TRIAL STATISTICAL ANALYSIS PLAN

Document No.:	**c44584904-01**
BI Trial No.:	**0248-0689**
Title:	Bioequivalence of two Sifrol® tablets following oral administration in healthy subjects (an open-label, randomised, single-dose, two-way crossover trial)
Investigational Product(s):	Sifrol®, pramipexole
Responsible trial statistician(s):	[redacted]
Date of statistical analysis plan:	23 AUG 2024
Version:	1.0

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LIST OF TABLES

2. LIST OF ABBREVIATIONS

See Medicine Glossary:
http://glossary

Term	Definition / description
[redacted]	
ANOVA	Analysis of variance
$AUC_{0-\infty}$	Area under the concentration-time curve of the analyte in plasma over the time interval from 0 extrapolated to infinity
AUC_{0-tz}	Area under the concentration-time curve of the analyte in plasma over the time interval from 0 to the last quantifiable data point
BP	Blood pressure
CI	Confidence interval
C_{max}	Maximum measured concentration of the analyte in plasma
CSD	Company Standard Displays
CV	Arithmetic coefficient of variation
gCV	Geometric coefficient of variation
gMean	Geometric mean
[redacted]	
Max	Maximum
Min	Minimum
N	Number non-missing observations
P10	10^{th} percentile
P90	90^{th} percentile
PKS	Pharmacokinetic parameter analysis set
PR	Pulse rate
Q1	1^{st} quartile
Q3	3^{rd} quartile
R	Reference treatment
RAGe	Report Appendix Generator system
SD	Standard deviation
T	Test treatment

[redacted]

Term	Definition / description
TS	Treated set

3. INTRODUCTION

As per ICH E9 (9.1), the purpose of this document is to provide a more technical and detailed elaboration of the principal features of the analysis described in the protocol, and to include detailed procedures for executing the statistical analysis of the primary and secondary variables and other data.

This TSAP assumes familiarity with the Clinical Trial Protocol (CTP), including Protocol Amendments. In particular, the TSAP is based on the planned analysis specification as written in CTP Section 7 'Statistical Methods and Determination of Sample Size'. Therefore, TSAP readers may consult the CTP for more background information on the study, e.g., on study objectives, study design and population, treatments, definition of measurements and variables, planning of sample size, randomization.

Study data as collected in the electronic case report form (eCRF) will be stored in a trial database within the RAVE electronic data capture (EDC) system. All study data (including external data) will then be uploaded to the Clinical Data Repository (CDR).

Pharmacokinetic (PK) parameters will be calculated using Phoenix WinNonlin™ software (version 8.1.1 or higher, [redacted] or SAS Version 9.4 (or later version).

The statistical analyses will be performed within the validated working environment CARE, including SAS™ (current Version 9.4, by [redacted]), and a number of SAS™-based tools (e.g., macros for the analyses of AE data or laboratory data; Report Appendix Generator system (RAGe) for compilation/formatting of the CTR appendices).

4. CHANGES IN THE PLANNED ANALYSIS OF THE STUDY

All analyses as planned in the CTP will be performed and are described in more detail in this TSAP.

5. ENDPOINTS(S)

5.1 PRIMARY ENDPOINT(S)

Section 2.1.2 of the CTP:

The following pharmacokinetic parameters will be determined for pramipexole:

- *AUC_{0-tz} (area under the concentration-time curve of the analyte in plasma over the time interval from 0 to the last quantifiable data point)*
- *C_{max} (maximum measured concentration of the analyte in plasma)*

5.2 SECONDARY ENDPOINT(S)

5.2.1 Key secondary endpoint(s)

This section is not applicable as no key secondary endpoints have been defined in the CTP.

5.2.2 Secondary endpoint(s)

Section 2.1.3 of the CTP:

The following pharmacokinetic parameter will be determined for pramipexole:

- *$AUC_{0-\infty}$ (area under the concentration-time curve of the analyte in plasma over the time interval from 0 extrapolated to infinity)*

- *Adverse events (including clinically relevant findings from the physical examination)*

6. GENERAL ANALYSIS DEFINITIONS

6.1 TREATMENT(S)

For basic study information on treatments to be administered and selection of doses, refer to CTP Sections 3 and 4.

This trial is designed as an open-label, randomized, single-dose, two-treatment, two-sequence, two-period crossover design in 28 healthy male and female subjects with a wash-out phase of at least 3 days between the single dose administrations of the two different Sifrol® tablets in the two periods.

For details of dosage and formulation, see Table 1 below.

Table 1 Treatments and labels used in the analysis

Treatment		Short label
R	Sifrol® Tabletten (manufactured in Ingelheim), IR tablet, 0.088 mg pramipexole (base), single dose in the morning of Day 1	Pra-Ing (R)
T	Sifrol® Tabletten (manufactured in [redacted]), IR tablet, 0.088 mg pramipexole (base), single dose in the morning of Day 1	Pra-Enn (T)

Subjects are randomly allocated to one of the two treatment sequences ‘R-T’ or ‘T-R’.

Section 1.2.3 of the CTP:

The Residual Effect Period (REP) of pramipexole is 2 days. This is the period after the last dose during which measurable drug levels and/or pharmacodynamic effects are still likely to be present.

Based on this, the following study phases, analysing treatment and actual treatment will be defined for the analysis of PK endpoints, adverse events (AEs), and other safety endpoints:

Table 2 Analysis phases for statistical analysis of AEs, and actual treatment for analysis of laboratory data, vital signs and concomitant therapies

Study analysis phase	**Treatment Period**	**Treatment short label**	**Start**	**End**
Screening	**Screening**		Date/time of informed consent	Date/time of administration of pramipexole in treatment period 1
On treatment	**Period 1, 2**	**Pra-Ing, Pra-Enn**	Date/time of administration of pramipexole (T or R) in the applicable treatment period	Date/time of next administration of pramipexole (T or R) in the applicable treatment period OR Date/time of previous pramipexole administration plus 2 days — whichever is earlier
Follow-up		**FU**	2 days (48 h) after date/time of previous administration of pramipexole (T or R)	If applicable: date/time of next administration of pramipexole (T or R) OTHERWISE Date/time of EoS examination

Section 7.2.5 of the CTP:

Note that AEs occurring after the last per protocol contact but entered before database lock will be reported to Pharmacovigilance only and will not be captured in the trial database.

The following AE displays will be provided in the CTR:

In Section 15.3 and Appendix 16.1.13.1.8 (for ClinicalTrials.gov and EudraCT only) of the CTR displays, the on treatment phase will be analysed (labelled with the short label for treatment at onset as in Table 2). The screening and follow-up phases will not be included in this analysis.

A total over all on-treatment phases **('Total on-treatment')** will be provided in addition for Section 15.3.

In Section 15.4 and Appendix 16.2 (Listings) of the CTR displays, the screening period, as well as the follow-up phases will additionally be included.

For detailed information on the handling of the treatments refer to Technical TSAP ADS (analysis data set) plan and Analysis Data Reviewers guide.

6.2 IMPORTANT PROTOCOL DEVIATIONS

Data discrepancies and deviations from the CTP will be identified for all treated subjects. Consistency check listings (for identification of deviations of time windows) and a list of protocol deviations (e.g., deviations in drug administration, in blood sampling times, etc.) will be provided to be discussed at the Report Planning Meeting (RPM). At this meeting, it will be decided whether a discrepant data value can be used in analyses or whether it must be corrected in the clinical database. Each protocol deviation must be assessed to determine whether it is an important protocol deviation (iPD).

For definition of iPDs, and for the process of identification of these, refer to the Boehringer Ingelheim (BI) SOP 'Identify and Manage Important Protocol Deviations (iPD)' (9.2)

IPD categories are provided in the DV domain sheet, iPDs will be identified no later than in the Report Planning Meeting, and the iPD categories will be updated as needed.

If any iPDs are identified, they are to be summarised into categories by study period and will be captured in the iPD specification file (DV domain) (9.3) and in the decision log (9.4). Both documents will be stored within the trial master file (TMF) in electronic document management system (EDMS).

The iPDs will be summarised by treatment and listed in the CTR.

6.3 INTERCURRENT EVENTS

This section is not applicable.

6.4 SUBJECT SETS ANALYSED

Section 7.2.1.1 of the CTP:

Statistical analyses will be based on the following analysis sets:

- *Treated set (TS): The treated set includes all subjects who were treated with at least one dose of trial drug. The treated set will be used for safety analyses.*
- *Pharmacokinetic parameter analysis set (PKS): This set includes all subjects in the treated set (TS) who provide at least one PK endpoint that was defined as primary or secondary and was not excluded due to a protocol deviation relevant to the evaluation of PK or due to PK non-evaluability (as specified in the following subsection 'Pharmacokinetics'). Thus, a subject will be included in the PKS, even if he contributes only one PK parameter value for one period to the statistical assessment. Descriptive and model-based analyses of PK parameters will be based on the PKS.*

Table 3 Subject sets analysed

	Subject set	
Class of analysis	TS	PKS
Primary and secondary endpoints		X
Further PK endpoints		X
Safety assessments	X	
Treatment exposure	X	
iPDs	X	
Disposition	X	
Demographics & baseline conditions	X	

[redacted]

6.6 HANDLING OF MISSING DATA AND OUTLIERS

Handling of missing data and outliers will be performed as described in the CTP:

Section 7.3.1 of the CTP: *It is not planned to impute missing values for safety parameters.*

The only exceptions where imputation might be necessary for safety evaluation are AE dates. Missing or incomplete AE dates are imputed according to BI standards (see 'Handling of Missing and Incomplete AE Dates') (9.5).

Section 7.3.2 of the CTP:

Handling of missing PK data will be performed according to the relevant BI internal procedures.

PK parameters that cannot be reasonably calculated based on the available drug concentration-time data will not be imputed.

Missing data and outliers of PK data are handled according to BI standards (see 'Standards and processes for analyses performed within Clinical Pharmacokinetics/Pharmacodynamics' (9.6) and 'Noncompartmental PK/PD Analyses of Clinical Studies' (9.7)).

6.7 BASELINE, TIME WINDOWS AND CALCULATED VISITS

The baseline value for vital signs analysis is defined as the last measurement before first pramipexole drug administration in each treatment period. This means, the baseline for the reference treatment period 'Pra-Ing' (R) and the test treatment period 'Pra-Enn' (T) is usually

the measurement at V2/V3 (as applicable) planned time -1:00 (Day 1) for the analysis of vital signs. Laboratory assessments are only planned at screening and at end of study examination, so the measurements at screening will be used for baseline.

Time windows are defined in Section 6.1 of the CTP. Adherence to time windows will be checked via the consistency check listings at the RPM.

Unscheduled measurements of laboratory data and vital signs data will be assumed to be repeat measurements of the most recent scheduled measurement (e.g., for follow-up or confirmation of a particular value). Therefore, unscheduled measurements will be assigned to the planned time point of the previous scheduled measurement.

7. PLANNED ANALYSIS

Safety analysis (refer to Section 7.8) will be performed by [redacted] and will be presented in Sections 15.1 to 15.4 of the CTR and in Appendix 16.2 and 16.1.13.1.

Inferential statistical analyses of PK endpoints (refer to Section 7.4) will be performed by [redacted] and will be presented in Section 15.5 of the CTR and in Appendix 16.1.13.3.

Descriptive data analysis of PK endpoints and concentrations will be performed by [redacted] and will be presented in Section 15.6 of the CTR and in Appendix 16.1.13.5.

The format of the listings and tables will follow the BI standards (see 'Standards for Reporting of Clinical Trials and Project Summaries' (9.8)) with the exception of those generated for PK-calculations following BI standards for PK/PD analysis (9.9).

The individual values of all subjects will be listed, sorted by treatment sequence, subject number and visit and time point. The listings will be included in Appendix 16.2 of the CTR.

The following standard descriptive statistical parameters will be displayed in summary tables of continuous variables:

N	number non-missing observations
Mean	arithmetic mean
SD	standard deviation
Min	minimum
Median	median
Max	maximum

For plasma concentrations and PK parameters, the following descriptive statistics will additionally be calculated (if applicable or needed):

Nobs	number of observations
CV	arithmetic coefficient of variation
gMean	geometric mean
gCV	geometric coefficient of variation
P10	10^{th} percentile
Q1	1^{st} quartile
Q3	3^{rd} quartile
P90	90^{th} percentile

The data format for descriptive statistics of concentrations will be identical to the data format of the respective concentrations. The descriptive statistics of PK parameters will be calculated using the individual values with the number of decimal places as provided by the evaluation program. Then the individual values as well as the descriptive statistics will be reported with three significant digits in the CTR.

Tabulations of frequencies for categorical data will include all possible categories available in the CRF and will display the number of observations in a category, as well as the percentage (%). Percentages will be given in integer numbers due to the small sample size of < 100. Percentages will be based on all subjects in the respective subject set whether they have non-missing values or not. The category 'missing' will be displayed only if there are actually missing values.

Units of variables should be given in the titles or column/row descriptors in brackets (e.g., '(mg)').

Exclusion of PK parameters

The ADS 'ADPP' (PK parameters) contains column variables APEX and APEXCO indicating inclusion/exclusion (APEX) of a PK parameter and an analysis flag comment (APEXCO). All analyses based on the PKS will include parameters if they are not flagged for exclusion, that is APEX is equal to 'Included'.

Exclusion of PK concentrations

The ADS 'ADPC' (PK concentrations per time-point or per time-interval) contains column variables ACEX and ACEXCO indicating inclusion/exclusion (ACEX) of a concentration and an analysis flag comment (ACEXCO). Exclusion of a concentration depends on the analysis flag comment ACEXCO. For example, if ACEXCO is set to

- 'ALL CALC', the value will be excluded for all types of analyses based on concentrations.
- 'DESC STATS' the value will be excluded from descriptive evaluations per planned time point/time interval.
- 'HALF LIFE', the value will be excluded from half-life calculation (and, as a consequence, any calculation that relies on λ_z) only; the value is included for all other analyses.

If ACEXCO contains the addition 'TIME VIOLATION' or 'TIME DEVIATION' the value can be used for further analyses based on actual times. Excluded concentration itself will be listed in the CTR associated with an appropriate flag. Further details are given in 'Noncompartmental PK/PD Analyses of Clinical Studies' (9.7) and 'Description of Analytical Transfer Files, PK/PD Data files and ADA files' (9.10).

7.1 DEMOGRAPHIC AND OTHER BASELINE CHARACTERISTICS

Only descriptive statistics are planned for this section of the CTR, based on the TS.

The data will be summarised by treatment sequence group and in total.

7.2 CONCOMITANT DISEASES AND MEDICATION

Only descriptive statistics are planned for this section of the CTR, based on the TS.

Concomitant diseases and non-drug therapies will be coded according most recent version of the coding system of the Medical Dictionary for Drug Regulatory Activities (MedDRA). Concomitant medications will be coded according to the most recent version of the World Health Organization Drug Dictionary (WHO DD). The coding version number will be displayed as a footnote in the respective output.

In the remainder of this document, 'therapy' will be used for non-drug therapies and concomitant medications.

Section 7.2.5 of the CTP:

Previous and concomitant therapies will be presented per treatment without consideration of treatment periods.

A therapy will be considered concomitant to a treatment, if it

- is ongoing at the time of study drug administration, or
- starts within the analysis phase of the respective treatment (see Section 6.1 for a definition of treatments and analysis phases).

The diagnoses and therapies will be listed. Subjects without any concomitant diagnoses or concomitant therapies will be marked with a 'No' in the respective column.

The relevance of the concomitant therapies to the evaluation of PK data will be decided no later than at the RPM.

7.3 TREATMENT COMPLIANCE

Treatment compliance will not be analysed as a specific endpoint, but judged by observed analyte concentrations (cf. Section 5.4).

Treatment exposure to pramipexole is defined as the number of doses and total dose of pramipexole per subject and treatment.

Any deviations from complete drug intake will be addressed in the RPM and described in the CTR.

7.4 PRIMARY OBJECTIVE ANALYSIS

Independent of the main objectives stated in the CTP, this section describes further details of the primary endpoint analyses outlined in the CTP.

7.4.1 Main analysis

Section 7.2.2 of the CTP:

The statistical model used for the analysis of the primary endpoints will be an analysis of variance (ANOVA) model on the logarithmic scale. That is, the PK endpoints will be log-transformed (natural logarithm) prior to fitting the ANOVA model. This model will include effects accounting for the following sources of variation: sequence, subjects within sequences, period and treatment. The effect 'subjects within sequences' will be considered as random, whereas the other effects will be considered as fixed. The model is described by the following equation:

$y_{ijkm} = \mu + \zeta_i + s_{im} + \pi_j + \tau_k + e_{ijkm}$, *where*

y_{ijkm} = *logarithm of response measured on subject m in sequence i receiving treatment k in period j,*

μ = *the overall mean,*

ζ_i = *the* i^{th} *sequence effect, i = 1, 2,*

s_{im} = *the effect associated with the* m^{th} *subject in the* i^{th} *sequence, m = 1, 2, ...,* n_i

π_j = *the* j^{th} *period effect, j = 1, 2,*

τ_k = *the* k^{th} *treatment effect, k = 1, 2,*

e_{ijkm} = *the random error associated with the* m^{th} *subject in sequence i who received treatment k in period j.*

where $s_{im} \sim N(0, \sigma_B^2)$ *i.i.d.,* $e_{ijkm} \sim N(0, \sigma_W^2)$ *i.i.d. and* s_{im}, e_{ijkm} *are independent random variables.*

Point estimates for the ratios of the geometric means (test/reference) for the primary endpoints […] *and their two-sided 90% confidence intervals (CIs) will be provided.*

For each endpoint, the difference between the expected means for log(T)-log(R) will be estimated by the difference in the corresponding adjusted means (Least Squares Means). Additionally, their two-sided 90% confidence intervals will be calculated based on the residual error from the ANOVA and quantiles from the t-distribution. These quantities will then be back-transformed to the original scale to provide the point estimate and 90% CIs for each endpoint.

Section 7.1 of the CTP:

The assessment of bioequivalence will be based upon two-sided 90% confidence intervals (CIs) for the ratio of the geometric means (test/reference) for the primary endpoints using an

acceptance range of 80.00 – to 125.00%. This method is equivalent to the two-sided t-test procedure, each at the 5% significance level.

The following hypotheses are tested:

Null hypothesis H_0 (Inequivalence): $\mu_T - \mu_R \leq -\delta \text{ or } \mu_T - \mu_R \geq \delta$

where μ_T and μ_R are the means of the log-transformed endpoint for the test and reference treatments, respectively, and δ is the bioequivalence limit that defines the acceptance range on the logarithmic scale.

Alternative hypothesis H_a (Equivalence): $-\delta < \mu_T - \mu_R < \delta$

In this trial, the bioequivalence limit δ is ln(1.25). By back-transforming (exponentiating), this translates to an acceptance range of 80.00 to 125.00% for the ratio of the geometric means (test/reference) for endpoints on the original scale.

The rejection of the null hypothesis at the $\alpha = 0.05$ level is equivalent to the inclusion of the 90% confidence interval for $\mu_T - \mu_R$ in the acceptance range (-δ, δ).

The implementation for this analysis will be accomplished by using the CSD macros based on the PKS. The SAS code given in the additional section 10.1.1 can be used.

7.5 SECONDARY OBJECTIVE ANALYSIS

Independent of the main objectives stated in the CTP, this section describes further details of the secondary endpoint analyses.

7.5.1 Key secondary objective analysis

This section is not applicable as no key secondary endpoint has been specified in the protocol.

7.5.2 Secondary objective analysis

Section 7.2.3 of the CTP:

The secondary endpoint […] *will be calculated according to the relevant BI internal procedures and will be assessed statistically using the same methods as described for the primary endpoints*.

7.7 EXTENT OF EXPOSURE

Descriptive statistics of number of doses of pramipexole, as well as total dose, are planned for this section of the CTR, based on the TS. The date and time of drug administration will be listed for each subject.

7.8 SAFETY ANALYSIS

All safety analyses will be performed on the TS.

The safety data for treated subjects who failed to complete the study (dropouts or withdrawals) will be reported as far as their data are available.

Section 7.2.5 of the CTP:

For all analyses, the treatment actually administered (= treatment at onset) to the subject will be used (any deviations from the randomised treatment will be discussed in the minutes of the Report Planning Meeting.

7.8.1 Adverse Events

AEs will be coded using MedDRA. The coding version number will be displayed as a footnote in the respective output.

Unless otherwise specified, the analyses of AEs will be descriptive in nature. All analyses of AEs will be based on the number of subjects with AEs and NOT on the number of AEs. BI

standards as presented in 'Analysis and Presentation of AE data from clinical trials – display templates' (9.11) and 'Analysis and Presentation of AE data from clinical trials' (9.12) will be applied.

The analysis of AEs will be based on the concept of treatment-emergent AEs. That means that all AEs will be assigned to 'screening', 'on-treatment' or 'follow-up' phases as defined in Section 6.1.

An overall summary of adverse events will be presented.

Section 5.2.6.1.4 of the CTP: *No AESIs have been defined for this trial.*

According to ICH E3 (9.13), in addition to deaths and serious adverse events, 'other significant' AEs need to be listed in the CTR. These will be any non-serious adverse event that led to an action taken with study drug (e.g., discontinuation or dose reduced or interrupted).

The frequency of subjects with AEs will be summarised by treatment, primary system organ class (SOC) and preferred term (PT). Separate tables will be provided for subjects with serious AEs, for subjects with investigator-defined drug-related AEs, for subjects with investigator-defined drug-related serious adverse events, for subjects with AESIs and for subjects with AEs leading to discontinuation. In addition, the frequency of subjects with AEs will be summarised by worst intensity, treatment, primary system organ class (SOC) and preferred term (PT). The system organ classes will be sorted by default alphabetically, PTs will be sorted by descending frequency (within SOC).

In addition, for disclosure of AE data on ClinicalTrials.gov, frequencies of subjects with non-serious AEs that had an incidence of > 5% (in preferred terms) for at least one treatment will be summarised by treatment, primary SOC and PT. The frequency of subjects with SAEs will also be summarised.

For disclosure of AE data in the EudraCT register, the frequency of AEs and the frequency of non-serious AEs with an incidence of greater than 5 % (in preferred terms) and the frequency of SAEs will be summarised.

7.8.2 Laboratory data

Laboratory assessments are only planned at screening and end of study examination, after the REP period, i.e., no on-treatment analyses are feasible. Instead, laboratory data will only be listed per planned timepoint. The corresponding CTR section 15 will include a reference to the listing in section 16 stating that no on-treatment measurements are available. Laboratory data will be analysed qualitatively via comparison of laboratory data to their reference ranges. Values outside the reference range will be flagged in the data listings (cf. Section 7.2.5 of the CTP).

Clinically relevant findings in laboratory data will be reported as baseline conditions (at screening) or as AEs (during the trial) if judged clinically relevant by the investigator, and will be analysed as such (checked at the RPM at the latest).

7.8.3 Vital signs

The analyses of vital signs (blood pressure, pulse rate) will be descriptive in nature. Descriptive statistics of vital signs over time and for the change from baseline (see Section 6.7) will be provided. The results will be reported by treatment, as defined in Table 2. In addition, the time profiles of median and (min, max) will be displayed graphically by treatment.

Descriptive statistics of vital signs including change from baseline will be calculated by planned time point based on the first value of the subject at that planned time point (or assigned to that planned time point). For baseline values, the last measurement before drug administration will be used.

Clinically relevant findings in vital signs data will be reported as baseline conditions (prior to first administration of study treatment) or as AEs (after first administration of study treatment) if judged clinically relevant by the investigator, and will be analysed as such.

7.8.4 ECG

ECG recordings will be checked by the investigator for pathological results. Clinically relevant abnormal findings for ECG will be listed under 'Relevant Medical History / Baseline Conditions' (prior to first administration of study treatment) or will be reported as AEs (after first administration of study treatment), and will be analysed as such. No separate listing or analysis of continuous ECG monitoring will be prepared.

7.9 OTHER ANALYSIS

7.9.1 Biomarker analyses

No biomarker analysis is planned.

7.9.2 PK / PD analyses

No PK/PD analysis is planned.

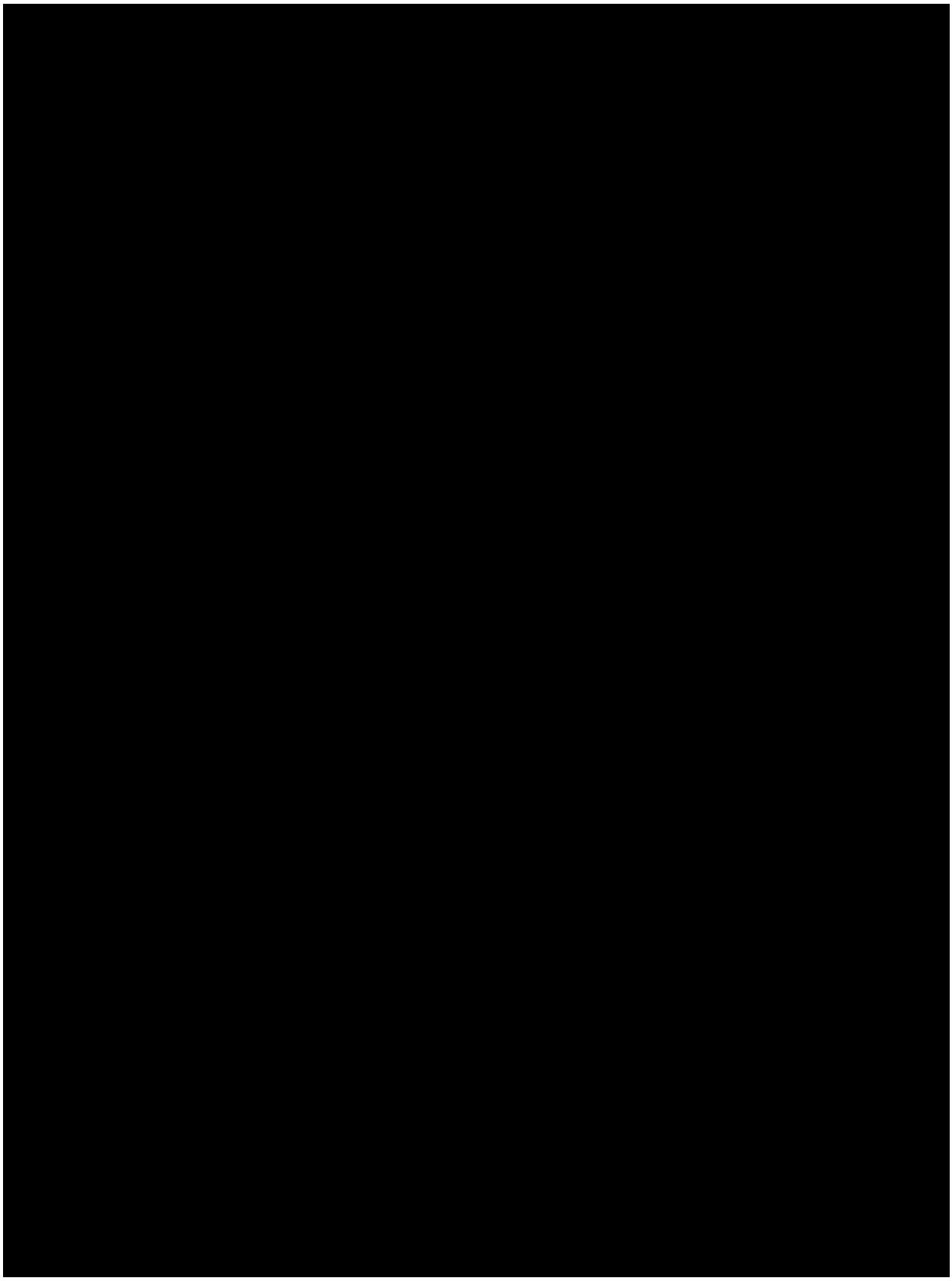
7.9.3 Physical examination

Physical examination findings will be reported as relevant medical history/baseline condition (i.e., if a condition already exists before first administration of study treatment) or as AE (if condition emerges after first administration of study treatment) and will be summarised as such.

No separate listing or analysis of physical examination findings will be prepared.

8. TIMEPOINT OF RELEASE OF TREATMENT INFORMATION

Not applicable due to open label fashion of the trial as described in the CTP Section 4.1.5. The treatment information will be loaded into the trial database at trial initiation.



11. HISTORY TABLE

Table 4 History table

Version	Date (DD-MMM-YY)	Author	Sections changed	Brief description of change
1.0	**23-AUG-24**	[redacted]	None	This is the final TSAP.