TRIAL STATISTICAL ANALYSIS PLAN

c34122641-01

BI Trial No.:	1407-0038
Title:	The effect of BI 730357 on the pharmacokinetics of rosuvastatin, digoxin, metformin and furosemide given as a cocktail – an open-label, non-randomised, 2-period fixed-sequence trial in healthy subjects Revised protocol #1, 2 (c32357152-03)
Investigational Products:	CRESTOR® (rosuvastatin), Lenoxin® (digoxin), MetfoLiquid GeriaSan® (metformin), Lasix® (furosemide), BI 730357
Responsible trial statistician:	[redacted] Phone: [redacted]
Date of statistical analysis plan:	12 Feb 2021 SIGNED
Version:	1

1. TABLE OF CONTENTS

[redacted]

[redacted]

LIST OF TABLES

2. LIST OF ABBREVIATIONS

Term	Definition / description
AE	Adverse event
[redacted]	
AESI	Adverse event of special interest
ANOVA	Analysis of variance
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
$AUC_{0-\infty}$	Area under the concentration-time curve of the analyte in plasma from 0 to infinity
[redacted]	
BI	Boehringer Ingelheim
[redacted]	
BP	Blood pressure
[redacted]	
C_{max}	Maximum measured concentration of the analyte in plasma
COVID-19	Coronavirus disease 2019
[redacted]	
CTP	Clinical trial protocol
CTR	Clinical trial report
ECG	Electrocardiogram
EoT	End-Of-Text
[redacted]	
ICH	International Council for Harmonisation
iPD	Important protocol deviations
IQRMP	Integrated quality and risk management plan
MedDRA	Medical dictionary for regulatory activities
[redacted]	
PK	Pharmacokinetics
PKS	Pharmacokinetic parameter analysis set

Term	Definition / description
PR	Pulse rate
REP	Residual effect period
RPM	Report planning meeting
SAE	Serious adverse event
SOC	System organ class
TS	Treated set
TSAP	Trial statistical analysis plan

3. INTRODUCTION

As per ICH E9, 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 trial statistical analysis plan (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.

SAS® Version 9.4 will be used for all analyses.

Pharmacokinetics (PK) parameters will be calculated using Phoenix WinNonlinTM software (version 6.3).

4. CHANGES IN THE PLANNED ANALYSIS OF THE STUDY

All analyses described in this TSAP are in accordance with the statistical methods described in the revised CTP.

5. ENDPOINTS

5.1 PRIMARY ENDPOINTS

As defined in Section 2.1.2 and 7.3.1 of the CTP, the primary endpoints are the following:

The change from visit 2 of

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

of rosuvastatin, digoxin, metformin and furosemide in plasma measured at visit 3 at steady state conditions of BI 730357.

5.2 SECONDARY ENDPOINT

5.2.1 Key secondary endpoint

Not applicable, as no key secondary endpoints were defined.

5.2.2 Secondary endpoint

The secondary endpoint is as defined in the Section 2.1.3 and 7.3.2 of the CTP:

The change from visit 2 of

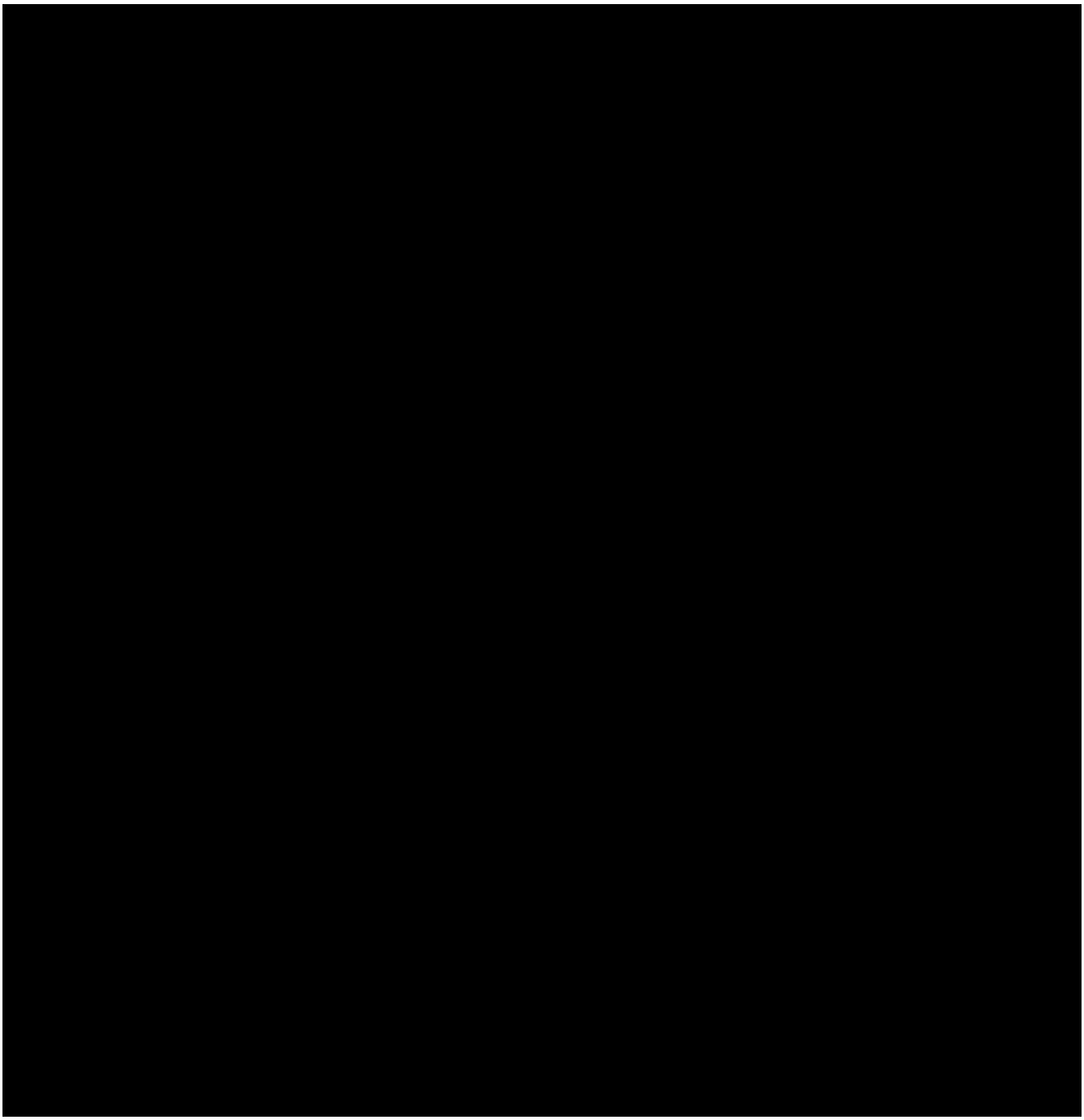
- AUC_{0-tz} (area under the concentration-time curve of the analyte in plasma over the time interval from 0 to the last quantifiable data point)

of rosuvastatin, digoxin, metformin and furosemide in plasma measured at visit 3 at steady state conditions of BI 730357.

5.3.3 Safety parameters

Further safety parameters will be used as defined in Section 2.2.2.5 and Section 5.2 of the CTP:

- Adverse events (AEs) (including clinically relevant findings from the physical examinations)
- Safety laboratory tests
- 12-lead Electrocardiograms (ECGs)
- Vital signs (systolic and diastolic blood pressures (BP), pulse rate (PR))



6. GENERAL ANALYSIS DEFINITIONS

6.1 TREATMENTS

For information on treatments to be administered, assignment of treatment groups, selection of doses, cf. Section 4 of the CTP.

The study will be performed as a non-randomised, open-label, 2-period fixed-sequence trial. In the first treatment period, subjects will receive a single dose of the cocktail (consisting of rosuvastatin, digoxin, metformin and furosemide) on Day 1 (reference treatment). In the second treatment period, the subjects will receive BI 730357 twice daily on Day -7 to Day 6 (13 days) with an administration of a single dose of the cocktail on Day 1 one hour after the morning administration of BI 730357 (test treatment). [redacted] The sequence of these treatment periods is fixed and the same for all subjects.

An overview of all relevant trial activities is provided in the Flow Charts in the CTP. For visit schedule and details of trial procedures at selected visits, refer to Sections 6.1 and 6.2 of the CTP, respectively.

For statistical analyses of safety, separate analysis phases will be defined for each subject as given in Table 6.1:1. The phase ‘Cocktail + BI’ refers to the time interval, where the cocktail REP of the first cocktail administration overlaps with the start of the BI dosing in period 2. The phase ‘BI + Cocktail’ refers to the time after the second cocktail administration with BI at steady state.

AE displays in CTR Section 15, Appendix 16.1.13.1.8.2 and Appendix 16.1.13.1.8.3 will present results for the four on-treatment phases only.

In CTR Section 15 AE tables (but not in Appendix 16.1.13.1.8.2 and Appendix 16.1.13.1.8.3 AE tables), the following total will be provided in addition:

- "Total on-trt", defined as the total over all on-treatment phases.

In Appendix 16.1.13.1.8.2 and Appendix 16.1.13.1.8.3 no total columns will be included in the tables.

CTR Appendix 16.1.13.1.8.1 displays will present results for the screening, on-treatment and follow-up phases.

Additionally to the total defined above, the following total will be provided in CTR Section 16.1.13.1.8.1 AE tables:

- "Total", defined as the total over all study phases (screening + on-treatment + follow-up).

Table 6.1: 1 Analysis phases

Study analysis phase	**Label**	**Start**	**End**
Screening	Screening	Date of informed consent	Date/time of first administration of the cocktail
On treatment cocktail	Cocktail	Date/time of first administration of the cocktail	Date/time of first administration of BI 730357 or date/time of first administration of the cocktail + Residual effect period (REP) (10 * 24h), whatever occurs earlier
On treatment Cocktail + BI 730357	Cocktail + BI	Date/time of first administration of BI 730357	Date/time of first administration of the cocktail + Residual effect period (REP) (10 * 24h)
On treatment BI 730357	BI	Date/time of first administration of BI 730357 or date/time of first administration of the cocktail + Residual effect period (REP) (10 * 24h), whatever occurs later	Date/time of second administration of the cocktail
On treatment Steady state BI 730357 + cocktail	BI + Cocktail	Date/time of second administration of the cocktail	Date/time of last administration of BI 730357 + Residual effect period (REP) (7 * 24h) Or 12:00 a.m. on day after subject's trial termination date, whatever occurs earlier.
Follow-up	F/U	Date/time of last administration of BI 730357 + REP (7 * 24h)	12:00 a.m. on day after subject's trial termination date

Statistical analyses of [redacted] PK parameters will be conducted by treatment period (cocktail vs. BI + cocktail).

More details on the technical implementation of these analyses are provided in the ADS Plan of this TSAP.

According to Section 1.2.6 of the CTP, the REP of the cocktail is 10 days. This is the period after the last dose with measurable drug levels and/or pharmacodynamic effects still likely to be present. [redacted]

6.2 IMPORTANT PROTOCOL DEVIATIONS

Table 6.2: 1 Handling of important protocol deviations (iPDs)

iPD code	iPD Category & Brief Description	Excluded from which analysis set
A1	Inclusion Criteria Not Met	None
A2	Exclusion Criteria violated	None
B1	Informed consent not available/not done	TS, PKS[redacted]
B2	Informed consent too late	None
C1	Incorrect trial medication intake	PKS[redacted]
C3	Non-compliance	PKS[redacted]
C4	Medication code broken inappropriately	None
C5	Incorrect intake of trial medication	PKS[redacted]
C6	Improper washout between treatments	PKS[redacted]
D1	Prohibited medication use	PKS[redacted]
D2	Mandatory medication not taken	PKS[redacted]
D3	Improper washout of concomitant medication	PKS[redacted]
E1	Certain violations of procedures used to measure primary or secondary data	PKS[redacted]
F1	Certain violations of time schedule used to measure primary or secondary data	PKS[redacted]
G1	Incorrect intake of meal	PKS[redacted]

Table 6.2: 2 Handling of non-important COVID-19 related protocol deviations

PD code	PD Category	Excluded from which analysis set
Q1	Missed examination	None
Q2	(Partially) missed visit	None
Q3	Drug shipment	None

Consistency check listings (for identification of deviations from 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 iPD and whether it is COVID-19 related. For definition of iPDs, and for the process of identification of these, refer to the BI reference document "Identify and Manage Important Protocol Deviations (iPD)" (1).
If any iPDs are identified, they are to be summarised into categories and will be captured in the RPM minutes. Categories which are considered to be iPDs in this trial are defined in the integrated quality and risk management plan (IQRMP). If the data show other iPDs, the definition in the IQRMP will be supplemented accordingly by the time of the RPM.

IPDs will be summarised and listed. Additionally, protocol deviations associated with COVID-19 disruption (non-important and important protocol deviations) will be summarised and listed.

6.3 SUBJECT SETS ANALYSED

The following subject sets will be defined for statistical analysis:

- Treated set (TS):

  The treated set includes all subjects who signed informed consent and were treated with at least one dose of study drug.

  The treated set will be used for safety analyses.

- Pharmacokinetic parameter analysis set (PKS):

  This set includes all subjects in the 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 subsection ‘Pharmacokinetics’ in section 7.3 in the CTP). Thus, a subject will be included in the PKS, even if he/she 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. Thus, the primary and secondary analyses will be based on the PKS.

Relevant protocol deviations may be

• Incorrect trial medication taken

• Incorrect dose of trial medication taken

• Use of restricted medications

Plasma concentrations and/or parameters of a subject will be considered as non-evaluable, if for example

• The subject experienced emesis that occurred at or before two times median tmax of the respective treatment (Median tmax is to be determined excluding the subjects experiencing emesis),

• A predose concentration of BI 730357 is >5% Cmax value of that subject in the respective treatment period

• Missing samples/concentration data at important phases of PK disposition curve

[redacted]

The discussion of all exceptional cases and problems and the decisions on the allocation of subjects to analysis sets will be made at latest at the RPM.

Table 6.3: 1 Subject sets analysed

	Subject set	
Class of endpoint	TS	PKS
Disposition	x	
Treatment exposure	x	
IPDs	x	
Demographics/baseline endpoints	x	
Safety parameters	x	
PK endpoints		x

[redacted]

[redacted]

6.5 POOLING OF CENTRES

This section is not applicable, because the study was performed in only one centre.

6.6 HANDLING OF MISSING DATA AND OUTLIERS

In CTP Section 7.5 the handling of missing data is defined.

Data of screened subjects who were withdrawn from the trial prior to first administration of any study drug will not be reported in the CTR.
Data of subjects who failed to complete all periods of the study (dropouts or withdrawals) will be reported in the CTR as far as their data are available. All withdrawals will be documented and the reason for withdrawal recorded in the CTR.

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 (2).

Missing data and outliers of PK data are handled 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. PK data will be excluded of analysis as described in Section 7.3 of the CTP.

6.7 BASELINE, TIME WINDOWS AND CALCULATED VISITS

Baseline will be defined as the last available value before the administration of the cocktail in period 1 (Visit 2). Note that this baseline definition will be used for evaluation of data of all on-treatment periods (“cocktail”, “BI + cocktail”, “BI”, “cocktail + BI”, as defined in Section 6.1).

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

Unscheduled measurements of laboratory data or 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. Descriptive statistics of on-treatment laboratory data will be calculated by planned time point based on the worst value of the subject at that planned time point (or assigned to that planned time point). For vital signs, descriptive statistics will be calculated by planned time point based on the last value of the subject at that planned time point (or assigned to that planned time point).

7. PLANNED ANALYSIS

The format of the listings and tables will follow the standards defined in the BI corporate guideline "Reporting of clinical trials and project summaries" (3).

For End-Of-Text (EoT) tables, the set of summary statistics of continuous variables will be:

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

For plasma concentrations as well as for all PK parameters the following descriptive statistics will additionally be calculated:

CV	arithmetic coefficient of variation
gMean	geometric mean
gCV	geometric coefficient of variation

For PK parameters the following descriptive statistics will additionally be calculated:

P10	10th percentile
Q1	1st quartile
Q3	3rd quartile
P90	90th percentile

The data format for descriptive statistics of plasma concentrations will be identical with 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 and will display the number of observations in a category as well as the percentage (%) relative to the respective treatment group (unless otherwise specified, all subjects in the respective subject set whether they have non-missing values or not).

Percentages will be rounded to integer numbers. The category missing will be displayed if and only if there actually are missing values.

[redacted]

7.2 CONCOMITANT DISEASES AND MEDICATION

Concomitant diseases will be coded according to the most recent version of Medical Dictionary For Regulatory Activities (MedDRA). Concomitant medication will be coded according to the most recent version of the World Health Organisation – Drug Dictionary.

CTP Section 7.3.4:
Previous and concomitant therapies will be presented per treatment group without consideration of time intervals and treatment periods.

Clarification: As all subjects belong to the same treatment group, previous and concomitant therapies will be presented for all subjects together.

A medication will be considered concomitant, if it

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

Concomitant medication and drug therapies will be grouped by preferred term.

Only descriptive statistics are planned for this section of the report.

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

7.3 TREATMENT COMPLIANCE

Treatment compliance will not be analysed as a specific endpoint.

7.4 PRIMARY ENDPOINTS

Analysis of relative bioavailability of primary endpoints $AUC_{0-\infty}$ and C_{max} of rosuvastatin, digoxin, metformin and furosemide in plasma will be performed in the PKS as defined in Sections 7.1 and 7.3.1 of the CTP. The analysis will be descriptive in nature.
There will be eight ANOVA models, as there are two primary endpoints and four drugs to be analysed.

The statistical model for the primary analysis defined in the CTP is an analysis of variance (ANOVA) model on the logarithmic scale including "treatment" as fixed effect and "subject" as random effect. [redacted]

[redacted]

Primary PK endpoints will be assessed descriptively. The analysis of standard PK parameters is calculated according to the relevant BI internal procedures. See Sections 7.3 and 7.5.2 of the CTP for details regarding exclusion of PK parameters and plasma concentrations.

7.5 SECONDARY ENDPOINT

7.5.1 Key secondary endpoints

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

7.5.2 Secondary endpoint

Analysis of relative bioavailability of the $AUC_{0\text{-}tz}$ of rosuvastatin, digoxin, metformin and furosemide in plasma will be performed in the same way as for the primary endpoints. This results in 4 ANOVA models: for rosuvastatin, digoxin, metformin and furosemide.

Additionally, the secondary PK endpoint will be assessed descriptively.

The analysis of standard PK parameters is calculated according to the relevant BI internal procedures. See Sections 7.3 and 7.5.2 of the CTP for details regarding exclusion of PK parameters and plasma concentrations.

[redacted]

7.6.1 Safety parameters

Safety and tolerability will be analysed as described in Section 7.8 of this TSAP.

[redacted]

[redacted]

7.8 SAFETY ANALYSIS

The safety analyses are described in the CTP Section 7.3.4. All safety analyses will be performed on the TS.

The safety analyses will be descriptive in nature. No hypothesis testing is planned.

CTP 7.3.4:
Measurements (such as ECG, vital signs, or laboratory parameters) or AEs will be assigned to treatments (see Section 4.1) based on the actual treatment at the planned time of the measurement or on the recorded time of AE onset (concept of treatment emergent AEs). Therefore, measurements planned or AEs recorded prior to first intake of trial medication will be assigned to the screening period, those between trial medication intake and end of REP (see Section 1.2.3) will be assigned to the treatment period. Events occurring after the REP but prior to next intake or end of trial termination date will be assigned to 'follow-up'. In case of two or more treatments, the follow-up will be summarized according to the previous treatment. These assignments including the corresponding time intervals will be defined in detail in the TSAP. Note that AEs occurring after the last per protocol contact but entered before final database lock will be reported to Pharmacovigilance only and will not be captured in the trial database.

7.8.1 Adverse Events

AEs will be coded with the most recent version of MedDRA. The analysis 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.

The analysis of AEs will be based on the concept of treatment emergent AEs. That means that all AEs occurring between first drug intake till last drug intake + REP will be assigned to the on-treatment phases as defined in Table 6.1:1. All AEs occurring before first drug intake will be assigned to 'screening' and all AEs occurring after the last drug intake and the REP will be assigned to 'follow-up'. For details on the treatment definition, see Section 6.1.

AEs will be analysed based on the actual treatment at the recorded time of AE onset. Any deviations from the planned treatment will be discussed in the minutes of the RPM.

Pre-defined adverse events of special interest (AESIs) are defined in the CTP Section 5.2.5.1.4. Hepatic injury, severe infections and opportunistic and mycobacterium tuberculosis infections are considered AESIs. The investigator classifies on the eCRF whether an observed AE was an AESI or not.

According to ICH E3 (3), in addition to Deaths and Serious Adverse Events and AESIs, 'other significant' AEs need to be listed in the clinical trial report. 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). SARS-CoV-2 infections and AEs related to SARS-CoV-2 will also be listed.

An overall summary of AEs will be presented.

The frequency of subjects with AEs will be summarised by treatment, primary system organ class (SOC) and preferred term. AEs which were considered by the investigator to be drug related (primary endpoint) will be summarised separately. A separate table will also be provided for subjects with serious adverse events (SAEs). A table will be provided for subjects with pre-specified AESIs, summarised by SOC and PT. The frequency of subjects with AEs will also be summarised by maximum intensity, primary SOC and preferred term.

The SOC will be sorted alphabetically, preferred terms will be sorted by frequency (within SOC).

Post-hoc AESIs may be defined at a later time point and summarised separately.

For further details on summarization of AE data, please refer to "Analysis and Presentation of Adverse Event Data from Clinical Trials" (4) and "Handling of missing and incomplete AE dates" (5).

For disclosure of AE data on ClinicalTrials.gov, the frequency of subjects with non-serious AEs occurring with an incidence of greater than 5 % (in preferred terms) will be summarised by treatment, primary SOC and preferred term. The frequency of subjects with SAEs will also be summarised.
For disclosure of AE data in the EudraCT register, the frequency of subjects with AEs, the frequency of subjects with non-serious AEs with an incidence of greater than 5 % (in preferred terms) and the frequency of subjects with SAEs will be summarised. Furthermore, the total number of treated subjects by country and by age group will be summarised.

For support of lay summaries, the frequency of subjects with drug-related SAEs will be summarised by treatment, primary system organ class and preferred term.

For the subgroup of subjects with SARS-CoV-2 infection while on treatment with study drug the number of subjects with AEs, with AEs by SOC and preferred term, with AEs leading to discontinuation by SOC and preferred term and with SAEs by SOC and preferred term will be summarised.

7.8.2 Laboratory data

CTP 7.3.4:
Laboratory data will be compared to their reference ranges. Values outside the reference range as well as values defined as possibly clinically significant will be highlighted in the listings. Additionally, differences from baseline will be evaluated.

The analyses of laboratory data will be descriptive in nature and will be based on BI standards ″Display and Analysis of Laboratory Data" (6).

Analyses will be based on normalised values, which means transforming to a standard unit and a standard reference range. The original values will be analysed if the transformation into standard unit is not possible for a parameter.

Descriptive statistics of laboratory values over time and for the difference from baseline (see Section 6.7) will be provided. Frequency tables of changes between baseline and last value on treatment with respect to the reference range will be presented.

Possibly clinically significant abnormal laboratory values are only those identified either in the Investigator's comments or at the RPM at the latest. It is the Investigator's responsibility to decide whether a lab value is clinically significant abnormal or not. Standard or project-specific rules for flagging clinically significant values in an automated manner will not be applied in this study.

Clinically relevant changes in laboratory test results will be reported as AE and summarised as such.

Urine creatinine will be excluded from the tables and listings of safety laboratory data. A separate listing of urine creatinine data will be provided.

7.8.3 Vital signs

Descriptive statistics including change from baseline (see Section 6.7) are planned for this section of the report.
Clinically relevant changes in vital signs will be reported as AE and summarised as such.

7.8.4 Others

Physical examination findings as well as relevant ECG findings will be reported as AE and will be summarised as such. No separate listing or analysis of physical examination or ECG findings will be prepared.

8. REFERENCES

1.	*001-MCS-40-413:* "Identify and Manage Important Protocol Deviations (iPD)", current version, Group "Clinical Operations", IDEA for CON.
2.	*BI-KMED-BDS-HTG-0035*: "Handling of missing and incomplete AE dates", current version; KMed
3.	*BI-KMED-BDS-HTG-0045*: "Standards for Reporting of Clinical Trials and Project Summaries", current version, KMed
4.	*BI-KMED-BDS-HTG-0066*: "Analysis and Presentation of Adverse Event Data from Clinical Trials", current version, Kmed
5.	*BI-KMED-BDS-HTG-0035*: "Handling of missing and incomplete AE Dates", current version, Kmed
6.	*BI-KMED-BDS-HTG-0042*: "Display and Analysis of Laboratory Data", current version, Kmed

10. HISTORY TABLE

Table 10: 1 History table

Version	Date (DD-MMM-YY)	Author	Sections changed	Brief description of change
1	**12-Feb-21**	[redacted]	None	This is the final TSAP.