STATISTICAL ANALYSIS PLAN

For:

Tempero Bio Inc.

PROTOCOL No. TMP-301-HNV-101

PROTOCOL VERSION: Final v3.3 (Amendment 03), 2023/07/18

A Phase 1, Randomized, Placebo Controlled, Multiple Ascending Dose (MAD) Study to Evaluate the Safety, Tolerability, and Pharmacokinetics of TMP-301 in Healthy Subjects

Altasciences Project No. TPM-P1-726

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STATISTICAL ANALYSIS PLAN APPROVAL

We have carefully read this statistical analysis plan and agree it contains the necessary information required to handle the statistical analysis of study data.

VERSION CONTROL

Version	Date	Author	Description of Changes
Final V 1.0	2023/10/23	Commercially Confidential Information	Not Applicable

TABLE OF CONTENTS

TABLE OF TABLES

ABBREVIATIONS

AE	adverse event
ANOVA	analysis of variance
aPTT	partial thromboplastin time
AUC0-TAU,FD	AUC from time zero to the time the end of the 12-hr dosing interval after the first dose
BID	twice daily
BMI	body mass index
BPRS	Brief Psychiatric Rating Scale
CADSS	Clinician-Administered Dissociative States Scale
CFB	change from baseline
CI	confidence interval
CL/F	total plasma clearance
Cmax,FD	maximum concentration after the first dose
Cmin,FD	minimum concentration after the first dose
CRF	case report form
CRU	clinical research unit
C-SSRS	Columbia Suicide Severity Rating Scale
CSR	clinical study report
CV%	percentage coefficient of variation
DMP	data management plan
DTS	deviation tracking system
ECG	electrocardiogram
E_{max}	maximum postdose effect
E_{min}	minimum postdose effect
EOS	end of study
FSH	follicle stimulating hormone
HIV	Human immunodeficiency virus
ICF	informed consent form
ICH	International Conference on Harmonisation
IP	Investigational product
λ_Z	elimination rate constant
LLOQ	lower limit of quantitation
ln	natural log
LSMeans	least-square means
MAD	multiple ascending dose
Max	maximum
Mean	arithmetic mean
MedDRA	Medical Dictionary for Regulatory Activities

Min	minimum
3MS	Modified Mini-Mental State Examination
n	number of available data
N	number of observations
NCA	non-compartmental analysis
NR	not reported
PK	pharmacokinetic(s)
PT	preferred term
Q1	first quartile
Q3	third quartile
R^2	coefficient of determination
SAE	serious adverse event
SAP	statistical analysis plan
SD	standard deviation
SE	standard error
SOC	system organ class
SOP	standard operating procedure
SS	steady-state
T_{max}	time of maximum concentration
Tmax,FD	time of maximum concentration after the first dose
TEAE	treatment-emergent adverse event
TE_{max}	time to maximum postdose effect
TE_{min}	time to minimum postdose effect
TFLs	tables, figures, and listings
TS	total score
UDS	urine drug screen
VAS	Visual Analogue Scale
Vz/F	volume of distribution
WHODrug	World Health Organization Drug Dictionary

1 INTRODUCTION

This study is a multiple ascending dose (MAD) study to evaluate the safety, tolerability, pharmacokinetics (PK) of TMP-301 in healthy subjects. There will be up to 4 dose cohorts, and within each cohort subjects will be randomly assigned to receive either active drug or placebo. Study subjects will be administered active drug or placebo once daily (qd) or twice daily (bid) for 14 days (note: for bid cohorts, only the AM dose is administered on Day 14).

This Statistical Analysis Plan (SAP) provides a detailed description of the statistical methods and procedures to be implemented for the analyses of data from the Clinical Study Protocol TPM-P1-726 (TMP-301-HNV-101). The analyses described in the SAP are based upon the final version of the Clinical Study Protocol 4.0 (Amendment 04), dated 2023/10/02.

2 STUDY OBJECTIVES

The objectives of the study and corresponding study endpoints are detailed in Table 1.

Table 1: Objectives and Related Endpoints

Objective	Endpoint
Primary	
• To assess the safety and tolerability of TMP-301 following multiple ascending oral doses	The primary endpoint is safety assessed after multiple-dose administrations by: • Adverse events (AE) • Clinical laboratory tests • Vital sign measurements • 12-lead electrocardiograms (ECG) • Physical examinations • Psychiatric assessments (Brief Psychiatric Rating Scale [BPRS], Clinician-Administered Dissociative States Scale [CADSS], Columbia Suicide Severity Rating Scale [C-SSRS], Modified Mini-Mental State Examination (3MS) and Alertness Visual Analogue Scale [VAS])
Secondary	
• To assess the pharmacokinetics (PK) profile of TMP-301 following multiple ascending oral doses	• Day 1: Partial areas defined as AUC_{0-12} and AUC_{0-24}; maximum concentration (C_{max}), time of maximum concentration (T_{max}), concentration at 12 h (C_{12}), concentration at 24 h (C_{24}) where $C_{24} = C_{trough\ Day2}$ • Day 14 (or last day of dosing): AUC_{ss} from time zero to the time the end of the (12 or 24 hour) dosing interval ($AUC_{0-TAU,ss}$. For BID cohorts, $AUC_{0-TAU,ss} = AUC_{0-12,SS}$, and for QD cohorts, $AUC_{0-TAU,ss} = AUC_{0-24,SS}$; $C_{avg,ss}$, $C_{max,ss}$, $T_{max,ss}$, $C_{12,ss}$, C_{24ss}, and $C_{min,SS}$, all at steady-state (SS), along with apparent total plasma clearance (CL/F) and apparent volume of distribution (V_z/F)
Exploratory	

• To assess endpoints related to TMP-301 mechanism of action • To evaluate CYP1A2 genotypes and their association with phenotypes for potential impact on metabolism and to identify biomarkers of potential response to TMP-301.	Accumulation ratio derived as: • AR_$AUC_{0\text{-}12\ BID}$= $AUC_{0\text{-}12,ss}$ / $AUC_{0\text{-}12,\ Day\ 1}$ • AR_$AUC_{0\text{-}24QD}$ = $AUC_{0\text{-}24,ss}$ / $AUC_{0\text{-}24,\ Day\ 1}$ • AR_C_{max} = $C_{max,ss}$ / $C_{max,FD,\ Day\ 1}$ • AR_C_{12} = $C_{12,ss}$ / $C_{12,\ Day\ 1}$ • AR_C_{24}= $C_{24,ss}$ / $C_{24,\ Day\ 1}$ • Estimations of dose proportionality at steady State. Dose–normalized PK parameters (C_{max}, C_{12}, C_{24}, and $AUC_{0\text{-}12}$ and $AUC_{0\text{-}24}$, when appropriate) will be assessed graphically for dose-proportionality For BID cohorts, $AUC_{0\text{-}TAU,SS}$ = $AUC_{0\text{-}12,SS}$, For QD cohorts, $AUC_{0\text{-}TAU,SS}$ = $AUC_{0\text{-}24,SS}$ • Estimation of time to achieve steady state (C_{trough} will be displayed graphically and summarized descriptively by day to assess for steady state) • As a marker of CYP1A2 activity, the ratio of paraxanthine to caffeine concentration (at 4 hours post caffeine dose) will be reported on Day -1 and Day 14. Change in CYP1A2 activity will be derived as CYP1A2_ratio = paraxanthine/caffeine concentration day 14 / paraxanthine/caffeine concentration day -1

Abbreviations: AE = adverse event; ECG = electrocardiogram; PK = pharmacokinetics

3 STUDY DESIGN

3.1 General Description

This study will be a randomized, double-blind, placebo -controlled, fixed -sequence, MAD study. The study will be conducted in a single clinical research unit (CRU). The study will consist of up to 4 cohorts. Each cohort will consist of 8 subjects (6:2 for active: placebo), for a maximum total sample size of approximately 32 subjects. Subjects will only participate in 1 cohort.

Screening will occur within approximately 28 days prior to the first scheduled study drug administration. Screening data will be reviewed to determine subject eligibility. Subjects who meet all inclusion criteria and none of the exclusion criteria and who consent to participation will be admitted to the CRU for baseline evaluations prior to dosing. All baseline safety results should be available prior to the first study drug administration.

In cohort 2, subjects will be fasted overnight for 10 hours prior to the morning dose, followed by a 2 hour fast. Subjects were fasted for 2 hours prior to dosing and 2 hours following the evening dose for the cohort 1 (50 mg bid). If dosing in a fed condition is selected for cohort 3 or 4, subjects will be fasted overnight for approximately 9 hours, then given breakfast followed by study treatment dosing 30 minutes after initiation of breakfast.

Each subject will be randomly assigned to 1 of the following cohorts:

Cohort 1:
- Group 1 (6 subjects): 50 mg of TMP-301 bid
- Group 2 (2 subjects): placebo (matching TMP-301) bid

Cohort 2:
- Group 3 (6 subjects): 50 mg of TMP-301 qd
- Group 4 (2 subjects): placebo (matching TMP-301) qd

Cohort 3:
- Group 5 (6 subjects): Dose regimen of TMP-301 to be determined
- Group 6 (2 subjects): placebo (matching TMP-301)

Cohort 4:
- Group 7 (6 subjects): Dose regimen of TMP-301 to be determined
- Group 8 (2 subjects): placebo (matching TMP-301)

Safety will be assessed and blood samples for PK will be collected throughout confinement. Subjects will be discharged from the CRU on Day 18. Subjects will return to the CRU on Day 25 for a follow-up visit and End-of-Study (EOS) procedures.

Caffeine (100 mg) will be included as probe CYP1A2 substrate in cohort 2 and subsequent cohorts. Ratio of plasma paraxanthine/caffeine will be determined in a blood sample 4 hours after administration of caffeine at baseline (Day-1) and on day 14.

The maximum duration of subject participation, including Screening, will be approximately 53 days. Subjects who terminate the study early will perform follow-up procedures at the time of Early Termination.

Based on safety and tolerability findings in any of the cohorts, a 14-day dose titration cohort to evaluate the impact on treatment-emergent adverse events (TEAEs) may be added as an additional cohort (7 days titration + 7 days stable dosing).

3.2 Treatments

The following treatments will be administered according to Table 2.

- IP: TMP-301 oral capsule
- Placebo: Matching placebo

Table 2: Dose Cohorts

Cohort	N (Active:Placebo)	Dose	Fasting Status	Drug Administration
1	6:2	Twice daily dose of 50 mg	Fasting Pre & 4 hours post dose	Twice daily TMP-301 or placebo dosing from Days 1 to 13 and once in the morning on Day 14 (for a total of 27 consecutive study drug administrations) or Once daily TMP-301 or placebo to be administered in the mornings of Days 1-14.
2	6:2	Once daily dose of 50 mg	Fasting Pre & 2 hours post dose or Fed 30 minutes pre-dose	
3 (Optional)	6:2	Dose and regimen to be determined		
4 (Optional)	6:2	Dose and regimen to be determined		

Caffeine (100 mg) will also be administered to enrolled subjects as part of the study on Day -1 and Day 14.

3.3 Study Procedures

For complete details on the study assessments to be performed for each cohort, refer to APPENDIX A.

3.4 Randomization and Unblinding Procedures

3.4.1 Method of Assigning Subjects to Treatment Groups

Altasciences will generate the randomization code with a computer program according to the study design, the number of subjects and the number of treatments. Within each cohort, subjects will be randomized (3:1) to receive TMP-301 or placebo. The random allocation of each IP to each subject will be done in such a way that the study is balanced. Once generated, the randomization code will be final and will not be modified.

Subjects who sign the ICF and are randomized but do not receive the study treatment may be replaced. Subjects who sign the ICF, are randomized and receive the study treatment, and

subsequently withdrawn as a result of AEs thought to be related to the study drug will not be replaced. Subjects who sign the ICF, are randomized and receive the study treatment, and are subsequently withdrawn for reasons not related to the study drug may be replaced.

3.4.2 Blinding

The randomization code may be made available to the personnel of the bioanalytical facility. The treatment assignment will not be known by the study participants.

Furthermore, the randomization code will not be available to the physician and clinical staff involved in the collection, monitoring, revision, or evaluation of AEs, as well as clinical staff who could have an impact on the outcome of the study, and including the pharmacokineticist (or delegate), until database lock, or designation that unblinding information is required by the SRC to determine the next dose.

The preparation of the products will be done by designated personnel that are not directly involved in the clinical aspects of the trial.

The randomization code must not be broken except in emergency situations where the identification of a subject's study treatment is required by an Investigator for further treatment to the subject or to complete a SAE report. Randomization information will be held by designated individual(s). The date and reason for breaking the blind must be recorded.

The results of the PK analyses will be made available only to the personnel responsible for evaluating the safety data before proceeding with the next dose level.

3.5 Determination of Sample Size

No formal sample size analysis was performed. It is estimated that approximately 24 subjects should be sufficient to meet the objectives of the study.

4 ANALYSIS POPULATIONS

- **Enrolled Population**

The Enrolled population will include all subjects who signed the informed consent form (ICF).

- **Screened Population**

The Screened population will include all subjects who meet the eligibility criteria.

- **Randomized Population**

The Randomized population will include all subjects who are assigned a randomization number.

- **Safety Population**

The Safety population will include all subjects who received at least 1 dose of one of the investigational product (IP) or placebo. The number of subjects who were included, who discontinued, and who completed the study will be tabulated. The primary reasons for discontinuation will be provided.

- **Pharmacokinetic (PK) Population**

The PK population will include all subjects who have received at least 1 dose of the investigational product and have sufficient PK data to derive at least 1 PK parameter.

Subjects who do not complete the sampling schedule may be included in the PK analysis for only the PK parameters that are judged not to be affected by the missing sample(s).

5 DATA HANDLING AND PRESENTATION

All safety and statistical outputs will be generated using SAS software, version 9.4 or higher. The PK outputs will be generated using WinNonlin version 8.0 or higher.

All programs used to generate statistical analyses will be validated according to Altasciences' Standard Operating Procedures (SOPs).

The analyses described in this plan are considered a priori, in that they have been defined prior to database lock and prior to breaking the blind. Any analyses performed subsequent to database lock and breaking the blind will be considered post hoc and exploratory, and will be identified in the Clinical Study Report (CSR).

5.1 Safety Analysis Presentation

Adverse events and medical history will be classified using the Medical Dictionary for Regulatory Activities (MedDRA) terminology as defined in the study Data Management Plan (DMP).

Prior and concomitant medications will be coded with the World Health Organization Drug Dictionary (WHODrug) as defined in the study DMP.

In general, all safety summary tables will be presented for the Safety population. Summaries for AEs will be presented by treatment. Summaries for other safety endpoints will be presented by treatment if the endpoints are measured end of Study only.

In general, the data listings will include all subjects in the Randomized population up to the point of study completion or discontinuation; exceptions will be listings pertaining to a subset of subjects only (eg, subjects with protocol deviations) or a subset of records/events (eg, abnormal laboratory values). Limited data for those not included in the Randomized population, eg, who failed screening, may be presented, as appropriate.

Categorical variables will be summarized using number of observations (N), number of available data (n), and the percentage of available data (%) for each class. Continuous variables will be summarized using descriptive statistics, including N, n, arithmetic mean (mean), standard deviation (SD), minimum (min), median, and maximum (max).

The following general considerations may be applied:

- Study Day will be derived from the reference date (eg, the day of the first dose of study drug) and date of event as:

  Study Day = (date of event – reference date) +1, if event is on or after the reference date;

  Study Day = date of event – reference date, if event is before the reference date.

- Duration will be calculated using the general formula: (end date - start date) +1.
- If the reported value of a clinical laboratory parameter cannot be used in a statistical summary table (eg, a character string is reported for a parameter of the numerical type), a coded value may be appropriately determined and used in the statistical analyses. In general, a value below the lower limit of normal range such as '<10' or '≤5' will be treated as half of the lower limit, '5' or '2.5', respectively, and a value above the upper limit of normal range such as '>100' will be replaced with a number deemed

scientifically reasonable, eg, 101. However, the actual values as reported in the database will be presented in data listings.

- Unscheduled assessments will not be included in descriptive summary, except for baseline consideration. When assessments are repeated for a given time point or performed at unscheduled times, only the result which is the closest to the dosing time will be included in summary tables.

In general, summary statistics for raw variables (ie, variables measured at the study site or central laboratory) will be displayed as follows:

- Minima and maxima will be displayed to the same number of decimal places as the raw data
- Means, medians, and quartiles (if presented) will be displayed to 1 additional decimal place
- Standard deviations will be displayed to 2 additional decimal places
- Percentages will be displayed to 1 decimal place; Percentages between 0 and 0.1 (exclusive) will be displayed as '<0.1'
- P-values will be displayed to 3 decimal places; P-values that are less than 0.001 will be displayed as '<0.001'

Derived variables (ie, variables that are not measured by the study site but are calculated for analysis based on other measured variables) will be displayed with the same precision as the raw data, as appropriated, then follow the similar rules in presentation of summary statistics.

5.2 Pharmacokinetic Analysis

In general, all PK summary tables will be presented for the PK population.

Individual raw PK concentrations will be displayed with the same precision as received from the bioanalytical laboratory.

Precision for individual PK parameters will be displayed as follows:

- C_{max}, $C_{min,}$ CL/F, Vz/F and AUCs will be displayed with the same precision as the raw PK concentration data
- Parameters associated with time (eg, time of maximum concentration [T_{max}]) will be displayed with 2 decimal places
- Percentages will be displayed with 2 decimal places
- Ratios will be displayed with 2 decimal places
- Coefficient of determination (R^2) and elimination rate constant (λ_z) will be displayed with 4 decimal places

Summary statistics for concentration and PK parameters will be displayed with the same precision as the individual values, with the exception of number of observations (n) and CV% which will be presented with 0 and 1 decimal place, respectively.

5.3 Baseline

Unless otherwise specified, the baseline value will be defined as the last non-missing evaluation prior to the first dose of study drug, including unscheduled assessment(s), if applicable.

5.4 Methods for Handling Missing Data

In general, no imputations of values for missing safety data (ie, blank, “Not Done”, “Not Applicable”, etc.) will be performed and data presentations will reflect the data point as it appears in the Case Report Form (CRF) or electronic data file.

6 STUDY SUBJECTS

Unless otherwise specified, all available data will be listed and summary table for disposition will be presented for all enrolled subjects.

6.1 Disposition

Subject disposition will be summarized by cohort (TMP-301 50 mg BID Fasted, TMP-301 50 mg QD Fasted, TMP-301 XX mg and TMP-301 XX mg), pooled placebo and overall, including:

- Number of subjects screened (if applicable)
- Number of subjects randomized (if applicable)
- Number of subjects who received study drug
- Number (%) of subjects who completed the study
- Number (%) of subjects discontinued from the study overall and by primary reason for discontinuation
- Number (%) of subjects included in each of the analysis populations

The percentages will be calculated using the number of subjects who received study drug.

Listings of subject's disposition, Randomization details, and subjects included in each of the analysis populations will be provided. In addition, a listing of screen failures including reason for failure will be presented.

6.2 Protocol Deviations

All protocol deviations related to study inclusion or exclusion criteria, conduct of the study, subject management, or subject assessment (including PK sample processing deviations) will be listed for the Randomized populations. Information for PK sampling time deviations will be derived programmatically, and presented in a listing.

Deviations will be collected in the clinic deviation tracking system (DTS) and presented in a general protocol deviation listing.

Table 3: Acceptable Windows for Timed PK Blood Specimen Collection Procedures

Elapsed Time	Accepted Window
Pre-dose	No window specified
> 0 hour to ≤ 30 minutes	± 10 minutes
> 30 minutes to ≤ 4 hours	± 10 minutes
> 4 hours to ≤ 12 hours	± 10 minutes
> 12 hours to ≤ 24 hours	± 10 minutes
> 24 hours to ≤ 72 hours	± 2 hours
> 72 hours	± 4 hours

7 DEMOGRAPHIC AND OTHER BASELINE CHARACTERISTICS

Unless otherwise specified, summary tables for demographics and other baseline characteristics, and all available data will be listed based on the Randomized population.

Demographic and baseline characteristics including age, sex, ethnicity, race, height, weight (at screening), and body mass index (BMI) will be listed and summarized by cohort (TMP-301 50 mg BID Fasted, TMP-301 50 mg QD Fasted, TMP-301 XX mg and TMP-301 XX mg), pooled placebo and overall. Descriptive statistics will be presented for age, height, weight, and BMI, while frequency counts and percentages will be presented for sex, ethnicity, and race.

The following will be presented in listings:

- Medical history
- Prior and concomitant medications
- Alcohol Habits
- Substance Use Habits
- Gynecological History
- Psychiatric History

8 SAFETY ANALYSIS

Unless otherwise specified, summary tables for safety assessments will be presented for the Safety population. All available data will be listed by cohort and subject based on the Randomized population.

8.1 Adverse Events

An AE is defined as any untoward medical occurrence in a clinical investigation subject administered an investigational product and which does not necessarily have a causal relationship with the treatment. An AE can therefore be any unfavorable and unintended sign (including a clinically significant abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of an investigational product, whether or not related to the investigational product.

Treatment-emergent AEs (TEAEs) are AEs not present prior to but reported following the exposure to study treatment or AEs already present that worsen in intensity or frequency following exposure to study treatment.

All TEAEs will be assigned to the last treatment taken by the subject where the date and time of the last treatment dosing is on or before of the start date and time of the event. Any TEAE started during follow-up period will be assigned to the last treatment that the subject has taken.

In case that the time of onset or time of resolution is unknown, treatment-emergent or assignment would be determined considering the worst case scenario and/or on case-by-case basis, as deemed appropriate. Treatment-emergent AEs with missing severity or relationship to study treatment will also be classified to the worst case (eg, severe and/or related, as appropriate) in corresponding summaries.

An overall summary of AEs will be presented by cohort (TMP-301 50 mg BID Fasted, TMP-301 50 mg QD Fasted, TMP-301 XX mg and TMP-301 XX mg), pooled placebo and overall active including:

- Number of AEs reported, overall only
- Number of TEAEs reported
- Number (%) of subjects with at least one TEAE
- Number (%) of subjects with at least one study drug-related TEAEs
- Number (%) of TEAEs by relationship to study treatment (ie, Reasonable Possibility or No Reasonable Possibility)
- Number (%) of TEAEs by severity
- Number of serious AEs (SAEs)
- Number (%) of subjects with at least one SAE
- Number (%) of subjects with at least one study drug-related SAE
- Number (%) of subjects with a TEAE leading to discontinuation
- Number (%) of subjects with outcome of death

Frequency tables will be presented by cohort, pooled placebo and overall:

- Number (%) of subjects with TEAEs by system organ class (SOC) and preferred term

(PT)

- Number (%) of subjects with drug-related TEAEs by SOC and PT
- Number (%) of subjects with TEAEs by relationship to study treatment
- Number (%) of subjects with TEAEs by severity
- Number (%) of subjects with SAEs by system organ class (SOC) and preferred term (PT)

All adverse event data will be presented in a listing.

8.2 Clinical Laboratory Evaluations

General biochemistry, hematology, and urinalysis assessments, and other laboratory tests are listed in Table 3.

Table 4: Clinical Laboratory Assessments

Clinical Laboratory Test Panel	Description
General biochemistry:	Alanine aminotransferase, albumin, alkaline phosphatase, aspartate aminotransferase, bilirubin total and conjugated bilirubin if total bilirubin is elevated, blood urea nitrogen, calcium, chloride, Creatinine phosphokinase (CPK), Bicarbonate (CO2), creatinine (including eGFR calculated using the CKD-EPI calculation, cholesterol total, glucose, LDH, phosphorous, potassium, protein total, sodium, triglycerides, and uric acid
Coagulation:	Prothrombin time INR and partial thromboplastin time (aPTT) levels
Endocrinology:	FSH[1]
Hematology:	White cell count with differential (absolute values of neutrophil, lymphocyte, monocyte, eosinophil, and basophil), red cell count, hemoglobin, hematocrit, mean corpuscular volume, and platelet count
Serology[1]	Human immunodeficiency virus (HIV) Ag/Ab Combo, Hepatitis B surface antigen, and Hepatitis C virus
Urinalysis:	Color, clarity, specific gravity, pH, leukocyte, protein, glucose, ketones, bilirubin, blood, nitrite, urobilinogen. Microscopic examination will only be performed if the dipstick test is outside of the reference range for leukocyte, blood, nitrite or protein
Urine drug screen:	Alcohol, amphetamines, barbiturates, benzodiazepines, cannabinoids, cocaine, cotinine, methadone, opiates, phencyclidine, and tricyclic antidepressants
Pregnancy test:	Serum pregnancy test or urine pregnancy test

[1]Screening visit only

Laboratory data will be summarized by the type of laboratory test, visit. Descriptive statistics (n, mean, SD, minimum, median, and maximum), and the number of subjects with laboratory test results below, within, and above normal ranges will be tabulated by laboratory test and visit.

All laboratory data will be listed by laboratory panel and test. Laboratory abnormalities and clinically significant abnormalities will also be listed.

8.2.1 Viral Screen

A screening viral screen will be done for HIV Ag/Ab Combo, Hepatitis B surface antigen, and Hepatitis C virus. The results of the viral screen will be listed.

8.2.2 Urine Drug Screen (UDS) and Urine Alcohol Testing

UDS will test for the following drugs of abuse: alcohol, amphetamines, barbiturates, benzodiazepines, cannabinoids, cocaine, cotinine, methadone, opiates, phencyclidine, and tricyclic antidepressants.

The results of UDS and urine alcohol testing will be listed.

8.2.3 Coagulation

A listing will be presented for coagulation data.

8.2.4 Pregnancy and Follicle Stimulating Hormone (FSH) Tests

A listing will be done for pregnancy and FSH tests. Serum pregnancy test will be done at screening. Urine pregnancy test will be done at check-in and discharge. For postmenopausal women, FSH test will be done at screening.

8.3 Vital Signs

Vital signs will include systolic and diastolic blood pressures, pulse rate, and oral body temperature.

Scheduled time points will include Days 1 and 7: Prior to morning dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, and 12.00 hours. Days 2, 4, 6, 8, 10, and 12: Prior to dosing and 4.00 and 12.00 hours postdose. Day 14: Prior to dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, 12.00, 24.00 (Day 15), 48.00 (Day 16), 72.00 (Day 17), 96.00 (Day 18) hours postdose.

At baseline on Day 1, predose vital sign measurements (except oral body temperature) will be conducted in triplicate (with an interval of approximately 10 - 30 minutes between each recording). The average value of each parameter will be considered as baseline value.

Vital signs will be summarized using descriptive statistics (n, mean, SD, minimum, median, and maximum). All vital signs, including oral temperature, will be listed. Clinically significant abnormal findings will be listed.

Summary tables on observed and change from baseline values will be presented by visit, time point, cohort and pooled placebo and overall.

8.4 Electrocardiograms

The 12-lead ECG data will include ventricular rate, PR, QRS, QT, and interpretation.

Scheduled time points will include Days 1, 7, and 14: Predose and at 4.00, 6.00, 8.00, and 12.00 hours postdose. Days 2, 4, 6, 8, 10, and 12: Predose and 4.00 and 12.00 hours postdose. Day 14:

Prior to dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, 12.00, 24.00 (Day 15), 48.00 (Day 16), 72.00 (Day 17), 96.00 (Day 18) postdose.

At baseline in Day 1 predose, ECG recordings will be conducted in triplicate at (-45, -30, and -15 minutes). The average value of each parameter will be considered as baseline value.

12-Lead ECG data absolute and change from baseline values in ventricular rate, PR, QRS, and QT intervals) will be summarized by parameter, visit, time point using descriptive statistics (n, mean, SD, minimum, median, and maximum). Clinically significant findings in ECG data will be listed.

8.5 Physical Examination Findings

Physical examination include a general review of the following body systems (at minimum): head and neck, cardiovascular, respiratory, gastrointestinal, brief neurological and general appearance, unless a symptom-oriented physical exam is indicated.

Physical examination findings will be presented in a listing by cohort, subject and visit and clinically significant physical examinations will be presented in a separate listing.

9 PSYCHIATRIC ASSESSMENTS AND ANALYSIS

Unless otherwise specified, all available psychiatric assessments and analysis will be presented for the Safety population. Listings will be based on the Randomized population.

Missing data for subjects who are administered all scheduled study treatments will be considered as random non-informative missing for analysis purposes. Missing values will be examined on a case-by-case basis to confirm this. Reasons for missing data will be displayed in the listings. No further imputation will be applied to any missing values.

Assessments done outside the acceptable visit windows may be excluded from by-time point summary statistics; this will be determined on a case-by-case basis prior to unblinding.

Analyses will be based on actual assessment times, except for predose (baseline) times, which will automatically be considered as time 0 as appropriate for analysis. If the actual time is missing, the nominal time will be used.

9.1 Brief Psychiatric Rating Scale (BPRS)

9.1.1 BPRS Assessment

The BPRS scale is designed for the assessment of psychiatric symptoms or disorders (e g., depression, anxiety, hallucinations, and unusual behavior).

The BPRS should be completed at Screening, Check-in on Day -2, 7 and 14. It will also be completed at approximately 6 and 24 h after dosing on Days 1, 7, and 14.

Baseline will be considered to be on Day -1.

Table 5: BPRS Details

Scale	Number of Items	Score Range Per Item	Total Possible Score	Scoring Interpretation	Data Collection
Brief Psychiatric Rating Scale (BPRS)	18 Items	1-7 point Likert scale 1=not present 2=very mild 3=mild 4=moderate 5=moderately severe 6=severe 7=extremely severe	126	Severity increases with increasing score A score of 18 is generally expected for healthy volunteers. Scores ≥ 25 reflect the presence of psychiatric symptoms and should be considered exclusionary Total Score cutoff criteria for patients diagnosed with schizophrenia or experiencing psychosis are: Mildly ill ≥ 31 Moderately ill ≥ 41 Markedly ill ≥ 53 In- or out patients with paranoid, disorganized or undifferentiated schizophrenia (DSM-IV), BPRS total score ≥36, BPRS psychotic subscore ≥23 and at least two BPRS psychosis items ≥4[1]	Score for each item (1-7) Score Range: 18-126 Cutoff Score: ≥25 If an item is not assessed, the BPRS Total Score will not be computed

9.1.2 BPRS Visit Windows

The acceptable windows for BPRS will be within 2 h following the 6 hours nominal time point and within 3 h prior to next dose for the 24 h time point. All the 24 h postdose assessments are intended to be completed prior to next day dosing as applicable.

Psychiatric assessments should be carried out in the following order when required: (1) 3MS, (2) CADSS, (3) BPRS, (4) C-SSRS.

9.1.3 BPRS Endpoints

Derived endpoints will include:

- $BPRS\_TS_{CFB}$ = Total Score Change from Baseline (CFB) = BPRS_TS at each postdose time point – $BPRS\_TS_{B}$ at baseline
- $BPRS\_TS_{\%CFB}$ = Total Score Percent Change from Baseline (CFB)

9.1.4 BPRS Analysis

Tables and listings will be presented for absolute values and derived endpoints described below. Inferential analysis will be provided for the derived endpoints (refer to SAP Section 9.6 for more detailed information).

In addition, a table and listing with Number of Subjects excluded at Screening (i.e., Number of subjects with BPRS_TS at Screening ≥ 25) will be presented.

9.2 Clinician Administered Dissociative States Scale (CADSS)

9.2.1 CADSS Assessment

The CADSS scale is designed for the assessment of dissociative states in adults.

The CADSS will be completed at Screening, Check-in on Day -2, and at approximately 6 and 24 h after dosing on Day 1. The CADSS will also be completed prior to dosing and at approximately 6 and 24 h after dosing on Days 7 and 14.

Baseline will be considered to be on Day -1.

Table 5: CADSS Details

Scale	Number of Items	Score Range Per Item	Total Possible Score	Scoring Interpretation	Data Collection
Clinician Administered Dissociative States Scale (CADSS)	23	0-4 point Likert scale 0=not at all 1=mild 2=moderate 3=severe 4=extreme	92	Severity increases with increasing score No official cut off criteria for scoring Baseline scores for normal healthy males were 1.67 (2.96)[2]	• Score for each item (0-4) Score Range: 0-92 (general dissociation) o Of note Items can be segregated into factors e.g. Altered visual and auditory perception (items 2, 9, 10, 11, 16, 18) o Altered time and perception (Items 1, 12, 15) Depersonalization (Items 3, 4, 5, 6, 20)[3]

9.2.2 CADSS Visit Windows

The acceptable window for CADSS will be within 2 hours following the 6 h nominal timepoint and within 3 h prior to next dose for the 24 h time point. All of the 24 h postdose assessments are intended to be completed prior to next day dosing as applicable.

Psychiatric assessments should be carried out in the following order when required: (1) 3MS, (2) CADSS, (3) BPRS, (4) C-SSRS.

9.2.3 CADSS Endpoints

Derived endpoints will include:

- $CADSS\_TS_{CFB}$ = Total Score Change from Baseline (CFB) = CADSS_TS at each postdose time point – $CADSS\_TS_B$ at baseline
- $CADSS\_TS_{\%CFB}$ = Total Score Percent Change from Baseline (CFB)

9.2.4 CADSS Analysis

Tables and listings will be presented for absolute values and derived endpoints described below. Inferential analysis will be provided for the derived endpoints (refer to SAP Section 9.6 for more detailed information).

9.3 Columbia-Suicide Severity Rating Scale (C-SSRS)

9.3.1 C-SSRS Assessment

The C-SSRS is a questionnaire designed for the assessment of suicidal ideation and behavior in adolescents and adults.

To monitor for a history of (the past 2 years to present) or for the emergence of suicidal ideation and behavior, subjects will undergo C-SSRS evaluations at Screening, Check-in on Day -2, and within 3 hours prior to dosing on Days 1, 3, 5, 7, 9, 11, 13, 15, 17 and on day 18 prior to Discharge, and at the follow-up/EOS visit.

The C-SSRS will be used to assess both behavior and ideation that tracks all suicidal events and provides a summary of suicidal ideation and behavior. It assesses the lethality of attempts and other features of ideation (frequency, duration, controllability, reasons for ideation, and deterrents), all of which are significantly predictive of completed suicide.

Two versions of the C-SSRS will be used in this study: the Baseline/Screening version (lifetime history and past 12 months) and the Since Last Visit version. The Screening version of the C-SSRS will be administered at the Screening visit. The Since Last Visit version of the C-SSRS will be administered at all subsequent assessment times.

9.3.2 C-SSRS Analysis

A frequency table will be used to summarize Baseline/Screening and Since Last Visit responses for each cohort, and C-SSRS Question, e.g. Wish to be Dead, Non-Specific Thought, Active Ideation (Not Plan) without Intent, Active Ideation with Some Intent, without Plan, Active Ideation with Plan and Intent.

A listing by Baseline/Screening and Since Last Visit responses will be output by cohort, subject, C-SSRS Category, and C-SSRS Question, e.g. Suicidal Ideation: Wish to be Dead, Non-Specific Thought, and Suicidal Behavior: Actual Attempt,, Engaged in Non-Suicidal Self-Injurious Behavior?, Interrupted Attempt, Aborted Attempt, Preparatory Acts or Behavior, Suicidal Behavior, Suicide.

9.4 Modified Mini-Mental State Examination (3MS)

9.4.1 3MS Assessment

The 3MS is an examination designed for the assessment of dementia in adults.

The 3MS will be completed at Screening, Check-in on Day -1, and at approximately 24 h after dosing on Day 1. The 3MS will also be completed prior to dosing and at approximately 24 h after dosing on Days 7 and 14.

Baseline will be considered to be on Day -1.

Table 6: 3MS Details

Scale	Number of Items	Score Range Per Item	Total Possible Score	Scoring Interpretation	Data Collection
Modified Mini-Mental State (3MS)	15 items with subscores	Variable Likert scoring for each item	100	Better performance correlated with increasing score. Scores ≤ 77 may be indicative of dementia Normal cognition = 78–100 Moderate cognitive impairment ≤ 77 Severe cognitive impairment ≤ 49	• Score for each item (0-7) • Total score/100

Scoring calculation:

1. When and where born?

1a. Date Day (0 or 1)

Month (0 or 1)

Year (0 or 1)

1a. Score = 0 to 3

1b. Place Town (0 or 1)

Province/Country (0 or 1)

1b. Score = 0 to 2

1 Score = Sum of 1a + 1b (ie, 0 to 5)

2. Three Words (Number of presentations ___)

2a. Shoes (0 or 1)

2b. Blue (0 or 1)

2c. Modesty (0 or 1)

2 Score = Sum of 2a + 2b + 2c (ie, 0 to 3)

3. 3a. Counting Forwards (0 or 1)

3b. Counting Backwards (0 to 2)

3c. Spell "World" (0 or 1)

3 Score = Sum of 3a + 3b + 3c (ie, 0 to 5)

4. First Recall

4a. Shoes (0 or 3)

4b. Blue (0 or 3)

4c. Modesty (0 or 3)

4 Score = Sum of 4a + 4b + 4c (ie, 0 to 9)

5. Today's Date

5a. Today's date (0 or 3)

5b. Year (0 or 3)

5c. Season (0 or 3)

5d. Month (0 or 3)

5e. Day of week (0 or 3)

5 Score = Sum of 5a + 5b + 5c + 5d + 5e (ie, 0 to 15)

6. Spatial Orientation

6a. Province (0 or 1)

6b. Country (0 or 1)

6c. City (0 or 1)

6d. Place (0 or 1)

6e. Street (0 or 1)

6 Score = Sum of 6a + 6b + 6c + 6d + 6e (ie, 0 to 5)

7. Naming

7a. (0 or 1)

7b. (0 or 1)

7c. (0 or 1)

7d. (0 or 1)

7e. (0 or 1)

7 Score = Sum of 7a + 7b + 7c + 7d + 7e (ie, 0 to 5)

8. Four legged animals Score 0 to 10

9. Similarities

9a. (0 to 2)

9b.(0 to 2)

9c. (0 to 2)

9 Score = Sum of 9a + 9b + 9c (ie, 0 to 6)

10. Repetition

10a. Province (0 to 2)

10b. Country (0 or 1)

10c. City (0 or 1)

10d. Place (0 or 1)

10 Score = Sum of 10a + 10b + 10c + 10d (ie, 0 to 5)

11. Obeys commands Score 0 to 3

12. Writing Score 0 to 5

13. Copy

13a. (0 to 4)

13b. (0 to 4)

13c. (0 to 2)

13 Score = Sum of 13a + 13b + 13c (ie, 0 to 10)

14. 3-Stage command

14a. (0 or 1)

14b. (0 or 1)

14c. (0 or 1)

14 Score = Sum of 14a + 14b + 14c (ie, 0 to 3)

15. Second Recall

15a.	Shoes	(0 or 3)
15b.	Blue	(0 or 3)
15c.	Modesty	(0 or 3)

15 Score = Sum of 15a + 15b + 15c (ie, 0 to 9)

Total 3MS Score = Sum of Scores for Items 1 to 15 = 0-100

9.4.2 3MS Visit Windows

The acceptable window for 3MS will be within 3 hours prior to next dose for the 24 hours time point. All the 24 hours postdose assessments are intended to be completed prior to next day dosing as applicable.

PD procedures should be carried out in the following order when required: (1) 3MS, (2) CADSS, (3) BPRS, (4) C-SSRS.

9.4.3 3MS Endpoints

Derived endpoints will include:

- $3MS\_TS_{CFB}$ = Total Score Change from Baseline (CFB) = CADSS_TS at each postdose time point – $3MS\_TS_B$ at baseline
- $3MS\_TS_{\%CFB}$ = Total Score Percent Change from Baseline (CFB)

9.4.4 3MS Analysis

Tables and listings will be presented for absolute values and derived endpoints described below. Inferential analysis will be provided for the derived endpoints (refer to SAP Section 9.6 for more detailed information).

9.5 Alertness Visual Analogue Scale (VAS)

9.5.1 Alertness VAS Assessment

The VAS alertness scale is designed to assess the alertness of subjects across a continuum scale with values from 0 to 10.

The VAS should be completed prior to TMP-301 administration at Screening, and at approximately 4, 6 and 12 h after dosing on Days 1, 7, and 14. This assessment will also occur on Day 18 prior to Discharge.

9.5.2 Alertness VAS Endpoints

Derived endpoints will include:

- E_{max}(Maximum postdose effect)
- E_{min}(Minimum postdose effect)
- TE_{max}(Time of postdose maximum effect)
- TE_{min}(Time of postdose minimum effect)

E_{max} and E_{min} will be calculated based on the postdose values; the predose value will be excluded.

TE_{max} and TE_{min} will be calculated as the time from time 0 to E_{max} and E_{min}, respectively, as appropriate. The first time of peak effect is used if the value is reported at more than one time point.

9.5.3 Alertness VAS Analysis

Alertness VAS values will be summarized and listed for absolute values and derived endpoints. No inferential analyses will be performed.

9.6 Psychiatric Assessments Analysis

Psychiatric measures at each time point will be summarized by treatment (placebo and each dose of TMP-301) using descriptive statistics and presented graphically. Derived endpoints will also be summarized by treatment using descriptive statistics. Only summary statistics will be output for E_{max} $TE_{max,}$ E_{min} and TE_{min}. Descriptive statistics will include n, mean, standard error (SE), minimum, first quartile (Ql), median, third quartile (Q3) and maximum for all psychiatric assessment values, and endpoints other than TE_{max} and TE_{min}. For TE_{max} and TE_{min}, minimum, Q1, median, Q3 and maximum will be output.

A mixed-effects model for a parallel study design will be used to compare the derived endpoints between treatments (e.g., BPRS_TS_{CFB}, BPRS_$TS_{\%CFB}$, CADSS_TS_{CFB}, CADSS_$TS_{\%CFB}$, 3MS_TS_{CFB}, and 3MS_$TS_{\%CFB}$). The model will include treatment (dosing group), day, and dose (1^{st} or 2^{nd}) as fixed effects, and the baseline measurement as a covariate. P-values for key components of variance, least-square means (LSMeans), and 95% CI for contrasts will be output. For each endpoint, the following comparisons will be made:

- Contrast 1: TMP-301 50 mg BID vs. Pooled Placebo
- Contrast 2: TMP-301 50 mg QD vs. TMP-301 50 mg BID
- Contrast 3: TMP-301 50 mg QDvs. Pooled Placebo
- Contrast 4: TMP-301 XX mg vs. TMP-301 50 mg BID
- Contrast 5: TMP-301 XX mg vs. TMP-301 50 mg QD
- Contrast 6: TMP-301 XX mg vs. Pooled Placebo

10 PHARMACOKINETIC ANALYSIS

The PK analysis will be carried out according to Altasciences SOPs.

10.1 Missing Values

The lack of concentration values due to failure to collect the sample, a lost or compromised sample or due to the subject's early termination from the study will be termed "missing" in the dataset, and no imputation will be done.

If the actual collection time of a postdose PK sample is unknown, but a valid concentration value has been measured, the sample will be set to missing in the PK analysis and will be presented in listing excluded from descriptive statistics.

10.2 Measurements Below the Lower Limit of Quantitation

Concentration values below the lower limit of quantitation (LLOQ) associated with predose and postdose collection times will be replaced with zero for the non-compartmental analysis (NCA).

Concentration values below the LLOQ will be replaced with zero for mean PK profile representations as well as for descriptive statistic calculations.

Concentration values below the LLOQ that are embedded between two quantifiable concentrations will be replaced with missing for mean PK profile representations as well as for descriptive statistic calculations.

10.3 Actual Time

The NCA will be based on actual sampling times.

The individual concentration-time profiles will be presented using actual sampling times whereas the mean concentration-time profiles and tables presenting summary statistics of concentration-time series will be presented using nominal sampling times.

10.4 Non-Compartmental Analysis

The following configuration for the NCA analysis (with Phoenix® WinNonlin® version 8.0, or higher) will be used:

- Data: Serial sampled data
- Model/Dose options Type: Plasma (200 -202) / Extravascular
- AUC Calculation Method: Linear Up Log Down
- Lambda $_z$ (λz) calculation: Best fit method for λz Linear-Log regression

Reasons for excluding PK parameters will include the following:

- AUC: AUC parameters will not be estimated if less than 3 consecutive measurable concentrations are observed
- PK parameters requiring λz estimation (eg, Vz/F) will be set to Not Reported (NR) in the tables and listings if they meet one of the following:
  - $R^2 < 0.8$

The PK parameters for TMP-301 are presented in Table 6.

Table 7: Pharmacokinetic Parameters of TMP-301

Parameter	Definition
PK Parameters Day 1	
C_{max}	Maximum observed concentration occurring at time T_{max} after the first dose
T_{max}	Time of maximum observed concentration after the first dose
$AUC_{0-12,}$ (For BID dosing)	Area under the concentration time curve over the dosing interval, calculated from 0 to 12 hours (dosing interval) after the first dose *(The **nominal time** will be used to estimate AUC using the NCA built-in tool in Phoenix® WinNonlin®. This means that actual times off by more than 1 minute from tau will be extrapolated/intrapolated as per Phoenix® WinNonlin®'s built-in formulas. If extrapolation/intrapolation is not possible, then no value is reported by the software.)*
$AUC_{0-24,}$ (For QD dosing)	Area under the concentration time curve over the dosing interval, calculated from 0 to 24 hours (dosing interval) after the first dose *(The **nominal time** will be used to estimate AUC using the NCA built-in tool in Phoenix® WinNonlin®. This means that actual times off by more than 1 minute from tau will be extrapolated/intrapolated as per Phoenix® WinNonlin®'s built-in formulas. If extrapolation/intrapolation is not possible, then no value is reported by the software.)*
C_{12}	Observed concentration occurring at time 12 h
C_{24}	Observed concentration occurring at time 24 h ($C_{24\ Day\ 1} = C_{trough\ Day\ 2}$)
PK Parameters Days 2, 5, 7, 9, 11 and 12	
C_{trough}	Observed concentration at the end of the dosing interval
PK Parameters Day 14	
$C_{max,\ ss}$	Maximum observed concentration at steady -state occurring at time $T_{max,\ ss}$
C_{avg}	Average concentration, calculated as $AUC_{0-Tau,\ ss}$/Tau
$T_{max,\ ss}$	Time of maximum observed concentration at steady-state
$C_{min,\ ss}$	Minimum observed concentration after the first dose of a multiple regimen
$AUC_{0-TAU,\ ss}$	Area under the concentration time curve over the dosing interval at steady-state. *(The **nominal time** will be used to estimate AUC using the NCA built-in tool in Phoenix® WinNonlin®. This means that actual times off by more than 1 minute from tau will be extrapolated/intrapolated as per Phoenix® WinNonlin®'s built-in formulas. If extrapolation/intrapolation is not possible, then no value is reported by the software.)* For QD dosing: $AUC_{0-TAU,SS} = AUC_{0-24,SS}$ For BID dosing $AUC_{0-TAU,SS} = AUC_{0-12,SS}$
$C_{12,\ ss}$	Observed concentration occurring at time 12 h

Parameter	Definition
$C_{24, ss}$	Observed concentration occurring at time 24 h
CL/F	Apparent total clearance at steady-state, calculated as Dose/ $AUC_{0\text{-}TAU,\ ss}$
V_z/F	Apparent volume of distribution at steady-state, calculated as Dose/λz * $AUC_{0\text{-}TAU, SS}$
AR_C_{max}	Accumulation ratio derived as $C_{max, ss}$ / $C_{max,FD\ Day\ 1}$
AR_$AUC_{0\text{-}12\ BID}$	Accumulation ratio derived as $AUC_{0\text{-}12,ss}$ / $AUC_{0\text{-}12,Day\ 1}$
AR_$AUC_{0\text{-}24\ QD}$	Accumulation ratio derived as $AUC_{0\text{-}24,ss}$ / $AUC_{0\text{-}24,\ Day\ 1}$
AR_C_{12}	Accumulation ratio derived as $C_{12,ss}/C_{12,Day\ 1}$
AR_C_{24}	Accumulation ratio derived as $C_{24,ss}/C_{24,Day\ 1}$ ($C_{24\ Day\ 1} = C_{trough\ Day\ 2}$)
CYP1A2_ratio	Ratio of CYP1A2 activity derived as paraxanthine/caffeine concentration day 14 / paraxanthine/caffeine concentration day -1
The following PK parameters will be used for PK calculation on Day 14 and presented in the PK listings only	
$\lambda_{z\ Upper}$	Upper limit on time for values included in the calculation of λ_z
$\lambda_{z\ Lower}$	Lower limit on time for values included in the calculation of λ_z
λ_z	Apparent elimination rate constant, estimated by linear regression of the terminal linear portion of the log concentration *versus* time curve
R^2	Goodness of fit for the terminal phase
Number of Points	Number of data points in computing λ_Z

10.5 Visit Windows

Windows for timed PK blood sample collections are presented in Table 7. PK samples collected outside of the pre-specified windows will be documented as protocol deviations. Since actual times are to be used for the PK analysis, deviations will be reflected in the analysis unless indicated otherwise upon review of the data.

Table 8: Acceptable PK Visit Windows

Elapsed Time	Accepted Window
Predose	No window specified
> 0 hour to ≤ 30 minutes	± 1 minute
> 30 minutes to ≤ 4 hours	± 2 minutes
> 4 hours to ≤ 12 hours	± 5 minutes
> 12 hours to ≤ 24 hours	± 10 minutes
> 24 hours to ≤ 72 hours	± 2 hours
> 72 hours	± 4 hours

10.6 Pharmacokinetic Statistical Methodology

All tables, figures, and listings (TFLs), when appropriate, will be stratified by treatment.

10.6.1 Summary Statistics

Summary statistics of the individual concentration data and derived parameters will be calculated with Phoenix® WinNonlin® for the PK population. Summary statistics will be calculated for concentration at each individual time point and for all PK parameters.

C_{trough} will be displayed graphically and summarized descriptively by day to assess for steady state.

Appropriate dose–normalized PK parameters ($C_{max, ss}$, $C_{12, ss}$, $C_{24, ss}$, and $AUC_{0\text{-}TAU, ss}$, when appropriate) will be assessed graphically for dose-proportionality.Concentration data will be summarized by group using the following statistics: number of observations (n), arithmetic mean (mean), SD, min, median, max, and CV%. The PK parameters will be summarized using these same statistics, as well as geometric mean and geometric mean CV%.

No comparison between fed and fast state will be provided.

10.6.2 Dose Proportionality

Natural log-transformed PK parameters ($C_{max, ss}$, $C_{12, ss}$, $C_{24, ss}$, and $AUC_{0\text{-}TAU, ss}$) will be assessed statistically for proportionality. Proportionality analysis will be done using a power model. The power model is defined as:

$$\ln(\text{PK parameter}) = \alpha + \beta \bullet \ln(\text{Dose}) + \varepsilon$$

where α is the intercept, β is the slope and ε is the error term. A linear model with ln-transformed dose as a continuous effect will be fitted. A point estimate and a 90% confidence interval will be derived for the slope (β).

The parameter can be considered to be dose-proportional if the 90% CIs for the slope coefficient that include 1 will suggest evidence of dose proportionality.

Dose proportionality may be assessed within different dose ranges if deemed appropriate with at least three doses.

Otherwise, if there are only two cohorts completed, Statistical inference will be based on a bioequivalence approach for a parallel group design and expressing the relative difference as a geometric mean ratio and 90% CI using ANOVA model based on dose normalized PK parameters.

No comparison between fed and fast state will be provided.

11 INTERIM ANALYSES AND DATA SAFETY MONITORING

No formal interim analyses will be performed; blinded safety data will be reviewed by an Investigator and the Sponsor's Medical Monitor following completion of each TMP-301 dose level.

12 CHANGES TO PROTOCOL-SPECIFIED ANALYSES

Added below analysis population definitions:

- **Enrolled Population**

The Enrolled population will include all subjects who signed informed consent.

- **Screened Population**

The Screened population will include all subjects who meet the eligibility criteria.

- **Randomized Population**

The Randomized population will include all subjects who are assigned a randomization number.

13 GENERAL INFORMATION RELATED TO DATA PRESENTATIONS

The formats and layouts of TFLs are provided in a separate document as common displays. Their numbering and general content follow the International Conference on Harmonisation (ICH) E3 guidelines. Actual formats and layouts may be altered slightly from those presented as necessary to accommodate actual data or statistics.

14 REFERENCES

1. Peuskens, J., Bech, P., Möller, H.J., Bale, R., Fleurot, O., Rein, W. (1999). Amisulpride vs. risperidone in the treatment of acute exacerbations of schizophrenia. Amisulpride study group. Psychiatry Res, 88(2), 107-17.
2. Piazza, G.G., Iskandar, G., Hennessy, V. et al (2022). Pharmacological modelling of dissociation and psychosis: an evaluation of the Clinician Administered Dissociative States Scale and Psychotomimetic States Inventory during nitrous oxide ('laughing gas')-induced anomalous states. Psychopharmacology, 239, 2317–2329.
3. van Schalkwyk, G. I., Wilkinson, S. T., Davidson, L., Silverman, W. K., Sanacora, G. (2018). Acute psychoactive effects of intravenous ketamine during treatment of mood disorders: analysis of the Clinician Administered Dissociative State Scale. Journal of affective disorders, 227, 11-16.

APPENDIX A STUDY SCHEDULE(S)

Study Procedures	Screening	Check-in	Baseline Caffeine test	Day 1	Days 2-3	Day 4	Days 5-6	Day 7	Days 8-13	Day 14	Days15-17	Discharge	Follow-up/EOS visit
Day	Days-28 to -1	Day -2	Day -1									Day 18	Day 25[z] (±2 days)
Informed Consent	X												
Inclusion/Exclusion Criteria	X	X											
Admission to Unit		X											
Demographic Data	X												
Medical History	X	X[a]											
Psychiatric History	X												
Physical Examination	X	X[b]										X[p]	
Psychiatric Examinations[c]	X[c]												
Urinary Drug Screen	X	X											
Alcohol Urine Test	X	X											
Serology	X												
Pregnancy Test[d]	X	X										X	
FSH[e]	X												
Height and body weight	X[f]											X	X
Genotyping sample[g]				**X**									
Exploratory Biomarkers sample				X[h]	X[h]			X[h]		X[h]		X	
Check-in		X											
Check-out												X	
Nonresidential visit	X												X

Study Procedures	Screening	Check-in	Baseline Caffeine test	Day 1	Days 2-3	Day 4	Days 5-6	Day 7	Days 8-13	Day 14	Days15-17	Discharge	Follow-up/EOS visit
Randomization		X											
TMP-301 or Placebo[i]				X	X	X	X	X	X	X			
Caffeine 100 mg oral			X							X			
Caffeine Plasma sampling			4 hr post dose							4 hr post dose			
Plasma PK Sampling[j,k,l]				X	X	X	X	X	X	X	X	96.00[aa]	X
Urine Sampling[l]				X						X	X		
Adverse Event Recording	X	X		X	X	X	X	X	X	X	X	X	X
Prior/Concomitant Medication Monitoring	X	X		X	X	X	X	X	X	X	X	X	X
Clinical chemistry, Hematology, and Urinalysis[m]	X	X		X	X	X	X		X	X	X	96.00 [aa]	X
Vital Signs: Blood Pressure, Pulse Rate and Oral Body Temperature[n,o]	X	X		X	X	X	X	X	X	X	X	96.00 [aa]	X
12-lead ECG[q,r]	X			X	X	X	X	X	X	X	X	96.00 [aa]	X
VAS Alertness Scale[s,t]				X				X		X		96.00 [aa]	
BPRS, CADSS[t,u,v,x]	X	X		X				X		X			
3MS[t,v,w,x]	X	X		X				X		X			
C-SSRS[y]	X	X		X	X		X	X	X		X	X	X
Discharge												X	

Abbreviations: BPRS = Brief Psychiatric Rating Scale; CADSS = Clinician-Administered Dissociative States Scale; 3MS = Modified Mini-Mental State; C-SSRS = Columbia-Suicide Severity Rating Scale**;** ECG = electrocardiogram; FSH = follicle-stimulating hormone; PK = pharmacokinetic(s).

[a] Interim medical history.
[b] Symptom directed physical examination and at Investigator discretion.

[c] Clinical Interview for DSM-5 (SCID-5 CT) will be conducted by appropriately trained staff. This psychiatric examination will include an assessment of premorbid personality, personal and developmental history, alcohol and substance use history, forensic history, past psychiatric history. The PRIME questionnaire will also be administered to screen for risk of psychosis. A psychiatric exam (full or partial) can take place at any time during the study at the discretion of the PI, if there is a relevant concern.

[d] In all females. Serum pregnancy test at Screening and urine pregnancy tests at all other times. A positive urine pregnancy test will be confirmed with a serum pregnancy test.

[e] In all females to determine postmenopausal status.

[f] Height measured at Screening only.

[g] The genotyping samples will be stored and may be processed and analyzed to assess genotyping of drug metabolizing systems.

[h] Day 1 at pre-dose, 1, 4, and 8 hours; day 2 pre-dose; and days 7 and 14 at pre-dose, 1, 4, and 8 hours.

[i] For BID Cohorts: Dosing twice daily interval from Day 1 to Day 13, two intakes separated by 12.00 hours (note: only the AM dose is administered on Day 14). For QD Cohorts: Dosing once daily in the morning from Day 1 to Day 14.

[j] Timing of PK blood samples may be changed based on emerging data.

[k] At each protocol specified timepoint for PK (including predose baseline) out to 12.00 hours postdose. (Section 6.2).

Serial blood will be collected on Day 1, and 14 at predose, 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, and 12.00 hours. Trough (predose AM) and 8 hr and 12 hr samples will be collected on Days 2, 7 and 11. Trough samples (predose AM) will be collected on Days 5, 9 and 12. Additional PK samples will be drawn after the last dose on Day 14 at 24.00 (Day 15), 28.00 (Day 15), 48.00 (Day 16), 52.00 (Day 16), 72.00 (Day 17), and 96.00 (Day 18) hours postdose and Day 25 at the follow-up EOS visit (Day25 ± 2 days). Urine will be collected on Day 1 (0 to 4, 4 to 8, 8 to 12, 12 – 24 hr) and Day 14 (0 to 4, 4 to 8, 8 to 12, 12 to 24, 24 to 48 (Day 15), and 48 – 72 hr (Day 16)). See Table 4.

[m] Day 1 predose, then 24.00 (Day 2) and 72.00 hours (Day 4) postdose, then every 2 days (Days 6, 8, 10, 12, 14, 16) post dose at approximately 10 am ± 1 hour.

[n] Days 1 and 7: Prior to morning dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, and 12.00 hours. Days 2, 4, 6, 8, 10, and 12: Prior to dosing and 4.00 and 12.00 hours postdose. Day 14: Prior to dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, 12.00, 24.00 (Day 15), 48.00 (Day 16), 72.00 (Day 17), 96.00 (Day 18) hours postdose.

[o] At baseline on Day 1, predose vital sign measurements (except oral body temperature) will be conducted in triplicate (with an interval of approximately 10 - 30 minutes between each recording). The average value of each parameter will be considered as baseline value.

[p] Symptom-directed physical examination.

[q] Days 1 and 7: Predose and at 4.00, 6.00, 8.00, and 12.00 hours postdose. Days 2, 4, 6, 8, 10, and 12: Predose and 4.00 and 12.00 hours postdose. Day 14: Prior to dosing and 0.5, 1.00, 2.00, 3.00, 4.00, 6.00, 8.00, 12.00, 24.00 (Day 15), 48.00 (Day 16), 72.00 (Day 17), 96.00 (Day 18) postdose.

[r] At baseline in Day 1 predose, ECG recordings will be conducted in triplicate at (-45, -30, and -15 minutes). The average value of each parameter will be considered as baseline value.

[s] Day 1, 7, and 14: Predose then 4.00, 6.00, and 12.00 hours postdose and Day 18.

[t] May also be conducted at other times if deemed appropriate based on emerging data.

[u] Check-in, then 6.00 and 24.00 hours postdose on Day 1, Day 7, and Day 14.

[v] Conducted as part of the psychiatric examination.

[w] Check-in, and 24.00 hours postdose on Day 1, Day 7, and Day 14.

[x] Subjects will be screened within 28 days prior to dosing. A psychiatric examination will be performed at Screening by a suitably trained psychiatrist or appropriately trained staff using a clinical interview. This will include an assessment of premorbid personality, personal and developmental history, alcohol and substance use history, forensic history, past psychiatric history, and mental state examination.

[y] Every other day from Day 1 to discharge.

[z] Approximately 1 week after last dose.

[aa] 96 hours after the last (Day 14) dose