Statistical Analysis Plan Amendment 3

Study ID: 218675

Official Title: A Phase 2a, Single Dose, Open-label, Dose Exploration Study to Assess the PK-PD Activity, Safety, and Tolerability of GSK4532990 in Adult. Participants With NASH or Suspected NASH

NCT ID: NCT06104319

Date of Document: 24-Oct-2025

Note: The 'Regulatory Agency Identifier Number' on Page 1 of the document was invalid and has been replaced with a black box.

TITLE PAGE

Information Type:	Statistical Analysis Plan (SAP)
Protocol Title:	A Phase 2a, single dose, open-label, dose exploration study to assess the PK-PD activity, safety, and tolerability of GSK4532990 in adult participants with NASH or suspected NASH
Study Number:	218675
Compound Number:	GSK4532990
Abbreviated Title:	Phase 2a dose exploration Study of GSK4532990 in Participants with NASH or suspected NASH.
Acronym:	SKYLINE

Sponsor Name: GlaxoSmithKline Research & Development Limited

Regulatory Agency Identifier Number(s)

Registry	**ID**
[redacted]	

©2023 GSK group of companies or its licensor.

SAP AMENDMENT SUMMARY OF CHANGES TABLE

DOCUMENT HISTORY		
Document	**Date**	**Document Identifier**
Amendment 03	24-Oct-2025	TBD- 22378172 v2.0
Amendment 02	15-Aug-2025	TMF- 22378172
Amendment 01	23-Sep 2024	TMF- 19989914
Original SAP	15-Jan-2024	TMF- 17428082

Amendment 03:

List of changes and rationale:

List of main changes in the SAP and their rationale:	**Description of Change**	**Brief Rationale**
CCI [redacted]		
4.6.3.1	Added the methodology for imputing non-detectable laboratory values	To pre-specify how to handle non-detectable values recorded as "<x" or ">x".
6.1.2	Added fibrinogen to the baseline characteristics table.	This parameter was added to provide additional information on the baseline characteristics of the study population.
6.2.1	INR moved from the liver enzymes to the haematology section.	INR is a haematology parameter, not a liver function parameter.
6.2.4	Weeks 10 and 14 were removed from the assessment window summary for all treatment arms.	Weeks 10 and 14 were not included in the schedule of activities in the latest protocol version.

TITLE PAGE 1

SAP AMENDMENT SUMMARY OF CHANGES TABLE 2

1. INTRODUCTION 6
   1.1. Objectives, Estimands and Endpoints 6
      1.1.1. Primary Objective Estimands 7
      1.1.2. Secondary Objective Estimands 8
   1.2. Study Design 9

2. STATISTICAL HYPOTHESES 12
   2.1. Primary Objective 12
   2.2. Multiplicity Adjustment 12

3. ANALYSIS SETS 12

4. STATISTICAL ANALYSES 13
   4.1. General Considerations 13
      4.1.1. Baseline Definition 13
      4.1.2. Participant Addition 13
      4.1.3. HSD17B13 protein quality and biopsy replacement 14
      4.1.4. Confidence intervals, statistical significance & summary statistics 14
   4.2. Primary Endpoint(s) Analyses 15
      4.2.1. Definition of primary estimands 15
      4.2.2. Main analytical and modelling approach 15
      4.2.3. Data exclusion and Sensitivity analyses 25
   4.3. Secondary Endpoints Analyses 25
      4.3.1. Secondary endpoints 25
      4.3.2. Main analytical approach 26
   CCI [redacted] 27
   27
   31
   4.5. Supportive information, and metrics 32
   4.6. Safety Analyses 33
      4.6.1. Duration of Study 33
      4.6.2. Adverse Events 33
      4.6.3. Additional Safety Assessments (if applicable) 35
   4.7. Other Analyses 37
      4.7.1. Subgroup analyses 37
      4.7.2. Other variables and/or parameters 37
      4.7.3. Analyses to Support Regional Submission 38
   CCI [redacted] 38
   4.9. Changes to Protocol Defined Analyses 40

5. SAMPLE SIZE DETERMINATION 41

6. SUPPORTING DOCUMENTATION 42
   6.1. Appendix 1 Study Population Analyses 42

6.1.1. Participant Disposition 42
6.1.2. Demographic and Baseline Characteristics 43
6.1.3. Protocol Deviations 44
6.1.4. Prior and Concomitant Medications 45
6.1.5. Prohibited Medications 45
6.2. Appendix 2 Data Derivations Rule 45
6.2.1. Criteria for Potential Clinical Importance 45
6.2.2. Study Period 47
6.2.3. Study Day and Reference Dates 47
6.2.4. Assessment Window 48
6.2.5. Multiple measurements at One Analysis Time Point 49
6.2.6. Handling of Partial Dates 50
6.2.7. Trademarks 52

7. REFERENCES 53

LIST OF TABLES

PAGE

Table 1, Summary of study objectives and endpoints. 6

Table 2, Intercurrent events and strategy used for primary endpoints. 8

Table 3, Intercurrent events and strategy used for secondary endpoints. 9

Table 4, Exploratory biomarker analysis summary. 28

Table 5, Summary of changes to protocol defined analyses plan. 40

Table 6, Summary of Baseline characteristics including demographics. 43

Table 7, Summary of parameters and criteria used to determine potential clinical importance. 45

Table 8, Analysis window ranges per visit and treatment arm. 48

1. INTRODUCTION

The purpose of this SAP is to describe the planned analyses to be included in the CSR for Study 218675. Details of the planned interim analysis, as well as the final analyses, are provided.

Additional details with regards to data handling conventions and the specification of data displays will be provided in the Output and Programming Specification (OPS) document. Participant-level data will be available interactively via RAPIDO Data Viewer at Statistical Analyses Complete (SAC).

This is a Phase 2a, single dose, open-label, multi-center, dose exploration study designed to assess the PK and PD activity, safety, and tolerability of GSK4532990 in adult participants with confirmed or suspected NASH. This study will further inform the dose exposure-target engagement relationship and potentially inform higher doses or alternate dosing regimen decisions in future clinical development of GSK4532990.

1.1. Objectives, Estimands and Endpoints

Table 1, Summary of study objectives and endpoints.

Objective(s)	Endpoint(s)
Primary	
• To assess the effect of a single dose of GSK4532990 on PK-PD target engagement in participants with NASH or suspected NASH	Dual endpoints: • Pre and post dose liver biopsy-derived HSD17B13 protein expression levels • Pre and post dose liver biopsy-derived HSD17B13 mRNA expression levels
Secondary	
• To evaluate GSK4532990 exposure parameters after a single dose in participants with NASH or suspected NASH	Plasma exposure parameters of GSK4532990: • Area under the concentration-time curve from time zero (pre-dose) to the last quantifiable concentration (AUC0-t) • Maximum observed concentration (Cmax)
• To evaluate the PD effect of GSK4532990 after a single dose in participants with NASH or suspected NASH	• Change from baseline in the observed HSD17B13 protein expression levels at each assigned post-dose biopsy time: Weeks 8, 12, or 28 • Change from baseline in the observed HSD17B13 mRNA expression levels at each assigned post-dose biopsy time: Weeks 8, 12, or 28

Objective(s)	Endpoint(s)
Safety	
• To assess the safety and tolerability of GSK4532990 after a single dose in participants with NASH or suspected NASH	Clinical assessments including: • Occurrence of AEs and SAEs • Change from baseline in vital signs at each visit assessment • Change from baseline in key laboratory measurements at each visit assessment • Occurrence of clinically significant ECG findings at each assessment
Exploratory	
CCI [redacted]	

1.1.1. Primary Objective Estimands

The primary study objective estimands will be assessed using the following attributes:

- Population: Adult participants with proven or suspected NASH

- Treatment: GSK4532990 – doses (mg): 25, 200, 400 and 800 mg

- Variables (dual endpoints):
  - Pre and post dose liver biopsy-derived HSD17B13 protein expression levels
  - Pre and post dose liver biopsy-derived HSD17B13 mRNA expression levels

- Population level summary measure
  - Predicted mean and 95% CI of percent change from baseline of HSD17B13 protein derived from the PK-PD target engagement model at each treatment arm and post-dose biopsy time
  - Predicted mean and 95% CI of percent change from baseline of HSD17B13 mRNA derived from the PK-PD target engagement model at each treatment arm and post-dose biopsy time

The intercurrent events and approach for managing intercurrent events for the primary objectives are described in the following table.

Table 2, Intercurrent events and strategy used for primary endpoints.

Intercurrent Events	Primary Estimands
Use of prohibited medication as defined in sections 5.1 & 5.2 of the protocol	Addressed with "Treatment policy" strategy, i.e., use available data regardless of use of prohibited medication
Increase in self-reported alcohol consumption which exceeds eligibility criteria as defined in the protocol	Addressed with "Treatment policy" strategy, i.e., use available data regardless of increased alcohol consumption
Delays in post-dose biopsy times for any reason	Addressed with "Treatment policy" strategy, i.e., data will be used even if the biopsy was outside the time window

The rationale for the primary estimands is that we are interested in estimating the mean and 95% CI of percent change from baseline based on the PK-PD target engagement model regardless of the use of prohibited medication, increased self-reported alcohol consumption or delays in post-dose biopsy times to reflect the real-world setting.

1.1.2. Secondary Objective Estimands

The secondary study objective estimands will be assessed using the following attributes:

- Population: Adult participants with proven or suspected NASH

- Treatment: GSK4532990 – doses (mg): 25, 200, 400 and 800 mg

- Variables and population summary measures:
  - As described in 4.3.1 and 4.3.2

The intercurrent events and approach for managing intercurrent events for the secondary objectives are described in the following table.

Table 3, Intercurrent events and strategy used for secondary endpoints.

Intercurrent Events	Secondary Endpoint Estimands	Additional Secondary Endpoint Estimands
Use of prohibited medication as defined in sections 5.1 & 5.2 of the protocol	Addressed with "Treatment policy" strategy, i.e., use available data regardless of use of prohibited medication	Addressed with "Whilst not on prohibited medication" strategy, i.e., use available data up until the use of prohibited medication
Increase in self-reported alcohol consumption which exceeds eligibility criteria as defined in the protocol	Addressed with "Treatment policy" strategy, i.e., use available data regardless of increased alcohol consumption	Addressed with "Whilst not exceeding the weekly limit of alcohol" strategy, i.e., use available data up until the week of increased alcohol consumption
Delays in post-dose biopsy times for any reason	Addressed with "Treatment policy" strategy, i.e., data will be used even if the biopsy was outside the time window	

For safety estimands, intercurrent events will be addressed with a "Treatment Policy" strategy, i.e., regardless of the intercurrent event occurring.

1.2. Study Design

Overview of Study Design and Key Features	
Design Features	Phase 2a multicentre study, single dose, open-label, four arm, dose-exploration study to assess the PK-PD activity, safety, and tolerability of GSK4532990 in adult participants with NASH or suspected NASH. The study consists of 3 periods: The screening and pre-dose, the intervention and intensive PK and the follow up period.

<table>
<tr><th colspan="2">Overview of Study Design and Key Features</th></tr>
<tr><td></td><td>
<u>Screening Period:</u> Participants will attend two screening visits to assess eligibility as described below:
• Screening 1 (Week -6 from dosing): Assessments to be performed as described in the schedule of assessments (SoA).
• Screening 2 (Week -2 from dosing): Assessments to be performed as described in the SoA.
• Pre-dose assessments (Day -1 to Day 1 from dosing): Assessments including liver biopsy to be performed as described in the SoA.

• <u>Intervention and intense PK period:</u> Day one through day three. Eligible participants will be enrolled into different cohorts to receive a dose of GSK4532990 following a sequence of 9 enrolment schedules with the schedules 2, 3, 6 and 7 to allow for randomisation. The study is designed to allocate/randomise 12 patients in each of the four treatment arms split equally into 6 patients with early and 6 patients with late biopsy time.

<u>Follow-up Period:</u> Week two through week eighteen for the 25, 200 and 400 mg treatment arms and week two through week thirty for treatment the 800 mg treatment arm. During the follow up period the participants will have several assessments and tests and a second liver biopsy either at 8, 12 or 28-weeks post-dose.
</td></tr>
<tr><td>Study intervention</td><td>
After a screening period of 6 weeks (4 to 10 weeks window), eligible participants will be enrolled in the study and followed up for up to 30 weeks after receiving a single dose of the intervention.

<u>Concomitant therapy:</u>

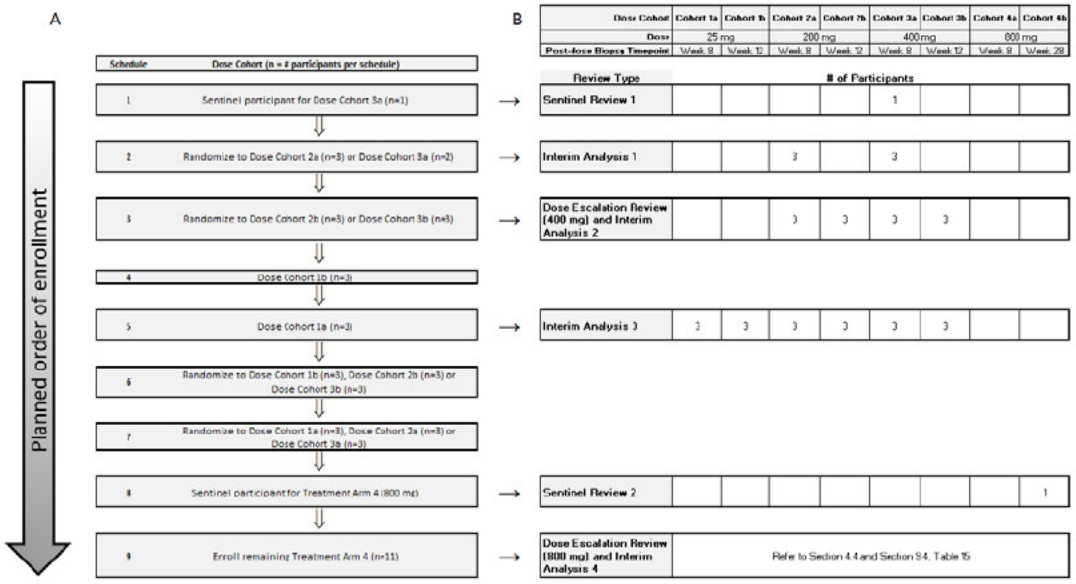
Any medication or vaccine (including over the counter or prescription medicines, recreational drugs, vitamins, and/or herbal supplements) or other specific categories of interest that the participant is receiving at the time of enrolment or receives during the study must be recorded along with: reason for use, dates of administration including start and end dates and dosage information including dose and frequency.
</td></tr>
<tr><td>Study intervention Assignment</td><td>
The details of the four treatment arms split into eight dose cohorts are provided below:

<table>
<tr><td>Arm Title</td><td colspan="2">GSK4532990 25 mg</td><td colspan="2">GSK4532990 200 mg</td><td colspan="2">GSK4532990 400mg</td><td colspan="2">GSK4532990 800mg</td></tr>
<tr><td>Arm Type</td><td colspan="2">1 Experimental</td><td colspan="2">2 Experimental</td><td colspan="2">3 Experimental</td><td colspan="2">4 Experimental</td></tr>
<tr><td>Arm Description</td><td colspan="2">Single dose of 0.125 mL of GSK4532990 200 mg/mL</td><td colspan="2">Single dose of 1.0 mL of GSK4532990 200 mg/mL</td><td colspan="2">Single dose of 2.0 mL of GSK4532990 200 mg/mL</td><td colspan="2">Single dose of 4.0 mL of GSK4532990 200 mg/mL</td></tr>
<tr><td>Biopsy time</td><td>Week 8</td><td>Week 12</td><td>Week 8</td><td>Week 12</td><td>Week 8</td><td>Week 12</td><td>Week 8</td><td>Week 28 (per IA2/3 iDRC guideline)</td></tr>
</table>
</td></tr>
</table>

Overview of Study Design and Key Features

The details of the enrolments schedules and randomisation are provided in the figure below. Once a schedule has been enrolled, the next commences. Rationale for the order of the dose cohorts and number of participants assigned to each schedule is due to prioritized timing of specific data for internal development planning and decision-making and optimization of timing for study enrolment and completion.

The study design includes a form of randomisation where possible. As shown in the following figure, randomisation is available in four schedules for 29 subjects. In each schedule with randomisation, the subjects are randomised in two or three out of the eight available cohorts.



CCI

[redacted]

Overview of Study Design and Key Features	
	CCI [redacted]

2. STATISTICAL HYPOTHESES

2.1. Primary Objective

The primary objective of the statistical analysis of the study is to predict the mean and 95% CI of percent change from baseline of HSD17B13 protein and mRNA expression levels, derived from the PK-PD target engagement model at each treatment arm and post-dose biopsy time. Estimating the mean change from the target engagement model does not feature hypothesis testing.

2.2. Multiplicity Adjustment

Given there are no planned hypothesis tests, there are no plans for multiplicity adjustment.

3. ANALYSIS SETS

Analysis Set	Definition / Criteria	Analyses Evaluated
Screened	• All participants who were screened for eligibility	• Study Population
Enrolled	• All participants who entered the study (who were randomized or received study intervention or underwent a post-screening procedure) • NOTE: screening failures (who never passed screening even if rescreened) and participants screened but never enrolled into the study (met eligibility but not needed) are excluded from the Enrolled Analysis set as they did not enter the study.	• Study Population

Analysis Set	Definition / Criteria	Analyses Evaluated
Safety/Exposed/Biomarkers	• All participants who received single dose of study intervention.	• Safety/Biomarkers
HSD17B13 Protein Concentration Analysis Set	• All participants in the Enrolled analysis set with evaluable* baseline biopsy protein assessment.	• PK/PD target engagement
HSD17B13 mRNA Concentration Analysis Set	• All participants in the Enrolled analysis set with evaluable* baseline biopsy mRNA assessment.	• PK/PD target engagement
Pharmacokinetic (PK)	• All participants in the Safety analysis set who had at least 1 evaluable PK assessment (non-quantifiable values will be considered as evaluable values).	• PK

*A biopsy assessment is considered evaluable when the results are available and above lower limit of quantification (LLOQ)

4. STATISTICAL ANALYSES

4.1. General Considerations

4.1.1. Baseline Definition

For all endpoints the baseline value will be the latest pre-dose assessment with an evaluable result, including those from unscheduled visits. If time is not collected, Day 1 will be treated as the baseline if additional information is available suggesting that the assessment took place before dosing. Unless otherwise stated, if baseline data is missing no derivation will be performed and baseline will be set to missing. For ALT and AST specifically, baseline will be set as the average of the two latest pre-dose observations (scheduled and unscheduled) with interval >=14 days. If there are fewer than 2 pre-dose observations with interval >=14 days, the latest evaluable pre-dose observation is considered as baseline.

4.1.2. Participant Addition

In the event that one or more participants within a dose cohort withdraws prior to the pre- or post-dose biopsy or a biopsy sample is not evaluable, additional participants with a unique study ID may be enrolled. This will ensure that each treatment arm has

a sufficient number of participants with evaluable biopsies for both pre- and post-dose biopsy time points in the study. Participants requiring more time until their post-dose biopsy and their last visit, will be prioritised, preferably, in a manner following the order of recruitment. A post-dose biopsy will not be required from participants with a non-evaluable pre-dose biopsy; these participants will be candidates for replacement as assessing HSD17B13 knockdown from the post-dose biopsy will not be possible with a non- evaluable pre-dose biopsy. To replace biopsies as needed, new participants will be added to the appropriate dose cohort by forced treatment allocation. The rationale for forced treatment allocation includes the small sample size per dose cohort, limiting exposure due to the invasive nature of biopsy, and the lack of existing safety exposure on doses above 200 mg. The recruitment of additional participants may take place when the study team identifies that a dose cohort is at risk of not reaching the minimum required number of 5 evaluable participants.



4.1.4. Confidence intervals, statistical significance & summary statistics

Confidence intervals (CI) will use 95% confidence levels unless otherwise specified. The alpha level for all the tests is at two sided 0.05 unless otherwise specified.

Unless otherwise specified, continuous data will be summarized using descriptive statistics: n, mean, standard deviation (std), median, minimum, and maximum. Categorical data will be summarized as the number and percentage of participants in each category.

4.2. Primary Endpoint(s) Analyses

4.2.1. Definition of primary estimands

The primary clinical questions of interest are:

- What is the predicted mean percent change from baseline of HSD17B13 protein levels, derived from the PK-PD target engagement model for each treatment arm in adult participants with proven or suspected NASH treated with GSK4532990 regardless of prohibited medication, increase in self-reported alcohol consumption, or delays in post-dose biopsy times?

- What is the predicted mean percent change from baseline of HSD17B13 mRNA levels, derived from the PK-PD target engagement model for each treatment arm in adult participants with proven or suspected NASH treated with GSK4532990 regardless of prohibited medication, increase in self-reported alcohol consumption, or delays in post-dose biopsy times?

Data following intercurrent events (use of of prohibited medication, increase in self-reported alcohol consumption and delays in post-dose biopsy) will be included in the primary estimand analysis.

The summary measures for the primary endpoints will be the model predicted mean percent change from baseline of HSD17B13 mRNA and protein derived from the PK-PD target engagement model at each treatment arm and post-dose biopsy time along with 95% C.I.

4.2.2. Main analytical and modelling approach

4.2.2.1. Summary of major steps

The data analysis will consist of the following major steps:

A base target engagement model will be developed without covariates.

- Covariates to the base target engagement model will be evaluated and incorporated as appropriate.

Target Engagement model development

The starting point of the GSK4532990 target engagement analysis will be based on the base mixed-effect model from the previous analysis including the corresponding

fixed effects and random effects parameters. This starter model will be evaluated and refined with all data available at final analysis to build the base structural model.

A population PK/PD model will be developed to describe the baseline and post dose changes in hepatic HSD17B13 protein and/or mRNA concentrations as a result of GSK4532990 at multiple timepoints and after different dose levels. Model development will be a sequential 2 step process. In the first step, PK data available on the PK analysis set will be used to build a population PK model with the aim of obtaining the empirical Bayes estimates (EBEs) of the individual PK parameters (apparent clearance, apparent volume of distribution, absorption rate constants, etc.). As a second step, the PK/PD model will be developed by fixing the EBEs of the PK section. Details about the population PK model development will be described on a separate modelling report.

The initial PK/PD structural model will test the inclusion of an effect compartment given the disconnect between the rapid systemic half-life of GSK4532990 and the delayed and sustained reduction of HSD17B13. Additional model structures might be tested to include a more mechanistic rationale of the temporal disconnect between PK and PD. An indirect response model will be tested to characterize the mechanism of GSK4532990 inhibiting translation (i.e. synthesis) of hepatic HSD17B13 as follows:

$$\frac{dHSD17B13}{dt} = K_{in} \bullet (1 - ER_{ij}) - HSD17B13 \bullet K_{out}$$

$$ER_{ij} = \frac{Imax \bullet Conc_{ij}}{IC_{50} + Conc_{ij}}$$

Where K_{in} is the zero-order synthesis rate of HSD17B13, K_{out} is the first-order elimination rate constant, ER_{ij} is an inhibitory E_{max} model to describe GSK4532990 effect in K_{in} reduction for subject i at time point j, Imax is the maximal decrease in K_{in}, and IC_{50} is the GSK4532990 concentration that produces half of maximal decrease in K_{in}. Finally, $Conc_{ij}$ is the GSK4532990 concentration in the site of action (e.g. effect compartment) for subject i at timepoint $_j$. The amount of hepatic HSD17B13 at baseline is equal to the ratio of K_{in} and K_{out}.

The fixed and random effects model parameters will be estimated for the base model. As a first step, inter-individual variability (IIV) will be included on all structural parameters; however, the number of random IIV parameters (η_P) may be reduced to ensure that the model minimizes successfully, and all parameters are adequately estimated.

If an inhibitory E_{max} model does not fit the data, other models will be evaluated as applicable (e.g. linear, power, etc.).

Modelling of inter-individual variability

Inter-individual variability in model parameters will be regarded as random quantities and will be modelled in terms of eta (η) variables. The etas across individuals (η_{i_P}) for each model parameter (P) are assumed to have a normal distribution with a mean of zero and a variance ($\omega_p{}^2$) which can be estimated. While the distribution of η is assumed to be normal, the distribution of P will depend on the mathematical expression relating the two.

For the population PK/PDmodeling activities, the distribution of P generally follows a log-normal distribution, and the mathematical equation is described as follows:

$$P_i = P_{TV} \cdot e^{\eta_{i_P}}$$

where: P_i is the parameter estimate for the ith individual, P_{TV} is the typical population estimate for parameter Pand η_{i_P} is the individual deviation from the population estimate.

The following equation converts the variance to a coefficient of variation (CV) for each P_{TV} in the original scale:

$$CV\ (\%) = \sqrt{e^{\omega_p{}^2} - 1} \cdot 100$$

An alternative additive form of IIV may be considered when P_i is known or suspected to come from a normal distribution:

$$P_i = P_{TV} + \eta_{i_P}$$

In such cases the inter-individual CV for P_{TV} will be computed as $\omega_P\ \omega_p/P_{TV}$where ω_p represent the square root of ω_p^2.

Shrinkage in each η_P (sh_{η_P} sh_{η_P}) will be calculated and reported for IIV parameters using:

$$sh_{\eta_P} = 1 - \frac{SD(EBE\eta_P)}{\omega_p}$$

where:

SD(EBE η_P) is the standard deviation of the *empirical Bayesian estimates (EBE)* of η_{i_P}.

The random effects for the parameters will be modeled according to a multivariate normal distribution (MVN), with mean value 0:

$$(\eta_1, \ldots, \eta_n) \sim \text{MVN}(0, \Omega)$$

in which Ω is the variance-covariance matrix of the random effects components η. Ω for all parameters with modelled IIV will first take the diagonal form:

$$\Omega = \begin{bmatrix} \omega_{P_1}^2 & & & \\ & \omega_{P_2}^2 & & \\ & & \ddots & \\ & & & \omega_{P_n}^2 \end{bmatrix}$$

where:

ω_{P1}^2 ω_{P1}^2, ω_{P2}^2, and ω_{Pn}^2 ω_{Pn}^2 represent the variances of the η_{i_P} associated with parameters P_1, P_2, through P_n, respectively.

After accounting for the influence of covariates, the covariance between pairs of random IIV parameters (*e.g.*, η_{i_P1} and η_{i_P2} for parameters P_1 and P_2, respectively) will be examined graphically by using pairs plot, and in case plausible correlations were found, its significance in the model would be checked by estimating the corresponding off-diagonal element(s). :

The inclusion of off-diagonal elements in Ω in the final model will depend on the numerical stability of the resulting model and its influence on goodness of fit plots.

Modelling residual variability

Residual variability accounts for the differences between observed data and the individual model predictions of the dependent variable .
The residual variability will be assumed to be a function of normally distributed random effects described in terms of epsilon (ε) with mean 0 and a variance σ^2:

$$(\varepsilon_1, \ldots, \varepsilon_n) \sim \text{MVN}(0, \Sigma)$$

in which Σ is the variance-covariance matrix of the residual variability components. Typically, any off-diagonal elements of this matrix will be assumed to be zero.
Models for residual error will generally take one of the following forms:
The additive error model, where the variance is constant:

$$Y_{obs,ij} = Y_{pred,ij} + \varepsilon_{1_{ij}}$$

The proportional error model, where the variance is proportional to the squared prediction:

$$Y_{obs,ij} = Y_{pred,ij} \cdot (1 + \varepsilon_{1_{ij}})$$

The combination of additive and multiplicative error model:

$$Y_{obs,ij} = Y_{pred,ij} \cdot (1 + \varepsilon_{1_{ij}}) + \varepsilon_{2_{ij}}$$

where: $Y_{obs,ij}$ is the jth observed value of the dependent variable in the ith individual $Y_{pred,ij}$ is the jth predicted value of Y in the ith individual ε_{1ij} and ε_{2ij} are independent, random variables describing the difference between $Y_{obs,ij}$ and $Y_{pred,ij}$ (both with a mean of zero and variances of σ_1^2 and σ_2^2 σ_2^2, respectively).

A proportional error model will initially be used to describe the residual variability. If the goodness-of-fit (GoF) plots reveal potential biases in the residual variability model, other residual error models will be considered as appropriate. Since the mRNA and protein data only include two timepoints per individual (pre and post-dose), the residual variability may be fixed at 10% if the data does not allow differentiation between IIV and residual error.

The square root of the residual error variance (σ) is expressed as the dependent variable units in an additive error model and as a coefficient of variation in a proportional error model for untransformed data. In some instances, it may be useful to fix the variance estimate of ε_1 (σ_1^2 σ_1^2) to a value of one and estimate CV directly as fixed effects $\left(\theta_{CV} \cdot \varepsilon_{1_{ij}}\right)$ If examination of individual fits and residual and inter-individual errors suggest that individual parameter estimates are unduly influenced by the exclusion of BLQ samples, consideration will be given to including these samples in the analysis dataset and M3 or M4 methods (Ahn et al. 2008) evaluated (for proportional error models or truncated normal distributions, respectively) to determine whether they provide more accurate individual parameter estimates as measured by the IIV for estimated η_P.

Estimation methods

First Order Conditional Estimation with Interaction (FOCEI) will be the preferred method of parameter estimation. Alternative methods such as Iterative Two Stage (ITS), Monte Carlo Importance Sampling EM (IMP), Monte Carlo Importance Sampling Expectation Maximization Assisted by Mode a Posteriori (MAP), and Stochastic Approximation Expectation Maximization (SAEM), may be applied if FOCEI fails to converge on reliable parameter estimates. ITS might be used to obtain initial estimates for FOCEI or SAEM if run times are an issue with these algorithms. "Mu Referencing" (Beal et al. 2009) will be used to improve the efficiency of computations when ITS, IMP, MAP, and SAEM methods are used. If the Beal M3 method needs to be applied to account for post-dose BLQ samples, the Laplacian estimation method will be used.

Covariate selection

Once the structural population PK/PD model has been identified, the predictive value of individual patient characteristics will be assessed as structural covariates. Structural covariates are major predictors for the variability in model parameters. Structural

covariates will be retained in the PK/PD model if they are statistically significant and provide biologically reasonable parameter estimates.

Graphical and statistical approaches along with consideration of the underlying scientific rationale will be used to identify which covariates will be examined on target engagement parameters and to assess the mathematical form of their relationships. The covariate effects will be considered for the model IC50 and may be explored for a limited number of other parameters deemed appropriate, depending upon the physiological plausibility. A non-exhaustive list of prospectively identified covariates will be evaluated for possible inclusion in the model.

CCI



Baseline covariates will be obtained from observations on the first day of dosing or at screening if this value is not available. For categorical covariates, each category should be represented by at least 10% of the population in order to be evaluated. Covariates with low representation (less than 10% of the population) that are not included in the initial full model may be tested in the semi-final models as exploratory covariates (to estimate trends rather than to provide precise parameter estimates). Correlated covariates may be tested in alternative semi-final models to provide the best description of the data.

Candidate covariates will only be tested for statistical significance in improving the model fit if they met one or more of the following criteria:

- plots of individual estimates (η_{i_P}) *versus* covariates demonstrate a trend;

- a statistically significant covariate effect is determined by univariate analysis of variance or regression analysis (for categorical and continuous covariates, respectively);
- physiological or pharmacological rationale; or
- information from prior analyses or published sources.

Covariate correlations will be assessed to detect confounding. If co-linearity is identified, only the most clinically relevant will be selected. Exploratory plots will be used to detect correlations between EBEs and covariates in the base model, only if ETA shrinkages are acceptable (<30%). Parameters that show excessive (>30%) shrinkage in IIV can be ill suited for graphical assessment of covariate effects.

Continuous covariates (*COV*) will be incorporated using a power model centered at their median values and P_{TV} expressed as:

$$P_{TV} = \theta_p \cdot \left(\frac{COV_i}{M(COV_i)}\right)^{\theta_{COV,P}}$$

where: P_{TV} is as previously defined, θ_P θ_P is the estimated parameter representing the typical value of model parameter *P* when the individual covariate (COV_i) is equal to the median of the covariate in the analysis population ($M(COV_i)$) , and $\theta_{COV,P}$ $\theta_{COV,P}$ θ_P is the estimated parameter representing the influence of covariate *COV* on model parameter *P*.

Alternative expressions may be considered for continuous covariates based on trends that are observed in covariate plots.

Categorical covariates (*CAT*) will be tested and incorporated in the model as a series of index variables taking on values of zero or one (*e.g.*, CAT_1, CAT_2, …, CAT_{n-1} representing the n-1 levels of *CAT*). Index variables will be included in the model as follows:

$$P_{TV} = \theta_P \cdot \prod_{i=1}^{n-1} (\theta_{CAT_{i,P}})^{CAT_i}$$

where: P_{TV} is as previously defined, θ_P θ_P θ_P is the estimated parameter representing the typical value of model parameter *P* for a reference category when all the individual categorical covariate index variables (CAT_i) are equal to zero, and $\theta_{CAT_1,P}$ $\theta_{CAT_{i,P}}$ is the estimated parameter representing the relative influence of a categorical covariate index variable on model parameter *P* when CAT_i is equal to one.

Alternative expressions may be considered for categorical covariates to facilitate the interpretation of the typical parameter estimates with respect to specific patient categories.

The full model with backwards deletion approach or the automated forward inclusion followed by backward elimination procedure will be the preferred methods for covariate modelling. In the full model approach, all covariate parameter relationships of interest will be entered in the model simultaneously. A backwards deletion will be carried out at the p=0.001 (increased objective function value [OFV] less than 10.83 points, d.f.=1) significance level where the relative influence of each covariate on the model will be reevaluated by deleting it from the full model on an individual basis.

On the stepwise covariate modeling approach on the other hand, an automated forward inclusion step will test each candidate covariate one by one and retained if they meet the statistical criteria of p=0.01. Then, a backwards deletion will be carried out at the p=0.001 significance level in a stepwise manner. The model that is the result of the backward elimination step will be considered the final PKPD model.

Where significant covariate effects are identified, assessment of effect magnitude over a relevant range, along with confidence intervals, will be provided.
A covariate may be retained in the final model, despite not meeting the criteria above, if there is a strong pharmacological or physiological rationale for its inclusion.

4.2.2.2. Model evaluation

Model goodness-of-fit

The goodness-of-fit (GoF) for a model will be assessed by a variety of plots and computed metrics. GoF plots may include data points for observed and/or predicted data, reference lines (identity, zero line, etc.), and smooth lines through the data. The GoF plots listed below may be used for graphical model evaluation.

- Plots of population (*PRED*) and individual (*IPRED*) predictions *versus* observations (*DV*) and *versus* time
- Plots of conditional weighted residuals (*CWRES*) *versus* population predictions (*PRED*) and *versus* time
- Histograms and QQ plots of *CWRES*
- Histogram and QQ plots of the etas (η)
- Scatter plots of eta (η) pairs
- Scatter plots of eta (η) *versus* modelled covariates
- Individual plots overlaying observed and population and individual predictions *versus* time

Plots of observed *versus* predicted concentrations will be examined for departures from the line of unity, which may be diagnostic of model misspecification. Plots of weighted residuals versus predicted concentration and time will be examined for homoscedasticity and curvature. Heteroscedasticity can indicate poor performance by the current residual error model while curvature is a sign of model misspecification. Histograms and QQ plots will be examined for evidence of departures from the assumptions of normality. Scatter plots of eta pairs will be reviewed for evidence of correlations and problems with shrinkage and model identifiability. Scatter plots of eta

(η) *versus* modelled covariates will be examined for homoscedasticity and curvature (for continuous covariates). Curvature will be an indication that alternative parameterization of the covariate effect might be useful. Individual plots of observed and predicted concentrations will be examined to assess individual GoF and to identify subjects and observations that may not be well characterized by the model under consideration.
Conditional weighted residuals (*CWRES*) are calculated in NONMEM as described by Hooker, et al.(Hooker, Staatz, and Karlsson 2007) Histograms of *CWRES* and individual plots comparing observed and individual predictions over time may highlight observations that are inconsistent with model predictions and the estimated magnitude of the residual error. Observations for which $|CWRES| > 6$ will be reviewed as potential outliers and may be excluded from the analysis when they are found to undue influence the parameter estimates or numerical stability of the estimation method. All observations excluded from the analysis will be identified and justified in the resulting report.

Model evaluation will also include relative standard errors (RSE) of the parameter estimates. 95% confidence intervals (CI) for each estimated parameter may be constructed based on its RSE. Shrinkage estimates will be examined for each η_P and for ε. Successful minimization and execution of a covariance step will be considered as part of the model evaluation for each run.

Model discrimination

The difference in the objective function value (ΔOFV) between models is proportional to minus twice the log-likelihood of the model fit to the data and will be used to compare competing hierarchical models (e.g., between highly correlated covariates). Models will be considered hierarchical if the more complex model can be reduced to the less complex model by removing (or fixing the value of) various of its estimated parameters. This ΔOFV is asymptomatically χ^2 distributed with degrees of freedom (d.f.) equal to the difference in number of estimated parameters between the two models. A ΔOFV with a χ^2 probability less than or equal to 0.01 (6.64 points of OFV, d.f.=1) will favor the model with the lower OFV. Backward elimination during covariate evaluation will use a more stringent criterion as described in Section 0.

Changes in OFV will be considered in conjunction with other GoF plots and metrics. Significant and meaningful reductions in the OFV are often accompanied by decreases in the RSE of estimated parameters, normalization of IIV, and residual error terms, reductions in their variance estimates, and reductions in shrinkage estimates for η and ε. Significant differences in OFV that are not associated with improvements in these metrics and relevant GoF plots will be critically reviewed for indications of model misspecification.

Final model evaluation

The final run should meet the following criteria:

- A "minimization successful" statement is issued by NONMEM.
- A covariance step is completed without warning messages by NONMEM.
- The number of significant digits is ≥ 3 for all estimated θ.
- Final estimates of θ are robust to initial estimates or close to boundaries.
- The RSE of estimated θ are less than 40%.
- 95% CI for estimated θ exclude values that would effectively reduce the hierarchical structure of the model.
- Correlations between individual η_{i_P} for all parameter pairs and ρ estimated for all off-diagonal elements in Ω are less than 0.95.
- GoF plots display no unexplained trends.

A visual predictive check (VPC) will be carried out on the final model; the final model will be used to simulate random individuals and calculate the corresponding prediction interval of the population, and the observed data will be superimposed for consistency. More complex VPC figures may also be generated if data permits.
Alternative methods of obtaining RSE (*e.g.,* $COV after IMP method at FOCEI final estimates) may be used if the NONMEM covariance step returns warning messages.
Final models that fail to meet these criteria will be accepted only after careful review and consideration of the modeling objectives. The justification for accepting a final model that does not meet these criteria will be detailed in the final report.

Simulations

The final target engagement model will be used for simulation purposes. Simulations will be conducted based on the findings of the final model and as data permits to answer the following questions:

- What is the percent HSD17B13 protein/mRNA knockdown from baseline at each time point with each dose?
- What covariates explain some of the variability in HSD17B13 protein/mRNA knockdown over a time?

Other objectives and simulations may also be considered based on final model and initial simulation results.



[redacted]

4.2.3. Data exclusion and Sensitivity analyses

Participants should be evaluable for both PK and PD in order to be included on the PK-PD target engagement model development. Any participants with HSD17B13 protein below the LLOQ at baseline will be excluded from model development. Additional subjects may be excluded, at the discretion of the CPMS study lead, if the PK concentration data and/or mRNA/protein levels are not deemed to be amenable to evaluation. Outlier detection will be based initially on visual examination of individual and pooled PK and PD profiles. Additionally, data points identified with a CWRES>6 during the initial model building process may be excluded from the analysis. However, the final model should be retested with and without inclusion of these data points. The exclusions and reason for exclusions will be included in the report. It should be noted that outliers in one model may not be an outlier in another and as such the declaration of an outlier is model dependent. Additional secondary endpoint PD summaries will be produced without the data that may be excluded from the main PK-PD target engagement model of the primary analysis as described in 4.3.1.

4.3. Secondary Endpoints Analyses

4.3.1. Secondary endpoints

The secondary endpoints will be assessed using the same elements of intervention, treatment, population and intercurrent events as the primary estimands. The main approach to managing intercurrent events will align with that of the primary estimands. Additionally, it will include supplementary estimands for self-reported alcohol consumption exceeding eligibility criteria and the use of prohibited medications, as outlined in Section 4.1.3.

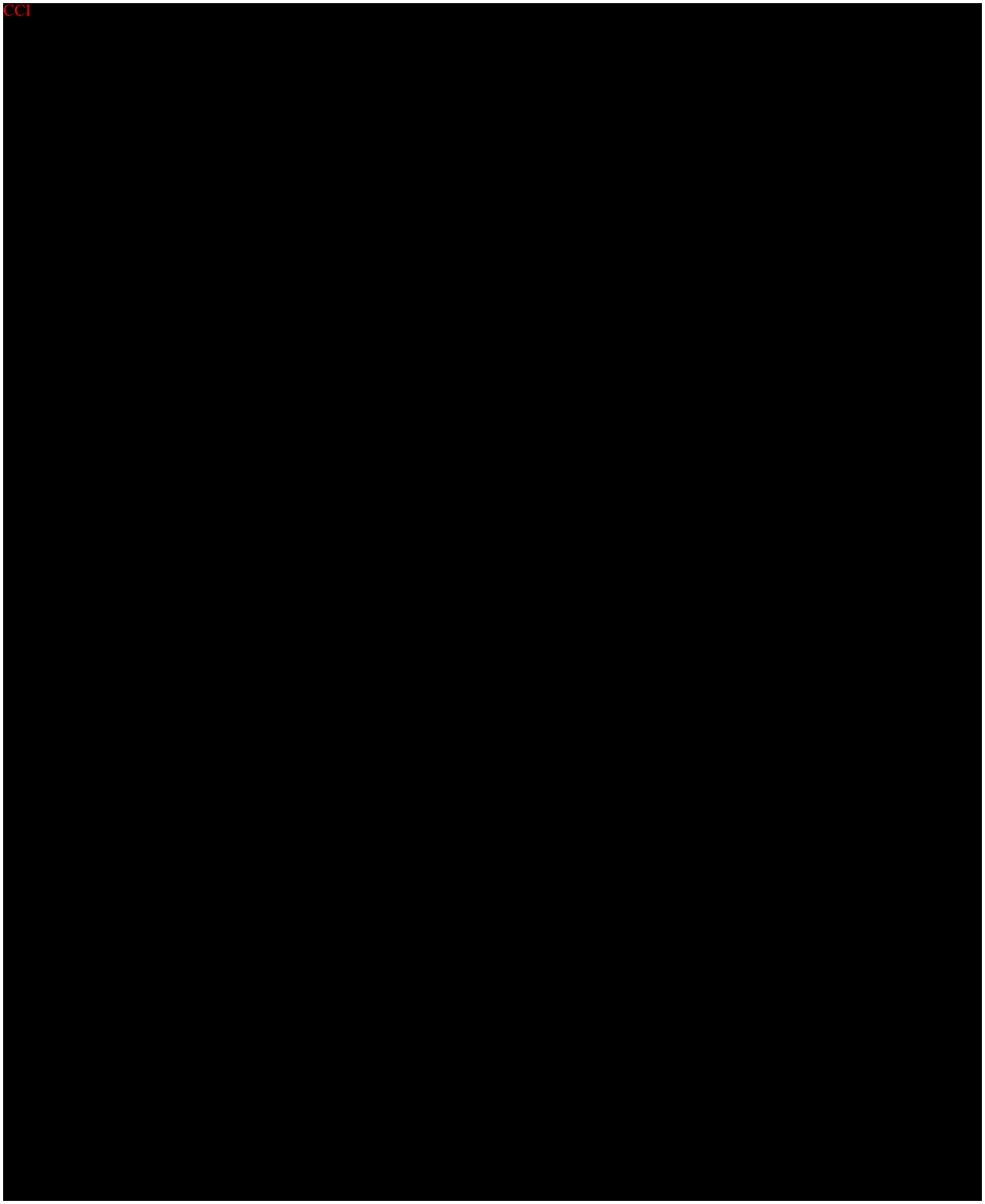
Secondary Objectives	Secondary Endpoints
• To evaluate GSK4532990 exposure parameters after a single dose in participants with NASH or suspected NASH (PK endpoints)	Plasma exposure parameters of GSK4532990: • Area under the concentration-time curve from time zero (pre-dose) to the last quantifiable concentration (AUC0-t) • Maximum observed concentration (Cmax)

Secondary Objectives	Secondary Endpoints
• To evaluate the PD effect of GSK4532990 after a single dose in participants with NASH or suspected NASH (PD endpoints)	• Percent change from baseline in the observed HSD17B13 protein expression levels at each assigned post-dose biopsy time: Weeks 8, 12, or 28 • Percent change from baseline in the observed HSD17B13 mRNA expression levels at each assigned post-dose biopsy time: Weeks 8, 12, or 28

4.3.2. Main analytical approach

CCI




CCI

CCI



CCI

[redacted]

CCI

[redacted]

CCI

[redacted]

CCI



4.5. Supportive information, and metrics

This section provides a synopsis of the supportive material that will be included in the CSR alongside their respective endpoints and is related to the model evaluation and provides support for the decision making that preceded certain analytical steps. This includes additional information, analytical metrics and model diagnostics.

Metric/Summary	Section/ Endpoint	Rationale	CSR
Model goodness of fit figures	4.2.2.2	To complement the primary PKPD model. Plots will include but are not limited to predicted vs observed values comparisons and various residual analyses. Model discrimination and selection metrics.	Appendix / Main body
Plots and metrics to identify covariates of importance	4.2.2.1	Graphical & statistical methods in conjunction with scientific rationale to identify covariates to examine on target engagement parameters	Main body / Appendix
Simulation-based metrics for the model predicted median, 2.5th and 97.5th	4.2.1	The additional simulation-based metrics will be provided, with their corresponding C.Is in order to	Main body

percentiles of the percent change from baseline of HSD17B13 mRNA and protein.		demonstrate the accuracy of model predicted variability.	
Population PK model information	4.2.2.1	Details about the population PK model development which is the 1st step in developing the PK/PD model.	Appendix

4.6. Safety Analyses

Safety analyses will be based on the Safety Analysis Set, unless otherwise specified. All analyses will be reported for each treatment arm. For all the adverse events analyses incidence proportions will be provided.

4.6.1. Duration of Study

The total study duration, including screening procedures, dosing and post-dose follow-up for each participant, is approximately 24 weeks for Treatment Arms 1-3 and 36 weeks for Treatment Arm 4. The observed exposure time will be calculated as the difference between the treatment start date and the date of the last follow up ((date of last follow up) - (treatment start date) +1). The number of days of exposure will be summarized by treatment arm with the following summary statistics:

- Duration of exposure (days): n, Mean, SD, Median, Min, Max.

4.6.2. Adverse Events

Adverse event analyses, including adverse events (AEs), serious adverse events (SAEs) and adverse events of special interest (AESIs), will be based on GSK core data standards. Adverse events will be mapped to system organ classes (SOC) and preferred terms (PT) using the standard Medical Dictionary for Regulatory Affairs (MedDRA dictionary). All AE and SAE summaries will be by SOC and PT unless otherwise specified.

An adverse event (AE) is considered treatment-emergent if the AE onset date is on or after study intervention start date. All AE summaries will be based on treatment-emergent events unless otherwise specified. SAE summaries will be based on treatment-emergent events, and SAEs that don't meet the definition of treatment-emergent will be summarised separately. Adverse events will be coded using Medical Dictionary for Regulatory Activity (MedDRA) and graded by the investigator according to National Cancer Institute (NCI)-CTCAE version 5.

AEs that are treatment-emergent will be summarized by treatment group including:

- counts and percentages of participants with any AE,
- common AEs defined as AEs with ≥ 15% incidence in any treatment group,
- common AEs defined as AEs with ≥ 25% incidence in any treatment group,
- AEs by maximum grade also using PTs by SOC and max CTCAE grade
- AEs related to study treatment,
- AEs related to the study procedures (e.g., biopsy) by SOC and PT,
- study treatment related AEs leading to permanent discontinuation of study intervention,
- non-serious AEs.

SAEs will be summarized by treatment group including:

- counts and percentages of participants with any SAE,
- SAEs related to study treatment,
- fatal SAEs and summary of deaths.

A study treatment-related AE is defined as an AE for which the investigator classifies the possible relationship to study treatment as "Yes". A worst-case scenario approach will be taken to handle missing relatedness data, i.e., the summary table will include events with the relationship to study treatment as 'Yes' or missing.

A by-subject AE data listing including but not limited to the verbatim term, PT, CTCAE severity grade, relationship to study treatment, and outcome will be provided. AEs leading to withdrawal from study and SAEs will be provided.

4.6.2.1. Adverse Events of Special Interest

The following will be considered adverse events of special interest (AESI) for the purpose of analyses:

- Injection Site Reactions (ISR)
- Hepatic Events.

ISRs will be identified by HLT (High Level Term) of Injection site reactions, while hepatic events by SMQ of Drug related hepatic disorders – comprehensive search (narrow and broad terms).

Summaries of event characteristics will be provided for each AESI respectively and by SOC and PT, per treatment group as applicable, including:

- Count and percentage of participants with any TEAE and number of events
  - Count and percentage of participants with any SAE and number of serious events
- Count and percentage of participants with any fatal and drug-related fatal SAEs and number of serious events
- Number of events that are related to study treatment (drug related)
  - Number of events leading to study withdrawal
  - Number of events by outcome
  - Number of events by action taken for the event

ISRs will be also summarized as:

- count and percentage of participants by maximum grade. Grade will be evaluated by CTCAE criteria (version 5.0)
- Count and percentage of participants by number of occurrences of ISRs (0, 1-2, 3-4, 5-6, ≥7)
- number of ISRs by duration of reaction (0-3 days, 4-7 days, 8-10 days, >10 days)

A worst-case approach will be applied at a participant level for the maximum grade, i.e., a participant will only be counted once as the worst case from all the events experienced by the participant. For action taken to an event, a participant will be counted once under each action.

4.6.3. Additional Safety Assessments (if applicable)

4.6.3.1. Laboratory Data

The observed and change from baseline of haematology, chemistry laboratory tests and the lipid profile will be produced by visit and by treatment group. Liver function laboratory tests will be included with chemistry lab tests. Blood fibrinogen will be included with haematology.

If a laboratory value which is expected to have a numeric value for summary purposes, has a non-detectable level reported in the database, where the numeric value is missing, but typically a character value starting with "<x" or ">x" (or indicated as less than x or greater than x in the comments field) is present, the number of decimal places in the observed values will be used to determine how much to add or subtract in order to impute the corresponding numeric value.

Example 1: 2 decimal places = "<x" becomes x-0.001 e.g.<0.05 becomes 0.049
Example 2: 1 decimal place = "<x" becomes x-0.01 e.g.<0.2 becomes 0.19
Example 3: 0 decimal places = ">x" becomes x+0.1 e.g.>99 becomes 99.1

Summaries of worst-case changes from baseline with respect to potential clinical importance range will be generated for haematology, chemistry, and urinalysis changes. Decreases to low, changes to within range or no changes from baseline, and increases to high will be summarized for the worst-case post-baseline. If a subject has a decrease to low and an increase to high during the same time interval, then the subject is counted in both the "to Low" categories and the "to High" categories. The denominator for calculating the percentage of participants with values of potential clinical importance will be the number of participants in each treatment arm with non-missing baseline and at least one non-missing post-baseline value.

Summaries of grade shifts from baseline grade will be provided for haematology, chemistry and lipid lab tests that are gradable by CTCAE v5.0. These summaries will display the number and percentage of participants with a post-baseline maximum grade compared to their baseline grade. Missing baseline grade will be assumed as grade 0. For laboratory tests that are graded for both low and high values, summaries will be done separately and labelled by direction.

Summaries of hepatobiliary laboratory events including possible Hy's law cases will be provided. Possible Hy's law cases are defined as any elevated alanine aminotransferase (ALT) >3×upper limit of normal (ULN), total bilirubin ≥2×ULN and alkaline phosphatase (ALP) <3×ULN/missing. Total bilirubin ≥2×ULN can be within 28 days following the ALT elevation and if direct bilirubin is available on the same day, it must be > 35% of total bilirubin. ALP <3×ULN/missing means it is satisfied unless the ALP is ≥3xULN at the time of bilirubin elevation. The summary will be produced for worst case post baseline only.

An e-DISH plot of maximum post baseline total bilirubin versus maximum post baseline ALT will be created. Scatter plots of maximum vs baseline ALT will also be created.

The count and percentage of participants who have met the increased liver and renal monitoring criteria as defined in the protocol will be summarized for each treatment group.

The following listings will be provided for laboratory data:

- A listing of participants who have met increased liver monitoring criteria.
- A listing of participants who have met increased renal monitoring criteria.
- A listing of participants with any clinical chemistry, hematology, coagulation or urinalysis laboratory results of potential clinical importance.

4.6.3.2. Vital Signs

Observed vital signs and the change from baseline in vital signs, including temperature, pulse rate (beats per min), respiratory rate (breaths per min), systolic blood pressure (mmHg) and diastolic blood pressure (mmHg), will be summarised by visit and by treatment group.

Summary of worst-case change from baseline with respect to potential clinical importance range will be produced for pulse rate, systolic and diastolic blood pressure.

Participants with any vital signs data of potential clinical importance will be listed and summarised.

4.6.3.3. ECG

The number of participants with ECG findings will be summarized as per clinical significance at each planned visit. Clinical significance will be presented as normal and abnormal, and abnormal will be further presented as clinically and not clinically significant.

Change from baseline will be also summarized for each ECG value: mean heart rate (beats/min), aggregate PR interval (msec), aggregate QRS interval (msec), aggregate uncorrected QT interval (msec), aggregate QTcF (msec).

Further QTc data analysis will use the collected values based on Fridericia formula.

QTc values based on Fridericia formula will be rounded to the nearest integer and the values will be categorized into the following CTCAE grade and ranges: Grade 0 (<450 msec), Grade 1 (450-480 msec), Grade 2 (481-500 msec), and Grade 3 (≥501 msec). Summaries of grade increase will be provided. These summaries will display the number and percentage of subjects with any grade increase, increase to grade 2 and increase to grade 3 for the worst-case post-baseline only. Missing baseline grade will be assumed as grade 0.

Changes in QTc values for post-baseline observations will be categorized into the clinical concern ranges which are specific to changes in QTc: ≤30, 31-60 and >60 msec. A summary of change in QTc value will display the number and percentage of subjects with a change within each range for the worst-case post-baseline observation only. Subjects with missing baseline values will be excluded from this summary.

4.7. Other Analyses

4.7.1. Subgroup analyses

N/A

4.7.2. Other variables and/or parameters

The binary variable of alcohol consumption that exceeds the exclusion criteria threshold will be based on the self-reported alcohol consumption at weeks 8, 12,16, 20, 24 & 28. This variable will be used to identify an intercurrent event as per the estimands definitions. It will be considered as an intercurrent event when it exceeds ≥14 standard drinks (24 units, 196 g ethanol) per week for females or ≥21 standard drinks (37 units, 294 g ethanol) per week for males.

One standard drink is equivalent to: 12 US fluid ounces (oz) beer (5% alcohol); 5 oz of wine (12% alcohol), or 1.5 oz of 80 proof spirits (40% alcohol).

The number and proportion of participants with self-reported alcohol consumption exceeding the threshold will be summarised at weeks 8, 12,16, 20, 24 & 28.

4.7.3. Analyses to Support Regional Submission

N/A

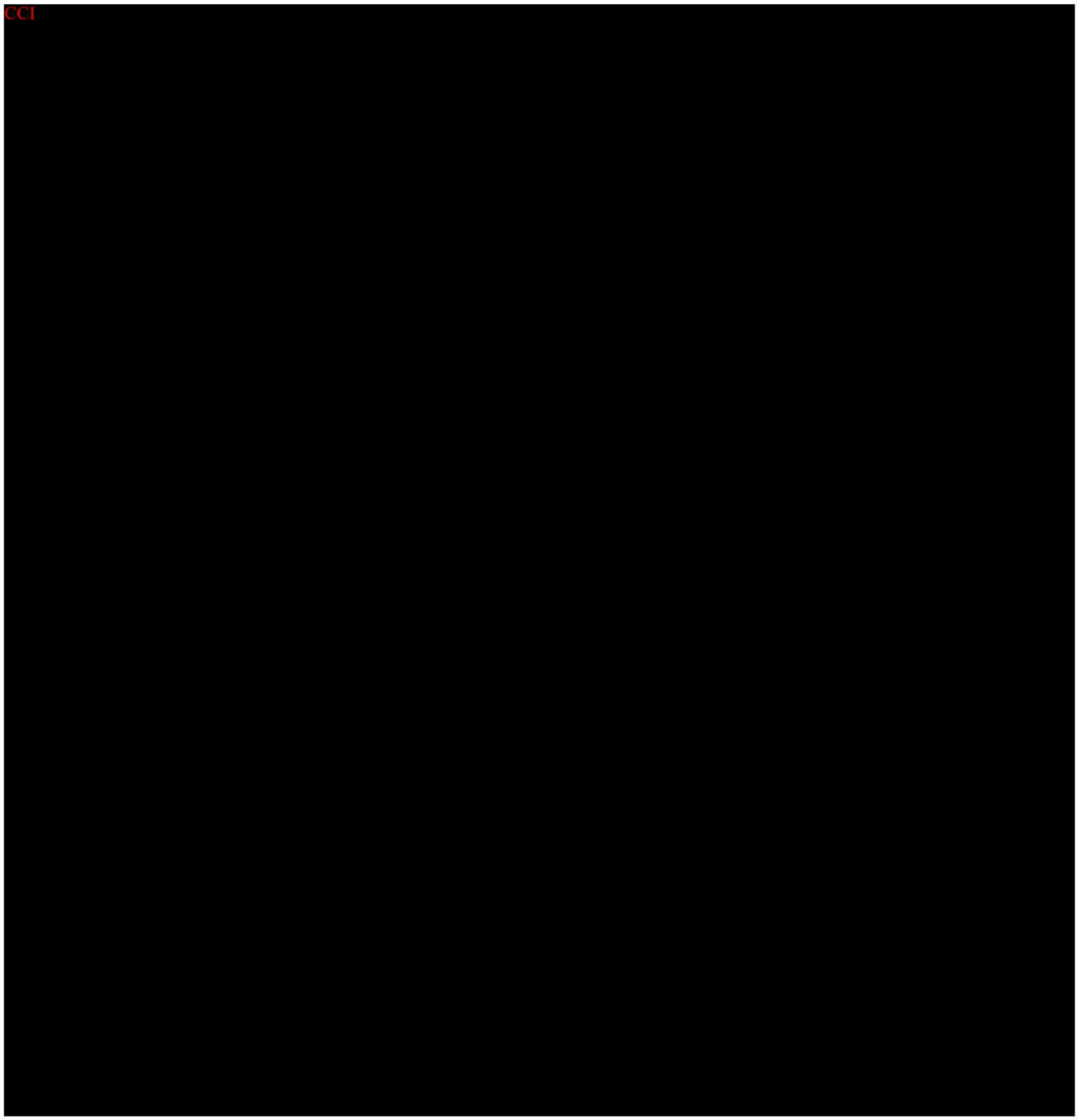
CCI

[redacted]

CCI

[redacted]

CCI



4.9. Changes to Protocol Defined Analyses

Table 5, Summary of changes to protocol defined analyses plan.

Protocol Defined Analysis	SAP Defined Analysis	Rationale for Changes
Per protocol (Section 3, protocol amendment 2), for the primary endpoint analysis, besides the 'treatment policy' strategy, there are additional estimands using 'whilst not on prohibited medication'	The additional estimands for the primary endpoint analysis were removed and only the 'treatment policy applies' (SAP sections 1.1.1 and 1.1.2).	Increased alcohol consumption and use of prohibited medications are intercurrent events that will be considered as candidate covariates in the PK-PD target engagement model.

Protocol Defined Analysis	SAP Defined Analysis	Rationale for Changes
and 'whilst not exceeding the weekly alcohol limit' strategies.		

5. SAMPLE SIZE DETERMINATION

The target sample size is 48 participants who will be included in the analysis of the primary estimand and provide sufficient precision to the study and is adjusted to account for missing biopsy data. This study design results in 12 participants in each treatment arm, and within each treatment arm, 6 participants will be assigned to one of two biopsy time points.

The sample size estimation was based on the minimum number of subjects and pre- and post-dose HSD17B13 protein and mRNA samples required to estimate the PK/PD target engagement model parameters with good precision and bias. The target engagement model is developed to describe the baseline and post-dose changes in hepatic HSD17B13 protein and mRNA concentrations as a result of GSK4532990 at multiple timepoints and at different dose levels. The ability of the target engagement model to calculate the change in HSD17B13 protein and mRNA is dependent upon the estimation of the fixed and random effect parameters with good precision, acceptable parameter bias and normalized root mean square error.

A target engagement model has been developed utilizing data from AROHSD1001 with assumptions for maximum drug effect and HSD17B13 degradation rates. Thus, the purpose of this study is to examine the validity of those assumptions by accurately estimating all parameters in the target engagement model which will lead to precise estimation of baseline and post GSK4532990 knockdown of HSD17B13 protein and mRNA levels. Therefore, this exercise involved optimal design evaluation using virtual trial simulations and target engagement model re-estimation. The simulations focused on the impact of sample size on the estimation of IC50, one of the fixed effect parameters of primary interest. Based on these simulations, summaries of the expected precision of the predicted percent change of the HSD17B13 protein and mRNA levels from baseline are provided in the following table for the different treatment arms and biopsy times of the study.

Table 6, Simulation-based summary of precision for the model-based predictions in HSD17B13 mean change from baseline for different combinations of dose arm and biopsy time.

Biopsy time	Dose:	25mg	200mg	400mg	800mg
Day 60 prediction range mean with 95%		9.9% (6.2%-14.1)	5.2% (3.1%-7.9%)	3.1% (1.8%-4.6%)	1.7% (1% - 3%)
Day 90 prediction range mean with 95%		7.8%(4.8%11.5%)	8.5% (5.2%-12.2%)	6% (3.5%-8.7%)	3.8% (2.2%-6.2%)
Day 120 prediction range mean with 95%		4.6% (2.7%-7.1%)	10.2% (6.3%-7.1%)	9.1% (5.5%-12.8%)	6.9% (4.1%-10.5%)

The model was used to generate a "global" dataset with baseline and daily changes in hepatic HSD17B13 protein due to GSK4532990 by simulating individual participant level PK and PD parameters ("true parameter estimates") for each participant over a single dose range of 25 mg to 800 mg. This dataset was used for random sampling without replacement to create virtual clinical trial designs varying the number of participants per treatment arm, the number of participants per timepoint, and the biopsy sample time points. Each clinical trial was used to estimate PK parameters in the target engagement model for each set of N patient/timepoint for 1000 trials. Results from the target engagement model estimation were compared to the "true parameter estimates" based on final fixed and random effect parameter estimates of the target engagement model estimated with good precision (% relative standard error) <10% for each simulated trial design.

6. SUPPORTING DOCUMENTATION

6.1. Appendix 1 Study Population Analyses

Unless otherwise specified, the study population analyses will be based on the Enrolled analysis set. A summary of the number of participants in each of the participant level analysis set will be provided.

6.1.1. Participant Disposition

A summary of the number and percentage of participants who completed the study as well as those who prematurely withdrew and/or added to the study will be provided. A study disposition table based on the screened analysis set with number and percentage of participants in each analysis set. Reasons for study withdrawal will be summarised. For those who have neither completed nor withdrawn, they will be categorized as on study intervention or in follow up at interim analyses, or as withdrawn at the final analysis.

A summary of study intervention status will be provided. This display will show the number and percentage of participants who have completed the scheduled study intervention, are ongoing with study intervention, or have discontinued study intervention. Reasons for study intervention discontinuation will be summarized. In the final analysis, participants who have not completed study intervention but don't have an early discontinuation date will be categorised as discontinued.

6.1.2. Demographic and Baseline Characteristics

The demographic and baseline characteristics will be summarised with descriptive statistics, based on the Enrolled Analysis Set.

Table 6, Summary of Baseline characteristics including demographics.

Baseline characteristics	Details
Age	• Mean (SD) in years • Categories (%): 18-49, 50-64 and ≥65
Gender	Categories (%): Male, Female, Unknown
Ethnicity	Categories (%): Hispanic or Latino, Not Hispanic or Latino, Not Reported, Unknown
Race	Categories (%): American Indian or Alaska Native, Asian, Black or African American, Native Hawaiian or Other Pacific Islander, White, Not Reported, Unknown
Body weight	Mean (SD) in kg
BMI	• Mean (SD) in kg/m^2 • Categories (%): < 35, ≥35
T2DM	Categories (%): Yes, No
HFF (hepatic fat fraction) by MRI-PDFF	Mean(SD) based on the average of all available locations in % for patients with results from 4 or more locations
Laboratory parameters	
ALT, AST, ALP, GGT	Mean (SD) in IU/L
Total bilirubin, Direct bilirubin	Mean (SD) in µmol/L
Platelet count	Mean (SD) in x 10^9/L
INR	Mean (SD)
Albumin	Mean (SD) in g/dL
Fibrinogen	Mean (SD) in g/L

Biomarkers of metabolic syndrome	
Total Cholesterol, LDL-C, HDL-C, non-HDL-C, triglycerides	Mean (SD) in mg/dL
Hemoglobin A1c	Mean (SD) in %
CCI [redacted]	
Alcohol consumption	
Self-reported	Categories (%): Exceeding threshold, Not Exceeding threshold, Unknown

Abbreviations: ALP=Alkaline phosphatase; ALT= alanine aminotransferase; AST= aspartate aminotransferase; BMI=body mass index; ELF™=Enhanced Liver Fibrosis score; EtG = ethyl glucuronide; GGT= Gamma-glutamyl transferase; HA = hyaluronic acid; HDL-C = High density lipoprotein cholesterol; HFF= Hepatic fat fraction; INR= International normalized ratio; LDL-C= Low density lipoprotein cholesterol; LSM=Liver stiffness measurement; CCI [redacted] CCI [redacted] PETH= phosphotidylethanol; PIIINP=amino-terminal pro-peptide of type III procollagen; SD= standard deviation; T2DM= Type 2 diabetes mellitus; TIMP-1=tissue inhibitor of metalloproteinases 1; VCTE™= Vibration-controlled transient elastography

6.1.3. Protocol Deviations

Important protocol deviations will be summarized.

Protocol deviations will be tracked by the study team throughout the conduct of the study. These protocol deviations will be reviewed to identify those considered as important as follows:

- Data will be reviewed prior to freezing the database to ensure all important deviations (where possible without knowing the study intervention details) are captured and categorised in the protocol deviations dataset.

- This dataset will be the basis for the summaries of important protocol deviations.

6.1.4. Prior and Concomitant Medications

Concomitant medications will be coded using both the GSK Drug and WHO Drug dictionaries. However, the summary will be based on GSK Drug dictionary only. The summary of concomitant medications will be provided by ingredient, i.e., multi-ingredient medications will be summarized for each individual ingredient rather than a combination of ingredients. The summary will be created using ingredient base names, i.e., ingredients with the same base name but different salt will appear under one base name in the summary.

A concomitant mediation may be included in more than one ATC level category and appear more than once. Summary tables will present the number and % of participants with at least one medication in an ATC/ingredient row.

Concomitant medications include any medication that was taken at some point during the on-intervention period.

6.1.5. Prohibited Medications

The use of prohibited medications by study participants will be assessed based on whether they have abstained from initiating any new classes of medications or investigational drugs specified in Sections 5.1 and 5.2 (Inclusion & Exclusion Criteria respectively) of the protocol during the study. The GSK medical monitor or their delegate will review the list of participants and timepoints associated with prohibited medication use.

6.2. Appendix 2 Data Derivations Rule

6.2.1. Criteria for Potential Clinical Importance

Reference ranges for all laboratory parameters collected throughout the study are provided by the laboratory. A laboratory value that is outside the reference range is considered either high abnormal (value above the upper limit of the reference range) or low abnormal (value below the lower limit of the reference range). Note: a high abnormal or low abnormal laboratory value is not necessarily of clinical concern.

The criteria displayed in the following tables will be used to flag potential clinical importance.

Table 7, Summary of parameters and criteria used to determine potential clinical importance.

Haematology				
Laboratory Parameter	Units	Category	Clinical Concern Range	
			Low Flag (< x)	High Flag (≥x)
Blood fibrinogen	g/L	Absolute	1.38	

INR	ratio	NA		1.5

Clinical Chemistry				
Laboratory Parameter	Units	Category	Clinical Concern Range	
			Low Flag (< x)	High Flag (>x)
Albumin	g/L	Absolute	30	
Albumin		% Change from Baseline (BL)	20% decrease	
Creatine kinase	IU/L	Absolute		250
Serum Creatinine	µmol/L	% Change from BL		50% increase
Serum Creatinine	µmol/L	Absolute increase from BL		44.2µmol/L (0.5mg/dL) from baseline
eGFR (CKD-EPI) (GFR from Creatinine Adjusted for BSA)	mL/min/ 1.73m^2	Absolute	<60	
eGFR (CKD-EPI) (GFR from Creatinine Adjusted for BSA)	mL/min/ 1.73m^2	% Change from BL	50% decrease	

Liver Function			
Test Analyte	Units	Category	Clinical Concern Range
ALT	IU/L	Multiples of BLN	≥2x
ALT	IU/L	Multiples of ULN	≥3x
ALT	IU/L	Absolute	≥ 300 IU/L
AST	IU/L	Multiples of BLN	≥2x
AST	IU/L	Multiples of ULN	≥3x
AST	IU/L	Absolute	≥ 300 IU/L
ALP	IU/L	Multiples of ULN	≥2x
Total Bilirubin	µmol/L	Multiples of ULN	≥ 1.5x

Urinalysis			
Test Analyte	Units	Category	Clinical Concern Range
Urine albumin: creatinine ratio	g/mol	Absolute	>=56.5 g/mol

Vital Signs		
Parameter (Absolute)	Units	Clinical Concern Range

		Lower	Upper
Systolic Blood Pressure	mmHg	< 85	> 160
Diastolic Blood Pressure	mmHg	< 45	> 100
Heart Rate	bpm	< 40	> 110

ECG			
Parameter	Units	Clinical Concern Range	
		Lower	Upper
QRS interval	Msec	< 75	> 110

6.2.2. Study Period

Assessments and events will be classified according to the time of occurrence relative to the study intervention period.

25, 200 & 400mg treatment arms	
Treatment State	**Definition**
Pre-Treatment	Assessment/Event Date < Date of start of study intervention
Post-Treatment	Assessment/Event Date > Date of dose of study intervention + 131 days

The definitions of treatment states for AEs are provided below:

25, 200 & 400mg treatment arms	
Treatment State	**Definition**
Pre-Treatment	AE start date < Date of start of study intervention
Post-Treatment	AE start date > Date of dose + 131 days

800mg treatment arm	
Treatment State	**Definition**
Pre-Treatment	Assessment/Event Date < Date of 0start of study intervention
Post-Treatment	Assessment/Event Date > Date of dose of study intervention + 215 days

The definitions of treatment states for AEs are provided below:

800mg treatment arm	
Treatment State	**Definition**
Pre-Treatment	AE start date < Date of start of study intervention
Post-Treatment	AE start date > Date of dose + 215 days

6.2.3. Study Day and Reference Dates

The safety reference date is the study intervention start date and will be used to calculate study day for safety measures.

The study day is calculated as below:

- Assessment Date = Missing → Study Day = Missing
- Assessment Date < Reference Date → Study Day = Assessment Date – Ref Date
- Assessment Date ≥ Reference Date → Study Day = Assessment Date – Ref Date + 1

6.2.4. Assessment Window

For data summaries by visit, if a scheduled visit is available, its nominal visit description will be used. Unscheduled and early withdrawal visit data will be slotted into a target visit based on the visit windows defined in the table below. A scheduled visit will always be prioritised over unscheduled visits. If there are multiple unscheduled assessments within the same window, the closest to the target timepoint will be prioritised. If all unscheduled assessments within the same window have the same distance from the target timepoint, the earliest one will be prioritised.

Table 8, Analysis window ranges per visit and treatment arm.

Target	Analysis Window treatment arms 25, 200 and 400 mg		
	Beginning Timepoint	Ending Timepoint	Analysis Visit
Day -42	Day -70	Day -28	Screening 1
Day -14	Day -27	Day -2	Screening 2
Day 1	Day -1	Day 1	Day 1
Day 2	Day 2	Day 2	Day 2
Day 3	Day 3	Day 5	Day 3
Day 14	Day 6	Day 21	Week 2
Day 28	Day 22	Day 35	Week 4
Day 42	Day 36	Day 49	Week 6
Day 56	Day 50	Day 70	Week 8
Day 84	Day 71	Day 98	Week 12
Day 112	Day 99	Day 119 /999 for non-WOCBP	Week 16/EOS
Day 126	Day 120	Day 999	Week 18/target EOS for WOCBP

Target	Analysis Window 800 mg treatment arm only		
	Beginning Timepoint	Ending Timepoint	Analysis Visit
Day -42	Day -70	Day -28	Screening 1
Day -14	Day -27	Day -2	Screening 2
Day 1	Day -1	Day 1	Day 1
Day 2	Day 2	Day 2	Day 2
Day 3	Day 3	Day 5	Day 3
Day 14	Day 6	Day 21	Week 2
Day 28	Day 22	Day 35	Week 4
Day 42	Day 36	Day 49	Week 6
Day 56	Day 50	Day 70	Week 8
Day 84	Day 71	Day 98	Week 12
Day 112	Day 99	Day 126	Week 16
Day 140	Day 127	Day 154	Week 20
Day 168	Day 155	Day 182	Week 24
Day 196	Day 183	Day 203 /999 for non-WOCBP	Week 28/EOS
Day 210	Day 204	Day 999	Week 30/target EOS for WOCBP

6.2.5. Multiple measurements at One Analysis Time Point

When triplicate ECG assessments are taken, the mean of the measurements will be calculated first and summary statistics will be based on the calculated mean. This will apply to both baseline and post baseline assessments.

If multiple assessments are taken from the same type of lab, the worst case (i.e., closest to PCI boundary values if not of potential clinical importance, and furthest away from PCI boundary values if of potential clinical importance) will be used.

Participants having both High and Low values versus Normal Ranges at any post-baseline visits for safety parameters will be counted in both the High and Low categories of "Any

visit post-baseline" row of related summary tables. This will also be applicable to relevant Potential Clinical Importance summary tables.

6.2.6. Handling of Partial Dates

<table>
<tr><th>Element</th><th>Reporting Detail</th></tr>
<tr><td>General</td><td>

Partial dates will be displayed as captured in participant listing displays.
However, where necessary, display macros may impute dates as temporary variables for sorting data in listings only. In addition, partial dates may be imputed for 'slotting' data to study phases or for specific analysis purposes as outlined below.
Imputed partial dates will not be used to derive study day, time to onset or duration (e.g., time to onset or duration of adverse events), or elapsed time variables (e.g., time since diagnosis). In addition, imputed dates are not used for deriving the last contact date in overall survival analysis dataset.

</td></tr>
<tr><td>Adverse Events</td><td>

Partial dates for AE recorded in the CRF will be imputed using the following conventions:

<table>
<tr><td>Missing start day</td><td>If study intervention start date is missing (i.e., participant did not start study intervention), then set start date = 1st of month.

Else if study intervention start date is not missing:

• If month and year of start date = month and year of study intervention start date, then

– If stop date contains a full date and stop date is earlier than study intervention start date, then set start date= 1st of month.

– Else set start date = study intervention start date.

Else set start date = 1st of month.</td></tr>
<tr><td>Missing start day and month</td><td>If study intervention start date is missing (i.e., participant did not start study intervention), then set start date = January 1.

Else if study intervention start date is not missing:

• If year of start date = year of study intervention start date, then

– If stop date contains a full date and stop date is earlier than study intervention start date, then set start date = January 1.

– Else set start date = study intervention start date.

Else set start date = January 1.</td></tr>
</table>
</td></tr>
</table>

Element	Reporting Detail	
	Missing end day	A '28/29/30/31' will be used for the day (dependent on the month and year).
	Missing end day and month	No Imputation
	Completely missing start/end date	No imputation
Concomitant Medications/ Medical History	• Partial dates for any concomitant medications recorded in the CRF will be imputed using the following convention:	
	Missing start day	If study intervention start date is missing (i.e., participant did not start study intervention), then set start date = 1st of month. Else if study intervention start date is not missing: • If month and year of start date = month and year of study intervention start date, then – If stop date contains a full date and stop date is earlier than study intervention start date, then set start date= 1st of month. – Else set start date = study intervention start date. Else set start date = 1st of month.
	Missing start day and month	If study intervention start date is missing (i.e., participant did not start study intervention), then set start date = January 1. Else if study intervention start date is not missing: • If year of start date = year of study intervention start date, then – If stop date contains a full date and stop date is earlier than study intervention start date, then set start date = January 1. – Else set start date = study. intervention start date. Else set start date = January 1.
	Missing end day	A '28/29/30/31' will be used for the day (dependent on the month and year).
	Missing end day and month	A '31' will be used for the day and 'Dec' will be used for the month.

Element	Reporting Detail		
	Completely missing start/end date	No imputation	

6.2.7. Trademarks

Trademarks of the [GlaxoSmithKline / ViiV Healthcare] Group of Companies
-

Trademarks not owned by the [GlaxoSmithKline / ViiV Healthcare] Group of Companies
CCI

7. REFERENCES

- Ahn, J.E., M.O. Karlsson, A. Dunne, and T.M. Ludden. (2008) Likelihood based approaches to handling data below the quantification limit using NONMEM VI. J Pharmacokinet Pharmacodyn. 35(4): p. 401-21.
- Bauer R (2019) NONMEM Tutorial Part II: Estimation Methods and Advanced Examples CPT Pharmacometrics Syst. Pharmacol. 8, p. 538–556
- Beal, S.L., L.B. Sheiner, A. Boeckmann, and R.J. Bauer, NONMEM User's Guides. (1989- 2009), 2009, Icon Development Solutions: Ellicott City, MD, USA.
- Bergstrand, M., A. Hooker, J. Wallin, and M. Karlsson. (2011) Prediction-corrected visual predictive checks for diagnosing nonlinear mixed-effects models. The AAPS Journal. 13(2): p. 143-151.
- European Medicines Agency (2007) Guideline on Reporting the Results of Population Pharmacokinetic Analyses.
- Hooker, A.C., C.E. Staatz, and M.O. Karlsson. (2007) Conditional weighted residuals (CWRES): a model diagnostic for the FOCE method. Pharmaceutical Research. 24(12): p. 2187-97.
- Li J, Karlsson MO, Sanhajariya S, Isbister GK, and Duffull SB (2021). A general model for cell death and biomarker release from injured tissues. J Pharmacokinet Pharmacodyn. 48:69-82.
- US Food and Drug Administration (2019) Draft Guidance for Industry: Population Pharmacokinetics.