSCT, every other month until 12 months after HSCT, and then every 3 months until end of study (EOS).

All the treated subjects then entered a long-term safety follow-up study (CADPT03A12001), for safety monitoring up to 15 years after HSCT, as per guidance from Health Authority.

Diagnosis and main criteria for inclusion: The study population comprised of male and female SCD subjects with globin typing (e.g., HbSS, HbSC, HbS/β0-thalassemia or others) from age 2 to 40 years.

Part A treated up to 10 adult subjects (age 18-40 years).

Part B was planned to treat up to 10 pediatric subjects (age 2-17 years).

Test and reference therapies, dose and mode of administration, batch number: In Part A, subject received intravenous infusion of OTQ923 cell suspension. When necessary, drug product from more than one manufacturing batch was combined to achieve the required dose. Part B planned to treat pediatric subjects with OTQ923 via i.v. infusion was not opened for enrollment.

No reference therapy was administered.

Protocol amendments and other changes to study conduct:

This CSR describes the conduct of the study according to the final protocol dated 14-Apr-2023 (including all 4 amendments). The key changes in all protocol amendments are described in Table 2-3.

Table 0-3 Protocol amendments

Version and date	Summary of key changes
Amendment 1, 17-Apr-2020	Amendment was initiated in response to requests by the US-FDA. The following changes were made: 1. The two sentinel subjects enrolled in Part A and Part B were to be monitored until engraftment and for a minimum of 28 days to provide sufficient safety data for review before treating the next subject. 2. Enrollment in Part C was to start after a minimum of 6 months of data was reviewed from at least 3 adult subjects post-HSCT. Based on these data, benefit/risk ratio for pediatric subjects in the study were to be assessed and existing protocol were then be amended to include adult data to provide rational supporting enrollment of pediatric subjects in the trial. The data from adult subjects and protocol amendment was subject to a regulatory review before the implementation of the protocol amendment. 3. Updated endpoints for the secondary objective “Overall and event-free survival” to provide clarity.

Version and date	Summary of key changes
	4. Inclusion of subjects who had failed, not tolerated or refused HU treatment. 5. Capping the maximum dose of the investigational drug product. 6. Capping the maximum volume and number of CD34+ cells collected as part of the apheresis in the study. 7. Updated the study stopping rules to enhance safety before resuming the study. 8. PRO instruments were streamlined to minimize the gathering of redundant or similar information and to reduce the burden on subjects. Added optional subject exit interview.
Amendment 2, 30-Apr-2021	Protocol amended mainly to incorporate a new screening window based on current clinical timelines, to allow combining more than one manufacturing batch for one subject, if needed.
Amendment 3, 25-May-2022	The main purpose of this protocol amendment was threefold: to reassign Part B of the study to explore OTQ923 in pediatric subjects with SCD, increase the screening window and increase the lower limit of apheresis harvest. Based on the initial apheresis and manufacturing experience in this trial, generating a protocol-defined dose relies on an adequate number of starting CD34+ cells and a successful manufacturing run. Therefore, the lower limit of the incoming apheresis material was increased. This proposed change increased the chances of successful manufacturing for each subject. Additionally, in consideration of the experimental nature of study treatment, the definition of graft failure was updated to reflect a longer timeframe, 45 days after HSCT. This amendment included the correction of typographical errors, and inconsistencies, to ensure data quality and minimize the risk of inconsistent interpretation.
Amendment 4, 14-Apr-2023	The main purpose of this amendment was threefold: • To allow for subjects to transition from this study after at least 1 year from OTQ923 infusion and enroll into a separate optional LTFU study (CADPT03A12001) instead of completing 2 years within the study. • The minimum OTQ923 dose for infusion was decreased. • The description of contraception measures was modified to adhere to the definition of highly effective contraception as per the Novartis Guideline on Prevention of Pregnancies in Participants in Clinical Trials.

Criteria for evaluation:

Efficacy: The primary and secondary efficacy assessment included the following:

1. Engraftment (ANC ≥ 500/microliter for 3 consecutive days)
2. Assessment of HbF expression
3. Proportion of subjects to achieve 30% of total fetal hemoglobin over time, time to achieve 30% of total fetal hemoglobin, and time to peak of total fetal hemoglobin.
4. Overall survival.

5. Chimerism at 12 months (peripheral blood leukocytes and bone marrow)
6. Number of VOC pain episodes and ACS episodes during the course of the trial
7. Change from baseline annualized VOC rate by 65% by 12 months
8. Change from baseline annualized SCD complications (aggregate of VOC, ACS, priapism and stroke) and, if relevant, rate of transfusions, by 65% by 12 months
9. Evaluation of effect on patient-reported outcomes from baseline and up to 24-month post-HSCT with age-appropriate patient reported outcomes (PRO) measures with ASCQ-ME, PROMIS Fatigue and PROMIS Physical Function.

Safety: Key safety assessments included reporting of adverse events (AEs) and serious adverse events (SAEs), physical examination, vitals and body measurements, ECG, and clinical laboratory tests (hematology, chemistry, and urinalysis).

Pharmacokinetics: The term pharmacokinetics (PK) was not applicable for genetically modified cell therapies, and hence the term *in vivo* cellular kinetics was used to describe the time course of engraftment and persistence of the gene edits.

Engraftment and persistence of genome-edited HSPC were monitored by measuring the CRISPR/Cas9 on-target editing frequency and editing pattern in bone marrow CD34+ cells and white blood cells using droplet digital polymerase chain reaction (ddPCR).

Biomarker: Expression level of HbF in blood was determined by high performance liquid chromatography (HPLC) (central laboratory) as one of the primary objectives to understand pharmacodynamics (PD) effect and efficacy of treatment.

Immunogenicity assessments: Immunogenicity samples were obtained and evaluated in all subjects to assess the presence and level of pre-existing and treatment induced anti-Cas9 antibodies and Cas9-reactive T cells.

Humoral immunogenicity was determined using an enzyme-linked immunosorbent assay method designed to detect the presence of anti-Cas9 antibodies in human serum.

Cellular immunogenicity was evaluated using an assay that detects interferon gamma (IFNg) expression of T cells that are activated specifically by Cas 9 peptides derived from the JFA065 Cas9 protein. Percent of IFNg expression positive cells by flow cytometry define the level of T cell activation, comparing Cas9 with two positive controls (Staphylococcal enterotoxin B, Cytomegalovirus, Epstein-Barr virus, and Influenza viruses peptide pool) and negative control (dimethyl sulfoxide (DMSO)).

Statistical methods: Study data were analyzed by Novartis personnel using the most updated SAS® version. The analysis sets used in the study are described below:

The Full Analysis Set (FAS) included all subjects who did not screen fail. Subjects were assigned to OTQ923.

The Safety set included all subjects who received any study treatment. Subjects were analyzed according to treatment received.

Safety analyses: The primary endpoints for the safety and tolerability objective included AEs, vital signs, ECGs, and laboratory results. All safety summaries were based on the Safety set (SAS). For safety analyses, all listings and tables were planned to be presented by treatment

group (subjects in the same study part were to be pooled into one single treatment group). Formal hypothesis testing was not performed.

Efficacy analyses: Efficacy analyses used the Safety set. Primary efficacy analyses were descriptive in nature, and formal hypotheses were not assessed.

Time to engraftment was listed by subject and was also summarized. HbF% along with change from baseline was listed by subject. Individual and summary plots for HbF% were provided over time. Summary statistics for HbF% and change from baseline were provided by visit/time. Additionally, the number and percentage of responders (HbF ≥ 30%) and time to peak HbF were summarized as secondary endpoints.

For all secondary endpoints, summary statistics were provided based on the Safety set. Continuous variables were summarized by number of observations, mean, standard deviation, median, minimum and maximum. Categorical variables were summarized by number and percentage of observations within each category of the parameter. Graphical summaries showing change over time were provided for select continuous variables: HbF.

Pharmacokinetic analyses: This study involved genetically modified cell therapies and hence the term *in vivo* cellular kinetics was used instead of pharmacokinetics to describe the time course of engraftment and persistence.

The engraftment and persistence of genome-edited HSPCs, along with their kinetic profiles, were monitored by measuring the CRISPR/Cas9 on-target editing frequency and editing pattern in bone marrow CD34+ cells and WBCs using droplet digital PCR and next-generation sequencing (NGS) amplicon-sequencing. The results are presented under the efficacy section.

Immunogenicity analyses: Pre-existing and treatment induced anti-Cas9 antibodies and Cas9-reactive T cells were analyzed and reported by subject and visit/time.

Changes to planned analysis: The following planned protocol analyses were not performed due to the limited number of subjects enrolled in the study (i.e. < 10 subjects) and the nature and modality of the data (such as low prevalence of treatment induced immunogenicity):

- Percentage of edited WBC and bone marrow cells by time points, derivation of kinetic parameters as appropriate and summary statistics for the extent and duration of engraftment and
- Impact of pre-existing and treatment-induced immunogenicity (humoral and cellular) on efficacy, safety, and chimerism kinetics.

Summary - Results

Subject disposition

Of 13 enrolled subjects, 4 (30.8%) subjects received OTQ923 cell suspension infusion and completed the study. Two (15.4%) subjects entered the long-term follow-up by 10-Apr-2025, while 2 subjects consented for the follow-up study after the data-base lock. The remaining 9 (69.2%) subjects discontinued the study without receiving OTQ923 infusion.

Demographic and background characteristics: At baseline, the median age of subjects was 21.5 years (range: 18 to 24 years), with an equal proportion of male (50.0%) and female (50.0%) subjects. All subjects were racially Black or African American (100%) and the majority had non-Hispanic or non-Latino ethnicity (75.0%).

Protocol deviations: Protocol deviations were reported in all the 4 (100%) subjects. All protocol deviation were of 'other' category that were related to study visit done outside of window period not due to COVID-19 (75.0%), failure to perform key procedures per protocol not due to COVID-19 (75.0%), and optional DNA collection prior to signing optional genetic ICF (sample collected was discarded and not analyzed) (25.0%). There was no impact on study outcomes or subject safety due to PDs.

Efficacy and Immunogenicity results:

- All 4 subjects (100%) achieved neutrophil engraftment with median time to neutrophil engraftment of 23 days.
- The median HbF remained consistently higher as compared to the baseline levels with HbF expression 6 months after HSCT at 20.6%.
- The maximum allelic editing in WBC (5Kb deletion: 33.5%, HBG1: 13.2%, HBG2: 16.7%) was observed at Week 8, while bone marrow editing for 5 Kb deletion, HBG1, and HBG2 remained comparable at Months 12 and 24.
- No subject achieved 30% HbF at 12 months after OTQ923 infusion.
- No deaths or transplant-related mortality were reported during the study.
- Of the 4 subjects, one subject achieved 65% reduction in annualized VOC rate, SCD complications, and annualized transfusion rate by 12 months.
- Overall, results of ASCQ-ME, PROMIS fatigue, and PROMIS Physical Functioning showed high variability in a the small number of subjects (n = 2 to 4 at different time points), which limits formal conclusion and highlights individual differences in response.
- Cellular immunogenicity responses remained consistently low (< 0.1%) for all subjects throughout the study, demonstrating that cellular immunogenicity did not increase over time. Further, all the subjects (100%) tested negative for anti-Cas9 antibodies post treatment administration. One subject had anti-Cas9 antibody at the baseline.

Pharmacokinetic results: Pharmacokinetic and imaging assessments for exploratory endpoints were not evaluated.

Safety results:

- Overall, the treatment with single infusion of OTQ923 was well tolerated by subjects with SCD.
- All subjects enrolled in the study experienced at least one AE, post-infusion of OTQ923.
- The most commonly reported AEs (any grade, irrespective of relationship to OTQ923) were arthralgia, constipation, stomatitis, anemia, febrile neutropenia, pain in extremity, decreased platelet count, and sickle cell anemia with crisis. Most of these AEs were severe.
- Three subjects experienced AEs (anemia, febrile neutropenia, stomatitis, decreased platelet count, and decreased neutrophil count decreased) suspected to be related to the conditioning regimen. Additionally, 2 subjects experienced AEs (anemia and pyrexia) suspected to be related to OTQ923.
- A total of 3 subjects reported SAEs, with only one SAE of pain in extremity suspected as related to the conditioning regimen. The most commonly reported SAE was sickle cell anemia with crisis (3 subjects). No SAEs were considered as related to OTQ923.

- No fatal SAEs were reported during the study.
- There were no clinically meaningful changes reported in laboratory parameters, vital signs, or ECGs during the study.

Conclusions: Overall, the treatment with a single infusion of OTQ923 following busulfan conditioning was well tolerated and demonstrated stable editing resulting in sustained increase in HbF levels over baseline and follow-up period, although did not meet the threshold of > 30% increase in HbF%. No unexpected serious safety concern was observed with OTQ923.

The treated subjects are being monitored for long-term safety in a separate study.