Protocol C5351005

An Open-label Extension Study to Evaluate the Long-term Safety of Osivelotor Administered to Participants with Sickle Cell Disease Who Have Participated in an Osivelotor Clinical Trial

Statistical Analysis Plan (SAP)

Version: **1.0**

Date: 23 JUN 2025

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1. VERSION HISTORY

Table 1. Summary of Changes

Version/ Date	Associated Protocol Amendment	Rationale	Specific Changes
1.0 / 23 Jun 2025	Protocol amendment 4.0 dated 06 Feb 2024	Not Applicable (N/A)	N/A

2. INTRODUCTION

This statistical analysis plan (SAP) provides the detailed methodology for summary and statistical analyses of the data collected in Study C5351005.

The primary analysis for this study will occur after all enrolled participants have completed End of Study (EOS) visit or have discontinued the study.

2.1. Modifications to the Analysis Plan Described in the Protocol

- All VOC data will be summarized including the events that occur after the inter-current events described in the protocol.
- All data for hemoglobin, reticulocytes, lactate dehydrogenase, and unconjugated bilirubin will be summarized including the data after the inter-current events described in the protocol.
- Updated definition of PK analysis set.
- Changed endpoint from “Incidence of SCD-related SAEs” to “Incidence of SCD-related AEs” to include all AEs.

2.2. Study Objectives, Endpoints, and Estimands

The table below has been taken verbatim from the protocol, with change described above.

Presentation of exploratory endpoints maybe modified and are described in the relevant sections (Section 6.3).

Objectives	Endpoints	Estimands
Primary:	**Primary:**	**Primary:**
To evaluate the long-term safety of osivelotor in participants with SCD.	• Incidence of TEAEs, changes in laboratory assessments, and changes in vital signs.	The safety estimand for the primary endpoints will consider all safety data collected. Specifically, all data collected on or after obtaining informed consent will be included in the analysis.
Secondary:	**Secondary:**	**Secondary:**
To evaluate frequency of VOCs and SCD-related complications. To evaluate the effects of long-term use of osivelotor on hemolytic anemia.	• Annualized rate of VOC. • Incidence of SCD-related AEs. • Change from baseline in hematological laboratory parameters, including hemoglobin, reticulocytes, lactate dehydrogenase, and unconjugated bilirubin.	Not Applicable

Objectives	Endpoints	Estimands
Exploratory:	**Exploratory:**	**Exploratory:**
To evaluate the long-term effects of osivelotor treatment on inflammation, erythropoietin levels, SCD specific biomarkers, and HRQOL assessments.	• Change from baseline in adhesion of whole blood to microfluidic channels. • Change from baseline in RBC sickling from peripheral blood smears. • Change from baseline in RBC deformability and RBC mitochondrial content. • Change from baseline in albumin-to-creatinine ratio and kidney injury molecule. • Change from baseline in erythropoietin levels. • Change from baseline in haptoglobin, hemopexin, and soluble C3. • Change from baseline in HRQOL assessments including PROMIS SF Fatigue 13a, PROMIS SF Pain Interference 8a, PROMIS NRS Pain intensity, PGI-C of fatigue and pain, CGI-C of SCD, PGI-S of fatigue and pain, and CGI-S of SCD, EuroQol EQ-5D-5L, and WPAI+CIQ:SCD.	Not Applicable
To evaluate the PK parameters of long-term exposure to osivelotor	Blood and plasma concentrations of osivelotor.	Not Applicable

Abbreviations: CGI-C, Clinician's Global Impression of Change; CGI-S, Clinician's Global Impression of Severity; PGI-C, Patient's Global Impression of Change; PGI-S, Patient's Global Impression of Severity; PK, pharmacokinetic; PROMIS, Patient-Reported Outcome Measurement Information System; HRQOL, health-related quality of life; RBC, red blood cell; SCD, sickle cell disease; SF, short form, WPAI+CIQ:SCD, Work Productivity and Activity Impairment Questionnaire plus Classroom Impairment Questions: Sickle Cell Disease, VOC – vaso-occlusive crisis.

2.2.1. Primary Estimand(s)

The safety estimands for the primary endpoints will consider all safety data collected. Specifically, all data collected on or after obtaining informed consent will be included in the analysis.

Population: Patients with SCD as defined by the inclusion/exclusion criteria.

Endpoints:

- TEAE
- Lab assessments
- Vital Signs

Intercurrent Events: NA

Population Level Summary: will be provided by group/dose arm (Section 5.3). Specifically,

- for continuous endpoints: Mean
- for categorical endpoints: Proportion

2.2.2. Secondary Estimand(s)

Not applicable

2.2.3. Study Design

C5351005 (OLE) is designed as a multicenter, global, open-label extension study. The primary objective is to assess the long-term safety of osivelotor in participants with SCD. This study is available to eligible participants who have previously enrolled in a Pfizer-sponsored osivelotor clinical study and meet the entry criteria for this study.

Eligible participants will receive open-label osivelotor at Baseline (Day 1). It is intended that Day 1 coincides with the final visit from the originating osivelotor study. End of Treatment (EOT) assessments from the originating study will serve as Baseline assessments for this study. If Day 1 for this study is greater than 7 days from the completion of the originating study, new baseline assessments will be obtained. If during the period between Day 1 of this study and completion of the originating study no dose is received for greater than 3 consecutive days, an appropriate loading dose will be administered prior to continued maintenance dosing.

Only participants from C5351004 Part A (also referred to as the “parent study”) were enrolled into the OLE. C5351004 Part A is a study which enrolled only adult participants. There are two sets of participants rolling over from C5351004 Part A to the OLE study – (i) participants without a delayed start (Date of first dose in C5351005 – EOT date from C5351004 + 1 ≤ 42 days) and (ii) participants with a delayed start of OLE (Date of first dose in C5351005 – EOT date from C5351004 + 1 > 42 days). Each set follows a distinct schedule of assessments as outlined in the C5351005 protocol.

3. ENDPOINTS AND BASELINE VARIABLES: DEFINITIONS AND CONVENTIONS

3.1. Primary Endpoint(s)

3.1.1. Incidence of TEAEs

A treatment-emergent adverse event (TEAE) is defined as an AE that occurs during the effective duration of treatment defined as the time from the first dose of study drug through last dose of study drug + 56 days.

TEAEs that occur in the parent study are not counted as TEAEs in this study. In case, the start date of AE is the same date as the treatment start date and the question in the AE CRF page “Is this AE continuing from the parent study?” = “Yes”, then the event is not a TEAE,

as long as it can be confirmed that the AE in question had a start date in the parent study and no end date in the parent study.

In addition, in case the start date of AE is unknown (partially or fully unknown) and the response to the question in AE CRF page “If complete date is not known or the start date is the same as Day 1, did the AE start prior to dosing?” = “No”, then the event is a TEAE. This particular CRF question will only be used when the AE start date is not completely known or the start date is the same as Day 1.

3.1.2. Change in laboratory assessments

Laboratory assessments include all hematology and chemistry parameters as collected in the study (excluding hemoglobin, reticulocytes, lactate dehydrogenase, and unconjugated bilirubin – these parameters belong to the secondary endpoints list. See Section 3.2.2).

For eGFR (estimated Glomerular Filtration Rate), data will be summarized as collected in mL/min/1.73m^2 unit. Data for participants whose baseline result (reported in mL/min) comes from the parent study will be excluded from the analyses.

Change will be described using change from baseline. Rules for defining baseline are described in Appendix 1.2.

3.1.3. Change in vital signs

Vital signs include systolic and diastolic blood pressure (mmHg), heart rate (beats per minute), respiratory rate (breaths per minute) and body temperature.

Change will be described using change from baseline. Rules for defining baseline are described in Appendix 1.2.

3.2. Secondary Endpoint(s)

3.2.1. Annualized rate of VOC

A VOC (Vaso-Occlusive Crisis) is defined as an acute episode of pain that:

- Has no medically determined cause other than a vaso-occlusive event, and
- Results in a visit to a medical facility (hospitalization, emergency department, urgent care center, outpatient clinic, or infusion center), and
- Requires parenteral narcotic agents, parenteral nonsteroidal anti-inflammatory drugs (NSAIDs), or an increase in treatment with oral narcotics.

VOC are categorized as:

- **Uncomplicated VOC**: a VOC that is NOT classified as an Acute Chest Syndrome (ACS), hepatic sequestration, splenic sequestration, priapism, or dactylitis.
- **Complicated VOCs**: includes ACS, hepatic sequestration, splenic sequestration, priapism, or dactylitis.

The annualized rate of VOC will be calculated as specified in Section 6.2.1.

3.2.2. Change from baseline in hematological laboratory parameters, including hemoglobin, reticulocytes, lactate dehydrogenase, and unconjugated bilirubin.

Hemoglobin will be reported in g/dL units.

Reticulocytes will be reported in 10^9/L units.
Lactate dehydrogenase will be reported in IU/L units.
Unconjugated bilirubin will be reported in umol/L units.

Rules for defining baseline are described in Appendix 1.2.

3.2.3. Incidence of SCD-related AEs

SCD-related AEs are defined as AEs with any of the Preferred Terms listed below:

- Sickle Cell Anemia with crisis
- Acute chest syndrome
- Hepatic sequestration
- Splenic sequestration
- Priapism
- Dactylitis
- Pneumonia
- Osteonecrosis
- Stroke
- Leg Ulcers

3.3. Exploratory Endpoints

3.3.1. Change from baseline in adhesion of whole blood to microfluidic channels.

Flow adhesion parameters include:

- result from whole blood flow adhesion on Vascular Cell Adhesion Molecules (VCAM) (cells/mm2)
- result from whole blood flow adhesion on P-selectin (cells/mm^2)
- result from isolated red blood cell flow adhesion on Vascular Cell Adhesion Molecules (VCAM) (cells/mm2)
- result from isolated white blood cell flow adhesion on P-selectin (cells/mm^2)

Rules for defining baseline are described in Appendix 1.2.

3.3.2. Change from baseline in RBC sickling from peripheral blood smears.

% sickled cells will be derived from the parameters obtained from the blood smear using the following equation:

$$\frac{total\ number\ of\ sickled\ cells\ per\ slide}{total\ number\ of\ cells\ per\ slide} x100\%$$

Rules for defining baseline are described in Appendix 1.2.

3.3.3. Change from baseline in RBC deformability

RBC deformability is measured by Elongation Index (EImin, EImax) and Point of Sickling (PoS)

where,

EI_{max} is the maximal elongation index, representing overall deformability under normoxic conditions.
EI_{min} is the minimal elongation index, representing overall deformability under hypoxic conditions.
PoS is point of sickling or point on the curve during deoxygenation when sickling begins.

Rules for defining baseline are described in Appendix 1.2.

3.3.4. Change from baseline in erythropoietin levels

Erythropoietin will be measured in IU/L units. Both raw erythropoietin and natural log-transformed erythropoietin will be summarized.

Rules for defining baseline are described in Appendix 1.2

3.3.5. Patient-Reported Outcome Endpoints

HRQOL will be assessed via patient-reported outcome endpoints and will be summarized.

Rules for defining baseline (when applicable) are described in Appendix 1.2.

- **PROMIS SF Fatigue 13a**

This questionnaire measures fatigue and consists of 13 questions. Each question is scored from 1-5 (1 = “Not at all” and 5 = “Very much”). After appropriate recoding, positively phrased questions (“I have energy” and “I am able to my usual activities”) are reverse scored (e.g. 5 = “Not at all” and 1 = “Very much”) and the raw total score is calculated by taking the sum of response values for each question. All questions must be answered in order to arrive at the raw total score.

- **PROMIS SF Pain Interference 8a**

This questionnaire measures the pain interference and consists of 8 questions. Each question is scored from 1-5 and a total raw score is calculated by taking the sum of response values for each question. All questions must be answered in order to arrive at the raw total score.

- **PROMIS NRS Pain intensity**

This questionnaire measures pain intensity and consists of a single question. Rating ranges from 0-10.

- **PGI-S of fatigue and PGI-S of pain**

These forms are not reflected in the SOA (Schedule of Assessments) of the protocol. However, these data are collected for participants who completed the same form in the parent study and rolled over to the OLE study.

Both forms have 2 questions each and the questions are based on a 5-point scale. For the purposes of summarization, the following numeric response will be assigned to each response category:

Not present / Not at all	0
Mild / A little bit	1
Moderate / Somewhat	2
Severe / Quite a bit	3
Very Severe / Very much	4

- **PGI-S**

This form is not reflected in the SOA (Schedule of Assessments) of the protocol. However, this data is collected for participants who completed the same form in the parent study and rolled over to the OLE study.

This form measures overall status on a 5-point scale. For the purposes of summarization, the following numeric response will be assigned to each response category:

None	0
Mild	1
Moderate	2
Severe	3
Very Severe	4

- **CGI-S**

The form is based on a 7-point scale. For the purposes of summarization, the following numeric response will be assigned to each response category:

Not assessed	Missing
Normal, Not at all ill	0
Borderline ill	1
Mildly ill	2
Moderately ill	3
Markedly ill	4
Severely ill	5
Among the most extremely ill	6

- **EuroQol EQ-5D-5L**

This form assesses the participant's health at the time of the clinic site visit focusing on categories of mobility, self-care, usual activities, pain/discomfort, anxiety/depression and on how good/bad their health is today.

- The visual analog scale (VAS) records the participant's health status score from 0 (worst health) – 100 (best health).
- The descriptive health questionnaire is comprised of 5 dimensions (mobility, self-care, usual activities, pain/discomfort, anxiety/depression), each having 5 levels (1 = no problems to 5 = extreme problems).
- An index score (based on US crosswalk calculator) is calculated for each participant based on the response across the 5 dimensions.

- **WPAI+CIQ:SCD**

WPAI+CIQ:SCD is the work productivity and activity impairment plus classroom impairment questionnaire. This form assesses the effect of SCD on the ability to work, attend classes, and perform regular daily activities.

3.3.6. Pharmacokinetic Concentration Data

Plasma and whole blood PK concentration data will be listed and summarized.

3.4. Baseline Variables

The following baseline variables will be summarized. No baseline variables will be used for statistical modeling.

- Demographics: age (years), sex, race, ethnicity, geographic region (North America, Sub-Saharan Africa).

Some of the following baseline data (unless mentioned otherwise) is not collected in C5351005 EDC and will be programmatically included from the parent study database and corresponding ADaM datasets:

- Baseline characteristics: weight (kg) (see Appendix 1.2), height (cm) and SCD genotype from the parent study.

- SCD history: HU use at parent study screening, L-glutamine use at parent study screening.

- VOC history

  VOCs (meeting definition in Section 3.2.1) within 12 months of parent study screening visit will be summarized in the following manner:

  a) Total number of VOCs in the past 12 months prior to parent study screening visit across all participants, total participant years in the past 12 months prior to parent study screening visit, annualized incidence rate of VOC in the past 12 months prior to parent study screening visit.

  b) Number of VOCs in the past 12 months prior to parent study screening visit per participant.

  c) Number of participants experiencing at least one VOC of specified type in the 12 months prior to parent study screening visit.

The 12-month VOC summaries will be based on counting individual events over 365 days prior to parent study informed consent date (screening visit).

- SCD complications history from parent study includes (i) historical SCD complications (including episodes ongoing at parent study screening) and (ii) concurrent SCD complications (episodes ongoing at parent study screening).

- Medical History (coded using MedDRA) from parent study.

4. ANALYSIS SETS (POPULATIONS FOR ANALYSIS)

Participant Analysis Set	Description	Applicable Analysis (for additional information refer to Section 6)
Enrolled	All participants who sign the informed consent document and enroll in this open-label extension (OLE) study.	Screening, Disposition, Protocol Deviations
Safety Analysis Set	All enrolled participants who have received at least one dose of study drug in this OLE study. Participants will be analyzed according to the assigned daily maintenance dose on study day 1 in the OLE study.	Demographics, Baseline characteristics, SCD History, Medical history, SCD complications history, Adverse events, Exposure, Prior and Concomitant medications, Labs, Potential Hy's Law cases, VOC, Pharmacodynamic endpoints, Weight, PROs
PK Analysis Set	All enrolled participants who receive at least one dose of study drug and have at least one measurable concentration data point (plasma or whole blood).	Whole blood and plasma concentrations

5. GENERAL METHODOLOGY AND CONVENTIONS

5.1. Definition and Terminology

- Day 1 is the day of first dose of C5351005 study drug administration.

- EOT information is collected on the C5351005 EOT eCRF page. The EOT date from this form will serve as the last dose of study (eg for defining TEAE etc).

- Date of study completion/discontinuation is the study completion/discontinuation date from C5351005 EOS eCRF page. If this date is missing, date of last known assessment will be used.

- Study Day is defined relative to the C5351005 date of first dose of study drug, i.e., Day 1.

For study assessments or events that occur on or after the date of first dose of study drug, the study day of an assessment/event is calculated as:

Study Day = event date – date of first dose + 1.

For pre-treatment events, the study day is calculated as:

Study Day = event date – date of first dose.

- Study Visit is the nominal visit as recorded on the CRF.
- Visit windows rules are defined in Appendix 1. No windowing rules will be applied to PK endpoints where only nominal visits will be used for presentation purposes.
- Treatment of Outliers for Pharmacokinetic Data

Individual blood and plasma concentration-time points, if considered anomalous, may be excluded from the analysis at the discretion of the pharmacokineticist following a review of the available documentation.

Entire individual treatment profiles for any participant, if considered anomalous, may be excluded following review of the available documentation and discussion with the Sponsor. However, results of analysis with and without the excluded profiles may be presented in the CSR.

Any such exclusion or treatment of anomalous PK data, as described above, will be discussed with the Sponsor's Clinical Pharmacologist and clearly described in the CSR along with justification for exclusion.

5.2. Hypotheses and Decision Rules

No formal statistical hypothesis testing will be performed in C5351005 study. All statistical analyses will be used for estimation purposes only and as such no decision rules are required. All p-values will be considered descriptive in nature and will not be used for inference.

5.3. General Methods

Data in general will be summarized as follows unless stated otherwise:

Participants without delayed start				**Participants with delayed start**				**All Participants**			
100 mg	**150 mg**	**200 mg**	**Total**	**100 mg**	**150 mg**	**200 mg**	**Total**	**100 mg**	**150 mg**	**200 mg**	**Total**

- Participants can be classified into two groups: (1) Participants without delayed start (2) Participants with delayed start.

- Participants with delayed start are those with a gap in dosing > 42 consecutive days from completing the originating study (Date of first dose in C5351005 – EOT date from parent study + 1 > 42).

- Participants without a delayed start are those with a gap in dosing ≤ 42 consecutive days from completing the originating study (Date of first dose in C5351005 – EOT date from parent study + 1 ≤ 42 days)

- Dose groups will be defined using the assigned daily maintenance dose in the C5351005 on study day 1 (SD_1 CRF form).

- Total columns will not be presented for PK outputs.

- All figures (unless stated otherwise) will be provided for each group and all participants. In each figure, data will be presented for each dose arm and total.

5.3.1. Analyses for Binary and Categorical Endpoints

Descriptive statistics for binary and categorical variables will be count and percentage and will be presented in the format 'n (%)'. Unless otherwise specified, the percentage in summary tables will be calculated using number of participants in the population (header N) for each dose arm as denominator.

5.3.2. Analyses for Continuous Endpoints

Descriptive statistics [n, mean, standard deviation (SD), median, range (minimum, maximum)] will be provided.

5.4. Methods to Manage Missing Data

5.4.1. Imputation of Incomplete Dates

An incomplete date is any date for which either the day, month or year is unknown, but all three fields are not unknown. An incomplete date occurs when the exact date an event occurred or ended cannot be obtained from a participant.

Any partial dates will be displayed in data listings without imputation of missing days and/or months.

For many of the analyses, a complete date is necessary to determine if the event should be included in the analysis or to establish the duration of an event. In such cases, incomplete dates will be imputed as follows.

5.4.1.1. Start Dates for Prior/Concomitant Medications

For prior/concomitant medications, the case report form permits the start date to have an unknown day and/or month.

- For missing start day only – Day will be imputed as the first day of the month (i.e., 1).

- For missing start day and month – Day and month will be imputed as the first day of the year (i.e., 1 January).

5.4.1.2. Stop Dates for Prior/Concomitant Medications

- For missing stop day only – Day only will be imputed as the last day of the month (i.e., 28, 29, 30, or 31).
- For missing stop day and month – Day and month will be imputed as the last day of the year (i.e., 31 December).

5.4.1.3. VOC start date (for purposes of summarizing parent study VOC history data)

- For missing start day only – Day will be imputed as the first day of the month (i.e., 1).
- For missing start day and month – Day and month will be imputed as the first day of the year (i.e., 1 January).

5.4.1.4. Start Dates for Adverse Events

For this study, there is no need for imputation of start dates of AE to define TEAE and to determine if the AE occurs prior to dosing based on the CRF design.

5.4.1.5. Stop Dates for Adverse Events

AE stop dates will not be imputed.

5.4.2. Imputation of AE data

If AE relationship is missing, relation status will be set to “Related”.

If AE severity is missing, severity will be imputed to “Severe” or the severity noted for the same AE during treatment period.

If AE seriousness is missing, no imputation of seriousness will be performed.

5.4.3. Imputation of Laboratory data

This imputation will follow the CDISC Pfizer safety rulebook. Specifically,

- Data in the form of “>10” or “<10” will be converted to numeric values by adding or subtracting, respectively, a “fuzz” factor of 0.001 (for example, <10 will be converted to 9.999).
- Expressions of the form” >=” or “<=” will be converted to the end point (for example, “>=10” will be converted to 10).

5.4.4. Pharmacokinetic Concentration Below the Limit of Quantification

In all data presentations (except listings), concentrations below the limit of quantification (BLQ) will be set to zero. In listings, BLQ values will be reported as “<LLQ”, where LLQ will be replaced with the value for the lower limit of quantification.

In summary tables, statistics will be presented for a particular dose arm when ≥ 3 evaluable measurements are available for a given timepoint and dose arm. Only minimum and maximum values will be presented when < 3 evaluable measurements are available for a given timepoint and dose arm.

6. ANALYSES AND SUMMARIES

6.1 Primary Endpoint(s)

The safety estimand for the primary endpoints will consider all safety data collected. Specifically, all data collected on or after obtaining informed consent will be included in the analysis.

6.1.1. Incidence of TEAEs

AEs will be mapped to system organ class (SOC) and preferred term using the Medical Dictionary for Regulatory Activities (MedDRA).

The following guidelines will be used to capture the different types of TEAEs based on the AE CRF:

- **Treatment-Related Adverse Events:** adverse events with relationship to study treatment (as recorded on the AE eCRF page, Relationship with study treatment = Related) reported by the investigator and those of unknown relationship (i.e., no answer to the question 'Relationship with study treatment').
- **Serious Adverse Events (SAE):** serious adverse events (as recorded on the AE eCRF page, Was the AE Serious = Yes).
- **Non-Serious Adverse Events:** non-serious adverse events (as recorded on the AE eCRF page, Was the AE Serious = No).
- **Adverse Events Leading to Interruption of Study Treatment**: adverse events leading to interruption of study treatment (as recorded on the AE eCRF page, Action taken with study treatment = Drug interrupted).
- **Adverse Events Leading to Dose Reduction**: adverse events leading to dose reduction (as recorded on the AE eCRF page, Action taken with study treatment = Dose Reduced).
- **Adverse Events Leading to Discontinuation of Study Treatment:** adverse events leading to discontinuation of study treatment (as recorded on the AE eCRF page, Action taken with study treatment = Drug Permanently Discontinued).
- **Adverse Events Leading to Death:** adverse event leading to death (as recorded on the AE eCRF page, Did the AE result in Death = Yes, as well as AEs of Grade 5).

TEAEs leading to study discontinuation will be derived based on the response to the following question in the AE CRF form: "Caused Study Discontinuation"

The overall incidence of TEAEs will be summarized (Section 5.3). This table includes:
TEAEs (number of TEAEs and participants with TEAEs),
Number of participants with:

TEAEs by maximum severity,
Treatment-related TEAE,
Treatment-related TEAE by maximum severity,
Treatment emergent SAE,
Treatment-related treatment emergent SAE,
Treatment emergent non-serious AE,
TEAEs leading to study discontinuation,
TEAEs leading to study treatment discontinuation,

TEAEs leading to study treatment interruption,
TEAEs leading to dose reduction,
TEAEs leading to death.

The TEAEs will be summarized and tabulated at participant level using counts and percentages:

- TEAEs by SOC and PT
- TEAEs by SOC, PT and maximum reported severity
- Treatment-related TEAEs by SOC and PT
- Treatment emergent SAEs by SOC and PT
- Treatment-related treatment emergent SAEs by SOC and PT
- TEAEs leading to study discontinuation by SOC and PT
- TEAEs leading to study treatment discontinuation by SOC and PT
- TEAEs leading to study treatment interruption by SOC and PT
- TEAEs leading to dose reduction by SOC and PT
- TEAEs leading to death by SOC and PT
- Treatment emergent non-serious AEs by SOC and PT

AEs will be sorted in alphabetical order for SOC and then overall descending frequency of PTs within each SOC by all participants.

The frequency of participants who experience each TEAE or treatment-related AE will be determined as follows: A participant experiencing the same AE multiple times will only be counted once for the preferred term. Similarly, if a participant experiences multiple AEs within the same SOC, that participant will be counted only once for that system. If changes in the severity of an AE are recorded in the eCRF, only the most severe incidence of the AE will be counted. If a participant experiences multiple occurrences of a TEAE, only the most severe related event or the worst severity (analysis dependent), will be counted for each participant within each SOC or preferred term for the summaries of treatment-related AEs.

Listing of all AEs will be provided.

A separate listing of deaths, if any, will be provided.

6.1.2. Changes in laboratory assessments

Descriptive statistics (Section 5.3) for each parameter will be presented for baseline, each evaluation post baseline, and change from baseline for each post baseline evaluation.

6.1.3. Changes in vital signs

Descriptive statistics (Section 5.3) for each will be presented for baseline, each evaluation post baseline, and change from baseline for each post baseline evaluation.

6.2. Secondary Endpoint(s)

6.2.1. Annualized rate of VOC

The summary and analyses of on-treatment VOCs will include the following:

- Duration of on-treatment period (weeks)
- Total participant-years

- Total number of VOCs
- Total number of uncomplicated VOCs
- Total number of complicated VOCs and total for each type (ACS, hepatic sequestration, splenic sequestration, priapism)
- Number of participants with VOCs
- Number of VOCs per participant
- Number of uncomplicated VOCs per participant
- Number of complicated VOCs per participant
- Annualized incidence rate of VOC
- Annualized rate of VOC along with 95% CI (using Negative Binomial Model described below)

A listing of VOC data will be provided.

Multiple VOC events will be counted per participant. Both complicated and uncomplicated VOC events will be included for the calculation of the VOC rate.

Annualized incidence rate of VOC on-treatment will be calculated at the group level using the formula below:

$$Annualized\ Incidence\ Rate\ of\ VOC\ on-treatment = \frac{sum\ of\ number\ of\ events\ on-treatment}{sum\ of\ participant\ years\ on-treatment}$$

where participant years on-treatment for each participant = (EOT date – date of first dose of study drug+1)/365.25. EOT date is the last known treatment date from the EOT page in CRF.

Annualized rate of VOC on-treatment using Negative Binomial Model

This analysis will not be performed by group but rather by combining all participants. A negative binomial model will be fitted to model the number of VOC events. The response variable is the number of VOC events for each participant. The independent variable is dose arm (100 mg, 150 mg) and the natural log of participant years (at participant level) will be used as the offset to account for the varying lengths of participant time on-treatment or on-study. The annualized rate and corresponding 95% CI will be presented for each dose arm and overall (100 mg + 150 mg).

6.2.2. Change from baseline in hematological laboratory parameters, including hemoglobin, reticulocytes, lactate dehydrogenase, and unconjugated bilirubin.

Descriptive statistics (Section 5.3) for each parameter will be presented at baseline and each evaluation post baseline. Change from baseline for each post baseline evaluation will also be presented.

Mean (SD) for both raw values and change from baseline values will be plotted over time. These figures will be presented for each parameter separately.

6.2.3. Incidence of SCD-related AEs

Treatment-emergent SCD-related AEs will be summarized as follows (additional details in Section 6.1.1).

- Treatment emergent SCD-related AEs by SOC and PT
- Treatment-related treatment emergent SCD-related AEs by SOC and PT
- Treatment emergent SCD-related Serious AEs by SOC and PT
- Treatment-related treatment emergent SCD-related Serious AEs by SOC and PT

6.3. Exploratory Endpoints

6.3.1. Change from baseline in adhesion of whole blood to microfluidic channels.

Descriptive statistics (Section 5.3) will be presented at baseline and each evaluation post baseline. Change from baseline and percent change from baseline for each post baseline evaluation will also be presented.

Mean (SD) of raw observed values will be plotted over time separately for the following parameters:

- result from whole blood flow adhesion on Vascular Cell Adhesion Molecules (VCAM) (cells/mm^2)
- result from whole blood flow adhesion on P-selectin (cells/mm^2).

6.3.2. Change from baseline in RBC sickling from peripheral blood smears.

Descriptive statistics (Section 5.3) will be presented at baseline and each evaluation post baseline. Change from baseline and percent change from baseline for each post baseline evaluation will also be presented.

For baseline, the first available record within the nominal visit (Day 1) will be used. All other records from this nominal visit will not be used in analysis. For post-baseline records, the average of all assessments that belong to same nominal visit will be used.

6.3.3. Change from baseline in RBC deformability.

Descriptive statistics (Section 5.3) will be presented at baseline and each evaluation post baseline. Change from baseline and percent change from baseline for each post baseline evaluation will also be presented.

For baseline, the first available record within the nominal visit (Day 1) will be used. All other records from this nominal visit will not be used in analysis. For post-baseline records, the average of all assessments that belong to same nominal visit will be used.

Mean (SD) of raw observed values will be plotted over time for the following parameters: PoS, EI_{max} (oxyscan only), EI_{min}.

6.3.4. Change from baseline in erythropoietin levels.

Descriptive statistics (Section 5.3) of baseline, each evaluation post baseline and change from baseline will be provided for observed and log-transformed erythropoietin values, respectively.

Mean (SD) for both log-transformed erythropoietin values and corresponding change from baseline values will be plotted over time.

6.3.5. Patient-Reported Outcome Endpoints

- **PROMIS SF Fatigue 13a, PROMIS SF Pain Interference 8a and PROMIS NRS Pain intensity**

  Descriptive statistics (Section 5.3) of the total raw score at each visit will be provided for each PRO. Total raw scores will be derived for PROMIS SF Fatigue 13a and PROMIS SF Pain Interference 8a.

  PROMIS-1a will also be summarized categorically (Section 5.3) using n (%) under the following categories: 0 = None, 1-3 = Mild, 4-6 = Moderate, 7-10 = Severe.

  All questions must be answered in order to arrive at the raw total score.

- **PGI-S, PGI-S of fatigue, and PGI-S of pain**

  Data collected under different forms will be summarized separately. Summaries will include categorical frequency distribution (Section 5.3) over time.

- **CGI-S**
  Summaries will include categorical frequency distribution and continuous descriptive summary statistics (Section 5.3) at each visit.

  "Not assessed" category will be assigned as missing for continuous descriptive summary.

- **EuroQol EQ-5D-5L**

  Summary of VAS score: Descriptive statistics (Section 5.3) will be provided at each visit for the VAS score.

  Summary of the 5 dimensions: For each dimension, the number and percentage of participants in each of the 5 response levels will be presented.

  Summary of EQ-5D-5L Index: Descriptive statistics (Section 5.3) will be provided at each visit for the index value.

- **WPAI+CIQ:SCD**

  - Number of participants employed, total number of hours missed from work because of problems associated with SCD, total number of hours missed from work due to other reasons and total number of hours worked will be summarized descriptively (Section 5.3) by visit.

  - Descriptive summary statistics (Section 5.3) for the observed SCD work productivity score will be presented at each visit.

- Number of participants who attend classes in an academic setting, total numbers of hours missed from class/school because of problems associated with SCD, total numbers of hours actually attended class/school will be summarized descriptively (Section 5.3) by visit.

- Descriptive summary statistics (Section 5.3) for the observed SCD regular daily activity score will be presented at each visit.

- Descriptive summary statistics (Section 5.3) for the observed SCD school productivity score will be presented.

6.3.6. Pharmacokinetic Concentration Data

Whole blood and plasms concentrations of osivelotor will be listed using the safety analysis set and summarized using the PK analysis set for each time point and visit. Summaries will include n, arithmetic mean, SD, coefficient of variation (CV) %, geometric mean (GM), geometric CV %, median, minimum and maximum.

In summary tables, concentrations below the limit of quantification (BLQ) will be set to zero. In listings BLQ values will be reported as "<LLQ", where LLQ will be replaced with the value for the lower limit of quantification.

In summary tables, statistics will be presented for a particular dose when ≥ 3 evaluable measurements are available for a given timepoint and dose group. Only minimum and maximum values will be presented when < 3 evaluable measurements are available for a given timepoint and dose group.

6.4. Subset Analyses

No subset analyses are planned for this study.

6.5. Baseline and Other Summaries and Analyses

6.5.1. Baseline Summaries

Demographic data, baseline characteristics, SCD history and VOC history (overall and by dose group) will be summarized using descriptive statistics (Section 5.3).

Historical SCD complications and concurrent SCD complications will be summarized.

For medical history, counts and percentages will be provided at the participant-level and any multiple occurrences in a participant for any given term will be counted only once.

6.5.2. Study Conduct and Participant Disposition

The number of participants screened based on all participants who signed informed consent, screen failures (n (%)) with percentage based on total participants screened and reasons for screen failure (n (%) with percentages based on total number of screen failures) will be summarized for all screened participants only.

The following summaries will be presented for disposition by dose arm and overall:

- Number (%) of participants treated

- Number (%) of participants completing treatment
- Number (%) completing study
- Number (%) of participants who discontinue study or discontinue treatment along with primary reason for discontinuation

Number (%) of participants in each analysis set will be summarized in a separate table.

6.5.3. Protocol Deviations

A summary of major protocol deviations (Section 5.3) as assessed by the study team will be provided.

6.5.4. Study Treatment Exposure

Duration of exposure (in weeks) will be summarized (Section 5.3).

Duration of exposure (in weeks) to study drug is defined as the number of weeks from initiation of study drug to the end of study drug treatment (refer to Section 5.1). If treatment end date is missing, then the data cut-off date will be used.

It is defined as below:

$$\textbf{Duration of exposure (weeks)} = \frac{(Date\ of\ last\ dose - Date\ of\ first\ dose + 1)}{7}$$

6.5.5. Prior and concomitant therapy

Concomitant medications will be coded using the WHO Drug Dictionary.

Concomitant medications are those medications taken on or after the initiation of study drug in C5351005 study. This definition includes medications started prior to the initiation of study drug including parent study but continue concurrently with study drug.

Prior medications are those medications taken prior to the initiation of study drug in the parent study.

Prior and Concomitant medications will be summarized descriptively (Section 5.3).

6.5.6. Potential Hy's Law

A listing of potential Hy's Law cases will be provided. Participants with AST or ALT≥3x ULN and Total Bilirubin ≥2x ULN will be included in this listing.

6.5.7. Weight

Weight (kg) will be listed.

7. INTERIM ANALYSES

Not applicable

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8. REFERENCES

Appendix 1. Data Derivation Details

Appendix 1.1. Definition and Use of Visit Windows in Reporting

Below are the analysis visit windows for different endpoints:

Table 2: Analysis visit windows for participants with delayed start: vital signs

Analysis Visit Time	Target Study Day	Per Protocol Study Day Range	Study Day Range for Statistical Analysis*
Baseline visit	1	[-29, 31]	[**,1]
Week 1	7	[4, 10]	[2, 10]
Week 2	14	[11, 17]	[11, 21]
Week 4	28	[25, 31]	[22, 35]
Week 6	42	[39, 45]	[36, 49]
Week 8	56	[53, 59]	[50, 70]
Week 12	84	[77, 91]	[71, 98]
Week 16	112	[105, 119]	[99, 126]
Week 20	140	[133, 147]	[127, 154]
Week 24	168	[161, 175]	[155, 210]
Week 36	252	[238, 266]	[211, 294]
Week 48	336	[322, 350]	[295, 378]
Week 60	420	[406, 434]	[379, 462]
Week 72	504	[490, 518]	[463, 546]
Week 84	588	[574, 602]	[547, 630]
Every 12 weeks	672	[658, 686]	…
…..			

*If multiple assessments occur within a visit window, then the average of those assessments will be used for summarization and analysis.
** Refer to Appendix 1.2 for baseline derivation rules

Table 3: Analysis visit windows for participants with delayed start: labs (hematology and chemistry)

Analysis Visit Time	Target Study Day	Per Protocol Study Day Range	Study Day Range for Statistical Analysis*
Baseline visit	1	[-29, 31]	[**,1]
Week 2	14	[11, 17]	[2, 21]
Week 4	28	[25, 31]	[22, 35]
Week 6	42	[39, 45]	[36, 49]
Week 8	56	[53, 59]	[50, 70]
Week 12	84	[77, 91]	[71, 98]
Week 16	112	[105, 119]	[99, 126]
Week 20	140	[133, 147]	[127, 154]
Week 24	168	[161, 175]	[155, 210]
Week 36	252	[238, 266]	[211, 294]
Week 48	336	[322, 350]	[295, 378]
Week 60	420	[406, 434]	[379, 462]
Week 72	504	[490, 518]	[463, 546]
Week 84	588	[574, 602]	[547, 630]
Every 12 weeks	672	[658, 686]	…
…..			

*If multiple assessments occur within a visit window, then the average of those assessments will be used for summarization and analysis.
** Refer to Appendix 1.2 for baseline derivation rules

Table 4: Analysis visit windows for participants with delayed start: EPO, pharmacodynamic endpoints and PROs

Analysis Visit Time	Target Study Day	Per Protocol Study Day Range	Study Day Range for Statistical Analysis*
Baseline visit	1	[-29, 31]	[**,1]
Week 12	84	[77, 91]	[42, 126]
Week 24	168	[161, 175]	[127, 210]
Week 36	252	[238, 266]	[211, 294]
Week 48	336	[322, 350]	[295, 378]
Week 60	420	[406, 434]	[379, 462]
Week 72	504	[490, 518]	[463, 546]
Week 84	588	[574, 602]	[547, 630]
Every 12 weeks	672	[658, 686]	…
…..			

Pharmacodynamic endpoints include RBC deformability, flow adhesion and blood smear.
*If multiple assessments occur within a visit window, then the average of those assessments will be used for summarization and analysis. For PROs and pharmacodynamic endpoints, if multiple assessments occur within a visit window, the assessment before or on the target study day will be used for summarization and analysis.
** Refer to Appendix 1.2 for baseline derivation rules

Table 5: Analysis visit windows for participants without delayed start: vital signs, EPO, pharmacodynamic endpoints, PROs, labs (hematology and chemistry)

Analysis Visit Time	Target Study Day	Per Protocol Study Day Range	Study Day Range for Statistical Analysis*
**Baseline visit	NA	NA	NA
Week 12	84	[77, 91]	[42, 126]
Week 24	168	[161, 175]	[127, 210]
Week 36	252	[238, 266]	[211, 294]
Week 48	336	[322, 350]	[295, 378]
Week 60	420	[406, 434]	[379, 462]
Week 72	504	[490, 518]	[463, 546]
Week 84	588	[574, 589]	[547, 630]
Every 12 weeks	672	[658, 686]	…
…			

Pharmacodynamic endpoints include RBC deformability, flow adhesion and blood smear.
*If multiple assessments occur within a visit window, then the average of those assessments will be used for summarization and analysis. For PROs and pharmacodynamic endpoints, if multiple assessments occur within a visit window, the assessment before or on the target study day will be used for summarization and analysis.
** This is taken from the Week 12 EOT analysis visit (See Appendix 1.2) from C5351004 parent study.

Appendix 1.2. Baseline Derivations

Baseline derivation for labs, EPO, pharmacodynamic endpoints, weight and vital signs

1. If the gap between EOT visit (EOT CRF form) from parent study and study day 1 from OLE study is < 7 days (date of first dose of study drug in OLE – EOT visit date from parent study + 1 < 7), use Week 12 analysis visit assessment from the parent study to serve as the baseline. If this assessment is not available, baseline is set to missing.

2. If the above defined gap in #1 is >=7 days, use the latest pre-dose day 1 assessment from OLE study to serve as the baseline.

Appendix 2. List of Abbreviations

Abbreviation	Term
ACR	albumin-to-creatinine ratio
ACS	acute chest syndrome
AE	adverse event
BLQ	below the limit of quantitation
CGI-C	Clinical Global Impression of Change
CGI-S	Clinical Global Impression of Severity
CRF	case report form
CSR	clinical study report
eCRF	electronic case report form
eGFR	estimated glomerular filtration rate
GMR	geometric mean ratio
EImin	elongation index minimum
EImax	elongation index maximum
EOS	End of Study
EPO	erythropoietin
GBT	Global Blood Therapeutics, Inc.
Hb	hemoglobin
HR	heart rate
HU	hydroxyurea
ICF	informed consent form
LDH	lactate dehydrogenase
LLOQ	lower limit of quantitation
MedDRA	Medical Dictionary for Regulatory Activities
NCI CTCAE	National Cancer Institute Common Terminology Criteria for Adverse Events
NRS	numeric rating scale
OLE	open-label extension
PD	pharmacodynamics
PGI-C	Patient Global Impression of Change
PGI-S	Patient Global Impression of Severity
PK	pharmacokinetic(s)
PoS	point of sickling

Abbreviation	Term
PRO	Patient-Reported Outcome
PROMIS	Patient-Reported Outcome Measurement Information System
PT	preferred term
QOL	quality of life
RBC	red blood cell
SAE	serious adverse event
SAP	statistical analysis plan
SCD	sickle cell disease
SD	standard deviation
SF	short form
TEAE	treatment-emergent adverse event
VCAM	vascular cell adhesion molecule
VOC	vaso-occlusive crisis
WHO	World Health Organization
WHODD	World Health Organization Drug Dictionary