Clinical Trial Protocol

	Document Number:	**c30179584-08**
EudraCT No.	2019-004836-51	
BI Trial No.	1447-0001	
BI Investigational Medicinal Product	BI 1569912	
Title	Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)	
Lay Title	A study in healthy men to test how well different doses of BI 1569912 are tolerated and how food influences the amount of BI 1569912 in the blood.	
Clinical Phase	I	
Clinical Trial Leader	[redacted] Phone: [redacted] Fax: [redacted]	
Principal Investigator	[redacted] Phone: [redacted] Fax: [redacted]	
Status	Final Protocol (Revised Protocol (based on global amendment 7))	
Version and Date	Version: 8.0	Date: 14 May 2021

CLINICAL TRIAL PROTOCOL SYNOPSIS

Company name	Boehringer Ingelheim
Protocol date	06 April 2020
Revision date	14 May 2021
BI trial number	1447-0001
Title of trial	Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
Principal investigator	[redacted]
Trial site	
Clinical phase	I
Trial rationale	As a transition from preclinical investigations to clinical development, safety, tolerability, pharmacokinetics and pharmacodynamics of BI 1569912 will be assessed in healthy male subjects using single rising oral doses in order to provide a basis for the further clinical development in the indication of major depressive disorder (MDD)
Trial objectives	SRD-Part: To investigate safety, tolerability, pharmacokinetics and pharmacodynamics following single rising doses of BI 1569912 BA/ FE-Part: To investigate (a) the relative bioavailability of BI 1569912 PfoS and tablet and (b) the influence of food on the relative bioavailability of the BI 1569912 tablet
Trial endpoints	Primary endpoint To assess SRD-Part: Safety and tolerability of BI 1569912 as the percentage of subjects with drug-related adverse events BA/ FE-Part: AUC_{0-tz} and C_{max} of BI 1569912 Secondary endpoints To assess SRD-Part: $AUC_{0-\infty}$ and C_{max} of BI 1569912 BA/ FE-Part: $AUC_{0-\infty}$ of BI 1569912
Trial design	SRD-Part: Single-blind, partially randomised within dose group, placebo-controlled, parallel-group design BA/ FE-Part: Open-label, randomized, single-dose, intra-individual, six-sequence, three-way crossover design

Number of subjects	84
total entered	SRD-Part: 72 healthy subjects* The addition of further dose groups exceeding the already tested dose levels for the evaluation of safety findings will be subject to a substantial amendment requiring approval. BA/ FE-Part: 12 healthy subjects
each treatment	SRD-Part: 8 per dose group (6 on active drug and 2 on placebo) BA /FE-Part: 4 per cohort * Additional subjects may be entered to allow testing of additional doses on the basis of experience gained during the trial conduct (e.g., preliminary pharmacokinetic data), under the condition that the planned and approved highest dose will not be exceeded. Thus, the actual number of subjects entered may exceed 72, but is not to exceed 88.
Diagnosis	Not applicable
Main criteria for inclusion	SRD- & BA/ FE-Part: Healthy male subjects, age of 18 to 45 years (inclusive), body mass index (BMI) of 18.5 to 29.9 kg/m2 (inclusive)
Test product (SRD-Part)	BI 1569912 PfoS of 0.625 mg/ mL strength
dose	0.25 mg, 0.75 mg, 2 mg, 5 mg, 10 mg, 20 mg, 30 mg, 40 mg, 50 mg QD
mode of admin.	Oral with 240 mL of water after an overnight fast of at least 10 h in the morning.
Test product (BA/ FE-Part)	BI 1569912 tablets of 5 mg or 2.5 mg or 0.5 mg strength
dose	Either 5 mg or 2.5 mg or 0.5 mg QD. The highest dose strength will be tested, as demonstrated in the SRD-Part to be safe and tolerable, reduced by a factor of at least 2.
mode of admin.	Oral with 240 mL of water after an ***overnight fast*** of at least 10 h in the morning.
Test product (BA/ FE-Part)	BI 1569912 tablets of 5 mg or 2.5 mg or 0.5 mg strength
dose	Either 5 mg or 2.5 mg or 0.5 mg QD. The highest dose strength will be tested, as demonstrated in the SRD-Part to be safe and tolerable, reduced by a factor of at least 2.
mode of admin.	Oral with 240 mL of water ***after a high-fat, high-calorie breakfast*** in the morning.
Test product (BA/ FE-Part)	BI 1569912 PfoS of 0.625mg/ mL strength
dose	Either 5 mg or 2.5 mg or 0.5 mg QD. The highest dose strength will be tested, as demonstrated in the SRD-Part to be safe and tolerable, reduced by a factor of at least 2.
mode of admin.	Oral with 240 mL of water after an ***overnight fast*** of at least 10 h in the morning.

Comparator product (SRD-Part)	Matching placebos: PfoS
dose	Not applicable
mode of admin.	Oral with 240 mL of water after an ***overnight fast*** of at least 10 h in the morning.
Duration of treatment	BI 1569912: SRD-Part: Single dose BA/ FE-Part: Three single doses
Statistical methods	SRD-Part: Descriptive statistics will be calculated for all endpoints. Dose proportionality of BI 1569912 will be explored using a regression model. A 95% confidence interval (CI) for the slope will be computed. Linearity index will be estimated using a linear model providing a two-sided 95% CI. BA/ FE-Part: Relative bioavailability and food effect will be estimated by the ratios of the geometric means (test/reference) for the primary and secondary endpoints. Additionally, their two-sided 90% confidence intervals (CIs) will be provided. This method corresponds to the two one-sided t-test procedure, each at a 5% significance level. Since the main focus is on estimation and not testing, a formal hypothesis test and associated acceptance range is not specified. The statistical model will be an analysis of variance (ANOVA) on the logarithmic scale including effects for sequence, subjects nested within sequences, period and treatment. CIs will be calculated based on the residual error from the ANOVA.

FLOW CHART – SINGLE RISING DOSE PART

Visit	Day	Planned time (relative to drug administration [h:min]	Approximate clock time of actual day [h:min]	Event and comment	Safety laboratory	PK blood [10, 11]	PK urine [10,12]	EEG/ qEEG	Physical /Neurological Examination	C-SSRS	CADSS	ERP	[redacted]	12-lead ECG	Continuous ECG monitoring	Vital signs (BP, PR, RR, T)	Questioning for AEs and concomitant therapy [6]
1	-21 to -4	-504:00	08:00	Screening (SCR)[1]	A			x[17,9]	x	x	x			x		x	
	-3	-72:00	08:00		B												
2	-2	-38:00	18:00	***Admission to trial site***	x[5]												
		-35:30	20:30	Snack (voluntary)													
	-1	-25:00	07:00											x[13]		x	x
		-24:00	08:00						x		x[18]	x					
		-23:00	09:00					x									
		-21:00	11:00					x									
		-20:00	12:00									x					
		-16:00	16:00					x									
	1	-1:00	07:00	Allocation to treatment[2,19]		x[2, 14]	x[2]										
		-0:45	07:15						x						x[2]		
		0:00	**08:00**	**Drug administration**			▲								▲		x
		0:05	08:05			x	|							|		▲[16]
		0:15	08:15			x	|							|	x	|
		0:30	08:30			x	|						x[20]	|	x	|
		0:45	08:45			x	|	x						|	x	|
		1:00	09:00			x	|			x				|	x	|
		1:10	09:10				|						x[20]	|		|
		1:15	09:15				|	x						|		|
		1:30	09:30			x	|						x[20]	|	x	|
		2:00	10:00	240 mL fluid intake			|				x			x[20]	|	x	|
		2:30	10:30			x	|							|		|

Visit	Day	Planned time (relative to drug administration [h:min]	Approximate clock time of actual day [h:min]	Event and comment	Safety laboratory	PK blood [10, 11]	PK urine [10,12]	EEG/ qEEG	Physical /Neurological Examination	C-SSRS	CADSS	ERP	[redacted]	12-lead ECG	Continuous ECG monitoring	Vital signs (BP, PR, RR, T)	Questioning for AEs and concomitant therapy [6]
		3:00	11:00			x[8]	┃	x						x[20]	┃	x	┃
		4:00	12:00	240 mL fluid intake, thereafter lunch[3]	B	x	┼		x		x[18]	x	▮	x[20]	▼	x	┃
		6:00	14:00			x	┃							x[20]		x	┃
		8:00	16:00	Snack (voluntary)[3]		x	┼	x						x[20]		x	┃
		10:00	18:00	Dinner[3]			┃										┃
		12:00	20:00			x	┼							x[20]		x	┃
	2	24:00	08:00		B	x	┼	x	x		x[18]	x	▮	x[20]		x	┃
		34:00	18:00			x	┃							x[20]		x	┃
	3	48:00	08:00			x	▼	x			x[18]	x	▮	x[20]		x	┃
	4	72:00	8:00	Breakfast (voluntary)[3] ***Discharge from trial site***	B[15]	x		x	x	x	x[18]	x	▮	x[20]		x	▼
5	4 to 14			End of trial (EoT) examination[4]	A				x					x		x	x[16]

1. Subject must be informed and written informed consent obtained prior to starting any screening procedures. Screening procedures include physical examination, check of demographics (including determination of body height and weight, smoking status and alcohol history), relevant medical history, concomitant therapy and review of inclusion/ exclusion criteria.
2. The time is approximate; procedures are to be performed and completed within the 3 h prior to drug administration. Allocation to treatment may be performed at any time following enrolment but must be completed prior to (first) drug administration.
3. If several actions are indicated at the same time, the intake of meals will be the last.
4. EoT examination includes physical examination, body weight, vital signs, ECG, safety laboratory, recording of AEs, and concomitant therapies.
5. Only urine drug screening and alcohol breath test will be done at this time.
6. AEs and concomitant therapies will be recorded throughout the trial, but will be specifically asked for at the times indicated in the Flow Chart above.
8. One additional blood sample for stability testing will be taken at this time (refer to Section 5.3.2.4).
9. qEEG not to be registered at SCR.
10. Sampling times and periods may be adapted based on information obtained during the trial (e.g., due to preliminary pharmacokinetic data) including addition of samples and visits as long as the total blood volume removed does not exceed 500 mL per subject.
11. Including blood sample for metabolite identification (refer to Section 5.3.2.2).
12. A blank urine sample (0.5 mL) is to be obtained prior to administration of trial medication. Other urine samples are to be collected over the stated post-dose intervals (◄—|—|—►) 0-4, 4-8, 8-12, 12-24, and 24-48 h.

13. At baseline (i.e., Day -1, prior to drug administration) 3 triplicate ECGs are recorded within approximately one hour. The recordings should be separated by at least 15 minutes.
14. One additional blood sample for pharmacogenomics analyses (see Section 5.6.1) will be taken either at Visit 2 or at a later visit.
15. If EoT is performed on Day 4, laboratory A instead of B will be performed.
16. Dissociative symptoms are reported as AEs and should be quantified with CADSS.
17. The EEG performed at the Screening Visit is to identify and exclude subjects with an unknown susceptibility for epileptiform abnormalities and/or seizures. Therefore, Screening EEG will include spontaneous EEG activity in resting condition as well as provocation maneuvers (intermittent photic stimulation and hyperventilation) . All EEGs will be reviewed by neurologists experienced in EEG reading.
18. CADSS questionnaire to be filled in before the performance of [redacted]
19. Preparation and randomisation of medication on Day -1 and allocation of treatment to subject only on Day 1.

Applicable for Amendment No. 7

20. ECG recordings as triplicate. ECG recording will always precede all other study procedures scheduled for the same time point (e.g., blood sampling, measurement of vital signs) to avoid compromising ECG quality.

FLOW CHART – BIOAVAILABILITY/ FOOD EFFECT PART

Period	Visit	Day	Planned time (relative to first drug administration) [h:min]	Approximate clock time of actual day [h:min]	Event and comment	Safety laboratory	PK $_{blood}$[15]	EEG/ qEEG	Physical /Neurological Examination	C-SSRS	CADSS	ERP	[redacted]	12-lead ECG	Vital signs (BP, PR, RR, T)	Questioning for AEs and concomitant therapy [6]
SCR	1	-21 to -4	-504:00	08:00	Screening (SCR)[1]	A		x[11, 7]	x	x	x			x	x	
		-3	-72:00	08:00		B										
2/3/4 (three identical periods , separated by a wash-out of at least 5 days	2/3/4	-2	***-38:00***	***18:00***	***Admission to trial site***	x[5]										
			-35:30	20:30	Snack (voluntary)											
		-1	-25:00	07:00										x[14]	x	x
			-24:00	08:00					x		x[12]	x	[redacted]			
			-23:00	09:00				x								
			-21:00	11:00				x								
			-20:00	12:00								x	[redacted]			
			-16:00	16:00				x								
		1	-1:00	07:00	Allocation to treatment[2, 13] (visit 2 only)		x[2, 8]									
			-0:45	07:15					x							
			-0:30	07:30	High fat, high calorie breakfast ***(for subjects assigned to treatment T1 only)***											
			0:00	**08:00**	**Drug administration**											x
			0:05	08:05			x									▲[10]
			0:15	08:15			x									
			0:30	08:30			x							x	x	
			0:45	08:45			x	x								
			1:00	09:00			x				x				x	
			1:05	09:05										x		
			1:15	09:15			x	x								
			1:30	09:30			x									
			2:00	10:00	240 mL fluid intake		x				x			x		
			2:30	10:30			x									

Period	Visit	Day	Planned time (relative to first drug administration) [h:min]	Approximate clock time of actual day [h:min]	Event and comment	Safety laboratory	PK $_{blood}$[15]	EEG/ qEEG	Physical /Neurological Examination	C-SSRS	CADSS	ERP	[redacted]	12-lead ECG	Vital signs (BP, PR, RR, T)	Questioning for AEs and concomitant therapy [6]
			3:00	11:00			x	x								
			4:00	12:00	240 mL fluid intake, thereafter lunch[3]	B	x		x		x[12]	x	[redacted]	x	x	
			6:00	14:00			x									
			8:00	16:00	Snack (voluntary)[3]		x	x						x		
			11:00	19:00	Dinner											
			12:00	20:00			x									
			14:00	22:00			x									
		2	24:00	08:00	Breakfast (voluntary)	B	x	x	x		x[12]	x	[redacted]	x	x	
			36:00	20:00			x									
		3	48:00	08:00			x	x			x[12]	x	[redacted]	x	x	
		4	72:00	08:00	Breakfast (voluntary) ***Discharge from trial site***	B[9]	x	x	x	x	x[12]	x	[redacted]	x	x	▼
EoT	5	16 to 26			End of trial (EoT) examination[4]	A			x					x	x	x[10]

1. Subject must be informed and written informed consent obtained prior to starting any screening procedures. Screening procedures include physical examination, check of demographics (including determination of body height and weight, smoking status and alcohol history), relevant medical history, concomitant therapy and review of inclusion/exclusion criteria.
2. The time is approximate; the procedure is to be performed and completed within the 3 h prior to drug administration.
3. If several actions are indicated at the same time, the intake of meals will be the last.
4. EoT examination includes physical examination, body weight, vital signs, ECG, safety laboratory, recording of AEs and concomitant therapies.
5. Only urine drug screening and alcohol breath test will be done at this time.
6. AEs and concomitant therapies will be recorded throughout the trial, but will be specifically asked for at the times indicated in the Flow Chart above.
7. qEEG not to be registered at SCR.
8. One additional blood sample for pharmacogenomics analyses (see Section 5.6.1) will be taken either at Visit 2 or at a later visit.
9. If EoT is performed on Day 16, laboratory A instead of B will be performed.
10. Dissociative symptoms are reported as AEs and should be quantified with CADSS.
11. The EEG performed at the Screening Visit is to identify and exclude subjects with an unknown susceptibility for epileptiform abnormalities and/or seizures. Therefore, Screening EEG will include spontaneous EEG activity in resting condition as well as provocation manoeuvres (intermittent photic stimulation and hyperventilation) . All EEGs will be reviewed by neurologists experienced in EEG reading.

12. CADSS questionnaire to be filled in before the performance of [redacted]
13. Preparation and randomisation of medication on Day -1 but allocation of treatment to subject only on Day 1
14. At baseline (i.e., Day -1, prior to drug administration) 3 triplicate ECGs are recorded within approximately one hour. The recordings should be separated by at least 15 minutes.
15. Sampling times and periods may be adapted based on information obtained during the trial (e.g., due to preliminary pharmacokinetic data) including addition of samples and visits as long as the total blood volume removed does not exceed 500 mL per subject.

TABLE OF CONTENTS

ABBREVIATIONS

ADME	Absorption, distribution, metabolism, and excretion
AE	Adverse event
AESI	Adverse events of special interest
Ae_{t1-t2}	Amount of analyte eliminated in urine over the time interval t_1 to t_2
ALT	Alanine aminotransferase
ANOVA	Analysis of variance
AST	Aspartate aminotransferase
$AUC_{0-\infty}$	Area under the concentration-time curve of the analyte in plasma over the time interval from 0 extrapolated to infinity
AUC_{0-24h}	Area under the plasma concentration-time curve from time zero to 24 h
$\%AUC_{tz-\infty}$	Percentage of $AUC_{0-\infty}$ obtained by extrapolation
AUC_{t1-t2}	Area under the concentration-time curve of the analyte in plasma over the time interval t_1 to t_2
$AUC_{\tau,ss}$	Area under the concentration-time curve of the analyte in plasma at steady state over a uniform dosing interval τ
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
$AUMC_{t1-t2}$	Area under the first moment curve of the analyte in plasma over the time interval t_1 to t_2
BA	Bioavailability
BI	Boehringer Ingelheim
BMI	Body mass index (weight divided by height squared)
BP	Blood pressure
CA	Competent authority
CI	Confidence interval
CL	Total clearance of the analyte in plasma after intravascular administration
CL/F	Apparent clearance of the analyte in plasma after extravascular administration
$CL_{R,\ t1-t2}$	Renal clearance of the analyte in plasma from the time point t_1 to t_2
C_{max}	Maximum measured concentration of the analyte in plasma
CRF	Case Report Form, paper or electronic (sometimes referred to as ‘eCRF’)
C-SSRS	Columbia Suicide Severity Rating Scale
CTP	Clinical trial protocol
CTR	Clinical trial report
CV	Arithmetic coefficient of variation

DILI	Drug induced liver injury
DNA	Desoxyribonucleic acid
CADSS	Clinician Administered Dissociative States Scale
ECG	Electrocardiogram
eCRF	Electronic case report form
eDC	Electronic data capture
EDTA	Ethylenediaminetetraacetic acid
EOT	End of trial
ERP	Event Related Potential
EudraCT	European Clinical Trials Database
F	Absolute bioavailability factor
FDA	Food and Drug Administration
FE	Food effect
fe_{t1-t2}	Fraction of administered drug excreted unchanged in urine over the time interval from t_1 to t_2
GCP	Good Clinical Practice
gCV	Geometric coefficient of variation
GLP	Good laboratory practice
gMean	Geometric mean
HPLC-MS/MS	High performance liquid chromatography with tandem mass spectrometry
HR	Heart rate
HSA	Human serum albumin
IB	Investigator's brochure
IEC	Independent Ethics Committee
IRB	Institutional Review Board
ISF	Investigator site file
λ_z	Terminal rate constant of the analyte in plasma
LC-MS/MS	Liquid chromatography with tandem mass spectrometry
LLOQ	Lower limit of quantification
MDA	Methylenedioxyamphetamine
MDD	Major depressive disorder
MDMA	Methylenedioxymethamphetamine
MedDRA	Medical Dictionary for Regulatory Activities
MRT	Mean residence time of the analyte in the body
$MRT_{,ex}$	Mean residence time of the analyte in the body, extravascular
NAM	Negative allosteric modulator
NDA	New drug application
NMDA	N-methyl-D-aspartate

NOAEL	No Observed Adverse Effect Level
NR2B	Negative allosteric modulator of subunit 2B
PD	Pharmacodynamic(s)
PE	Polyethylene
PfoS	Powder for reconstitution of an oral solution
PK	Pharmacokinetic(s)
PKS	Pharmacokinetic set
PP	Polypropylene
PR	Pulse rate
QT	Time between start of the Q-wave and the end of the T-wave in an electrocardiogram
QTc	QT interval corrected for heart rate using the method of Fridericia (QTcF)
R	Reference treatment
REP	Residual effect period
RR	Respiratory rate
SAE	Serious adverse event
SCR	Screening
SOP	Standard operating procedure
SRD	Single-rising dose
SUSAR	Suspected unexpected serious adverse reaction
T	Test product or treatment
T	Temperature
TMF	Trial master file
$t_{1/2}$	Terminal half-life of the analyte in plasma
t_{max}	Time from (last) dosing to the maximum measured concentration of the analyte in plasma
TS	Treated set
t_z	Time of last measurable concentration of the analyte in plasma
TSAP	Trial statistical analysis plan
ULN	Upper limit of normal
V_{ss}	Apparent volume of distribution at steady state after intravascular administration
V_z/F	Apparent volume of distribution during the terminal phase after extravascular administration
XTC	Ecstasy

1. INTRODUCTION

1.1 MEDICAL BACKGROUND

BI 1569912 is a negative allosteric modulator of the subunit 2B (NR2B), contained in N-methyl-D-aspartate (NMDA) receptors, to be developed for major depressive disorder (MDD).

MDD is a common, severe, and frequently recurrent mental illness with an estimated global point prevalence of approximately 5% [R14-3147]. MDD poses a serious social and economic threat to modern societies, as it is a major cause of disability according to the Global Burden of Disease Study [R19-0778]. First-line antidepressants targeting the monoamine system alleviate symptoms in only 50% of patients after 12 weeks [R06-0086], and the overall cumulative remission rate with multiple treatment trials including drug switch, combination, and/ or augmentation is only 67% after up to 1 year of treatment [P06-11895]. Moreover, current treatments have a long onset of action, usually 3 to 4 weeks.

The role of glutamate in depression became apparent after it was reported that tricyclic antidepressants blocked the cation (sodium and calcium) channel associated with the NMDA glutamate receptor [R19-0681] and that functional antagonists of the NMDA receptor had antidepressant-like behavioural effects in animals. These effects were confirmed by a trial in humans in which single intravenous infusion of ketamine, an unspecific NMDA receptor antagonist used for anaesthesia, alleviated depressive symptoms in patients with depression within hours after administration of sub-anaesthetic doses, peaking some days later [R19-0772].

Since then, ketamine has demonstrated efficacy in multiple exploratory trials in patients with treatment-resistant depression. The responder rate in these trials was approximately 50%, the onset was fast and the average antidepressant effect lasted for 1 week after a single infusion [R19-0553]. Meanwhile, the intranasal S-enantiomer esketamine received NDA approval by the FDA [R19-0829].

However, transient perceptual disturbances (dissociative reaction), sedation, blood pressure increases, and an abuse potential (being a scheduled drug) require controlled distribution as well as cardiovascular and behavioural monitoring after drug application. Those unwanted effects may, at least in part, derive from ketamine's lack of selectivity, as ketamine blocks the cation channel across all NMDA subtypes [R19-0555].

Based on genetic mouse models, the NR2B subunit was identified as a key mediator of ketamine efficacy [R19-0549]. In a small Phase II study with traxoprodil, an NR2B-specific negative allosteric modulator (NAM), a rapid and robust antidepressant response without eliciting a dissociative reaction was observed [R17-3810]. These data indicate that NR2B-selective NAMs (like traxoprodil) might have a better therapeutic window compared with non-selective NMDA inhibitors (esketamine).

For further information, see current Investigator's Brochure (IB) [c29289852].

1.2 DRUG PROFILE

1.2.1 Nonclinical pharmacology

BI 1569912 is a potent negative allosteric modulator (NAM) of NR2B containing NMDA receptors with an EC_{50} of 16 nM [n00272786]. BI 1569912 is similarly potent at human, rodent and canine channels [n00272786]. Receptor binding indicates also that BI 1569912, like other known NR2B NAMs, interacts with the prodil-binding site on the NR2B subunit [n00273070]. In vivo, BI 1569912 leads to a modulation of the EEG signature in rats, indicating a physiological brain response to the treatment [n00273097].

In the forced swim test (FST) in mice BI 1569912 demonstrated efficacy with a comparable effect size as the technical control ketamine [n00273364] at a minimal effective concentration (MEC) of 52 nM. Repeated dosing over 3 days does not indicate any pharmacodynamics drug tolerance with respect to effects observed in the FST.

For further information, see current IB [c29289852].

1.2.2 Safety pharmacology

BI 1569912 was tested in a comprehensive set of safety pharmacology studies covering the ICH S7-defined core battery of tests.

Cardiovascular function

The IC_{50} for the inhibition of hERG-mediated potassium current by BI 1569912 was >30 µM [n00273899]. The compound had no effect on cardiovascular and ECG parameters in rats [n00272959] and dogs [n00273900] when tested up to 3 mg/kg, corresponding to maximum plasma levels of 7200 nM in rats and up to 6500 nM in dogs. The pro-arrhythmic potential of BI 1569912 was considered low.

Respiratory function

In rats, respiratory rate and minute volume transiently increased at 20 mg/kg and more prominently and long lasting at 300 mg/kg. The dose of 2 mg/kg was free of respiratory effects and was associated with maximum plasma levels of 3420 nM [n00271550].

CNS function

BI 1569912, given as single oral dose up to 300 mg/kg, did not affect behaviour, physiological state, or body weight in rats and had no meaningful effect on body temperature.

No effect on locomotor activity was recorded at 2 and 300 mg/kg. Considerable increases of locomotor activity were noted in males and females 0.5 h and 2 h after dosing at 20 mg/kg [n00271186]. This finding, although not dose related, was confirmed in the respiratory safety study where increased locomotor activity was noted at 20 and 300 mg/kg [n00271550].

Gastrointestinal function

In rats, BI 1569912 had no effect on gastric emptying, intestinal transit or the consistency of intestinal content. [n00267795].

Renal, hepatic and metabolic function

BI 1569912 did not relevantly influence renal function or metabolic homeostasis and did not induce the risk of renal or liver injury at clinically relevant plasma levels [n00267798]. For further information, see current IB [c29289852].

1.2.3 Toxicology

1.2.3.1 Single dose toxicity

Single dose toxicity studies have not been performed with BI 1569912.

1.2.3.2 Repeated dose toxicity

Rat and dog were selected as animal models for repeat-dose toxicity testing as the NR2B receptor is highly conserved and the prodil-binding site has 100% sequence identity across these species. BI 1569912 is similarly potent at the human, rat and dog NR2B channel with EC_{50} values of 267 nM, 147 nM, and 198 nM, respectively [n00272786]. This indicates that the pharmacological responsiveness in rats and dogs should be similar in humans. All human metabolites of BI 1569912, identified by in vitro investigations, should be covered with both toxicological species.

Toxicokinetics demonstrated exposure to BI 1569912 in all treated groups of rodents and in all dosed dogs. In both species, maximum plasma levels were similar in males and females. Therefore, subsequently, it is referred to gender averaged toxicokinetic data.

The relevant toxicity studies conducted with BI 1569912 are listed in Table 1.2.3.2: 1.

Table 1.2.3.2: 1 Relevant toxicity studies conducted with BI 1569912

Species	Study type	Applied Doses [mg/kg/day]	GLP	Dose [mg/kg/day]	NOAEL	gMean C_{max} [nM]*	gMean AUC_{0-24h} [nM*h]*	Reference
Rat	6-week oral	0, 2, 20, 300	**Yes**	**20**	**Yes**	**53600**	**78500**	[n00269064]
Dog	6-week oral	0, 0.3, 1, 3	**Yes**	**1**	**Yes**	**1560**	**2490**	[n00269216]
				3	No	4490	7850	
Dog	Dose range finding	0, 1.8, 5, 15	No	1.8	Not defined	2980**	5090**	[n00266469]
				5	No	8230	14600	
Dog	Dose escalation	1.8, 5, 15, 25, 50	No	1.8	Not defined	2770**	4990**	[n00265114]
				5	Not defined	7880**	12600**	

* Gender-averaged values ** At Day 1 only

6-week oral toxicity GLP study in rats

Male and female rats were exposed to once daily oral doses (by gavage) of 0, 2, 20, and 300 mg/kg/day for 6 weeks. Additional animals were dosed with 0 or 300 mg/kg/day and left for a 4-week recovery period after the end of dosing [n00269064].

BI 1569912 induced no effects at dose levels of 2 and 20 mg/kg/day.

At 300 mg/kg/day, clinical signs were limited to the first 8 days of dosing and were most prominent on Day 1. These were salivation, swaying gait, paddling movements (males), head shaking, somnolence, and muscle relaxation. Most signs started at time of t_{max} (1 h post-dose) and lasted for few minutes or up to 2.5 h. Chromodacryorrhea (flow of blood-stained tears) was limited to few animals 7.5 h post-dose on Day 2. Increased activity and excessive gnawing was noted on Days 2 to 8 about 15 to 90 min post-dose. There were no BI 1569912-related clinical signs from Day 9 onwards.

Slight biochemical and (histo-) pathological changes were noted. There were no BI 1569912-related effects noted during or at end of the recovery period.

The dose of 20 mg/kg/day was defined as the NOAEL with a C_{max} of 53600 nM and an AUC_{0-24h} of 78500 nM*h on Day 1.

6-week oral toxicity GLP study in dogs

Male and female dogs were exposed to once daily oral doses (by gavage) of 0, 0.3, 1, and 3 mg/kg for 6 weeks. Additional animals were dosed with 0 or 3 mg/kg/day and left for a 4-week recovery period after end of dosing [n00269216].

There were no BI 1569912-related findings at 0.3 and 1 mg/kg/day.

At 3 mg/kg/day, behavioural signs of uncoordinated movements, circling, shivering/ trembling, somnolence, and/or restlessness were noted 10 min to 2 h post-dose until Day 29. There were no clinical signs from Day 30 onwards.

At (histo-) pathology, a slight focal ulcer was present in the urinary bladder of 1 female dosed at 3 mg/kg/day. The finding was interpreted as being of uncertain relation to treatment with BI 1569912. The compound had no effect on body weight and food consumption. Cardiovascular, ophthalmology, and biochemical investigations revealed also no effect at any dose level of BI 1569912.

The dose of 3 mg/ kg (free of focal seizures or convulsions) showed systemic exposures at a mean C_{max} of 4490 nM and a mean AUC_{0-24h} of 7850 nM*h.

The dose of 1 mg/kg/day was defined as the NOAEL (free of behavioural signs) with a C_{max} of 1560 nM and an AUC_{0-24h} of 2490 nM*h on Day 1. Maximum plasma levels were achieved between 0.25 and 0.375 h after administration. Drug plasma levels were similar at end of dosing when compared with those on Day 1.

Dose range finding non-GLP study in dogs

Male and female dogs were exposed to one oral dose (by gavage) of 0, 1.8, 5, and 15 mg/kg. Marked adverse neurological signs indicative of convulsions were noted after the dose of 5 and 15 mg/kg. This led to a premature sacrifice of one male and female animal being dosed with 15 mg/kg on Day 2. Animals, surviving the treatment, were observed until Day 7 [n00266469].

Dogs, dosed at 1.8 mg/kg, showed no BI 1569912-related clinical signs. No formal NOAEL was defined. Systemic exposures were at 2980 nM for mean C_{max} (*with a minimal C_{max} value of 2360 nM*) and at 5090 nM*h for mean AUC_{0-24h} (*with a minimal AUC_{0-24h} value of 4650 nM*h*).

In one male animal, dosed with 5 mg/kg, a single episode of generalised convulsion was noted 28 h post-dose. All animals dosed with 15 mg/kg showed uncoordinated movements, circling, increased motor activity, and excitement starting about 15 min post-dose and lasting up to 7 h post-dose. Starting 15 min post-dose, two animals showed, in addition, repeated short episodes of focal seizures. In between focal seizure episodes, the dogs were excessively sniffing, shivering/ trembling, and had an uncoordinated gait. Starting about 24 hours post-dose, these animals developed repeated generalised convulsions which lasted for up to 15 min. Biochemical and pathology assessments performed in these two animals revealed findings secondary to convulsions, in particular moderately increased plasma CK and LDH activity and acute haemorrhages.

The dose of 5 mg/ kg (generalised convulsion) showed systemic exposures at a mean C_{max} of 8230 nM and a mean AUC_{0-24h} of 14600 nM*h.

Dose escalation non-GLP study in dogs

Male and female dogs were exposed to once daily oral doses of 1.8, 5, 15, 25, and 50 mg/kg for up to 13 days. [n00265114].

The male dog dosed at 50 mg/kg was prematurely sacrificed due to pertinent clinical signs of repeated convulsions, uncoordinated gait, lateral recumbency, and muscle rigidity between episodes of convulsions. These signs started approximately 24 h post-dose. Clinical pathology findings revealed signs of dehydration, stress responses, and moderate increases of AST, ALT, LDH, CK, and GLDH activities.

Starting at 15 mg/kg, uncoordinated movements, stereotypic circling associated with an increased motor activity, lambency, and salivation were observed in all animals.

BI 1569912 induced no effect at dose levels of 1.8 and 5.0 mg/kg.

Plasma concentrations did not change after repeated doses of 15 or 25 mg/kg, indicating neither marked accumulation nor enzyme auto-induction.The dose of 1.8 mg/kg (no BI 1569912-related clinical signs, no formal NOAEL defined) showed systemic exposures at a mean C_{max} of 2770 nM (*with a minimal C_{max} value of 2750 nM*) and a mean AUC_{0-24h} of 4990 nM*h (with a *minimal AUC_{0-24h} value of 4050 nM*h*).

For further information, see current IB [c29289852].

1.2.3.3 Genotoxicity

BI 1569912 was negative in the Ames test. However, it induced numerical aberrations in form of polyploid cells in human lymphocyte cultures. It also induced a predominantly aneugenic response in the long-term experiment in the in vitro micronucleus test. Therefore, it was concluded that the detected polyploidy is related to aneugenicity. BI 1569912 was clearly negative for the induction of micronuclei in peripheral blood. Aneugenicity is considered a threshold event and the free dose in rats is associated with an AUC of 239000 nM*h. The safety margin to the predicted therapeutic human exposure of 330 nM*h (see Section 1.2.5) was regarded as sufficiently large in view of this threshold-related effect to conclude that BI 1569912 is safe with regard to genotoxicity.

For further information, see current IB [c29289852].

1.2.3.4 Reproductive and developmental toxicity

Histopathology in all repeat dose toxicity studies revealed no effect of BI 1569912 on the male reproductive tract of mice, rats, and dogs. Dedicated reproductive toxicity studies have not been performed with BI 1569912.

For further information, see current IB [c29289852].

1.2.3.5 Carcinogenicity

Carcinogenicity studies have not been conducted with BI 1569912.

1.2.3.6 Phototoxicity

BI 1569912 was negative in BALB/c 3T3 Neutral Red Uptake tests and is therefore considered not phototoxic. No direct phototoxicity is anticipated in humans.

For further information, see current IB [c29289852].

1.2.3.7 Neurotoxicity

NMDA receptor antagonists are known to induce neurotoxicity in rats, i.e., vacuolation of the cingulate and retrosplenial cortex, named 'Olney's lesions' [R97-2207]. Therefore, special attention was paid in all repeat-dose toxicity studies that the brain was sectioned as recommended by Bolon et al. [R17-1266] and it was assured that cingulate and retrosplenial cortex were present in the slides examined.

In a dedicated neuropathology study [n00273820] with single administrations to rats of doses up to 300 mg/kg, BI 1569912 induced no vacuolation of neurons ('Olney's lesions') and no neuronal death at systemic exposures with C_{max} levels of 614000 nM and $AUC_{0\text{-}24h}$ levels of 4510000 nM*h.

For further information, see current IB [c29289852].

1.2.3.8 Abuse and dependency

BI 1569912 did not show significant interaction to a broad panel of receptors, channels and enzymes in vitro, including targets of abuse potential [n00272895]. When compared with esketamine, BI 1569912 showed only a mild increase of psychomimetic-like behaviour in an in vivo tolerability study in mice at a dose of 1 mg/kg.

Neurobehavioural assessment of dogs revealed no effect after a single dose of up to 3 mg/kg [n00273900]. Locomotor activity was increased at dose levels of 20 mg/kg or higher. In repeat-dose toxicity studies in rats, behavioural signs were limited to clinically irrelevant high drug plasma levels. In both rats and dogs, the behavioural signs ameliorated with repeated dosing and completely disappeared until study end, indicating habituation. Drug withdrawal did not induce any effects in the recovery animals of the 6-week rat and dog study.

The preclinical data reveal only weak and inconclusive evidence for an abuse potential of BI 1569912 and its overall risk to induce abuse or dependency is considered low.

For further information, see current IB [c29289852].

1.2.4 Nonclinical pharmacokinetics

Validated HPLC-MS/MS assays are available for the quantification of BI 1569912 plasma concentrations in rat [n00272730],[n00272731] and dog [n00272732]. The lower limit of quantification of the assays is 1.00 nM. [^{14}C] BI 1569912 was synthesised for use in in vitro and in vivo studies.

1.2.4.1 Drug Absorption and Disposition

Dedicated pharmacokinetic studies in animals to characterize the absorption of BI 1569912 have not been performed.

Plasma protein binding

Plasma protein binding of BI 1569912 was assessed in vitro by equilibrium dialysis [n00272974]. Plasma protein binding was similar across the investigated species, with 72.2% (fraction unbound (fU): 27.8%) in man, 74.0% (f_U: 26.0%) in mouse, 88.7% (f_U: 11.3%) in rat and 65.5% (f_U: 34.5%) in dog. Both, HSA and hAGP contribute to the total protein binding of BI°1569912 in human plasma with HSA as the predominant binding partner and a minor contribution of hAGP.

In vivo distribution studies

Whole body autoradiography in rats [n00272820] demonstrated that the overall extent of distribution of BI 1569912 from plasma to tissues was predominantly low with tissue-to-plasma ratios of 0.21 to 0.59. Radioactivity was detectable and quantifiable in the central nervous system only at 0.5 h after oral dosing with percentages of radioactivity concentrations in different parts of CNS (related to whole blood) amounting to about 40%. High affinity and long-term (yet reversible) exposures of radioactivity to ocular tissues were found with a half-life of drug-related radioactivity in the eye (melanin containing tissue and total) of about 80 h. There was no discernible affinity and only short-term exposure of radioactivity to the integumentary system.

Metabolism

Relevant metabolites, formed by metabolic Phase I reactions in human hepatocytes, were also formed in hepatocytes of mouse, rat, dog, and Göttingen minipig hepatocyte incubations (research data on file). Dedicated in vivo metabolism studies have not yet been performed.

Excretion

Studies investigating the excretion (ADME study) have not yet been performed.

For further information, see current IB [c29289852].

1.2.4.2 Potential Pharmacokinetic Interaction

In vitro studies investigating potential drug-drug interactions on drug metabolism or drug transport have not yet been performed.

1.2.5 Prediction of human pharmacokinetics

A human clearance of 2.89 mL/min/kg was predicted based on in vitro-in vivo correlation. A human volume of distribution at steady state of 0.678 L/kg was predicted based on mean of animals. These resulted in a predicted human disposition mean residence time and effective half-life of 3.91 h and 2.7 h, respectively, and single therapeutic dose of 2.0 mg (free base) to achieve a C_{max} of 52 nM in human.

Human therapeutic dose of 2.0 mg is predicted based on a minimum effective total plasma concentration of 52 nM in the mouse FST. Considering similar potency in human compared with the mouse, and no significant difference in protein binding between both species, this also translates into an anticipated effective total plasma concentration of approximately 52 nM, which was set as target C_{max} for the prediction of human pharmacokinetics and dose.

A summary of the predicted human pharmacokinetic parameters and therapeutic dose is presented in Table 1.2.5: 1.

Table 1.2.5: 1 Summary of the predicted pharmacokinetic parameters and therapeutic dose of BI 1569912 in human

Parameters	Prediction
CL [mL/min/kg]	2.89
V_{ss} [L/kg]	0.678
MRT disp [h]	3.91
F [%]	58.2
MAT [h]	2.09
k_a [h-1]	2.47
MRT tot [h]	5.99
t_{max} [h]	1.39
$t_{1/2}$,eff [h]	2.7
Exposure target (C_{max}) [nM]	52
$AUC_{0\text{-}inf}$ [nM*h]	330
C_{max} [nM]	52
Therapeutic dose [mg]	2.0

Source data: [n00272965]

For further information, see current IB [c29289852].

1.2.6 Clinical experience in humans

To date, no clinical studies with BI 1569912 have been performed.

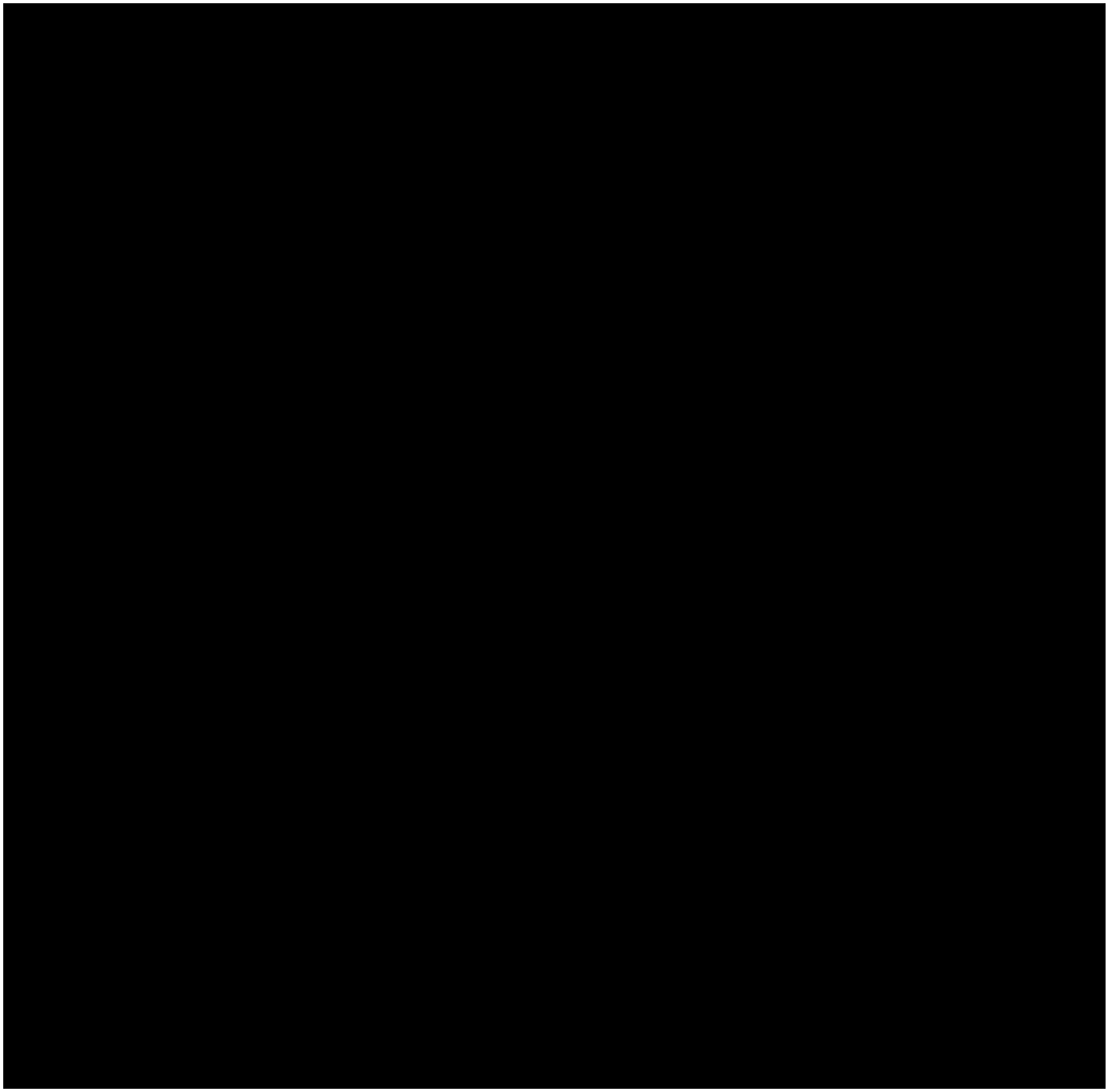
However, clinical information on compounds of the same (NR2B NAM) or related (unselective NMDA inhibitors) pharmacological classes are available.

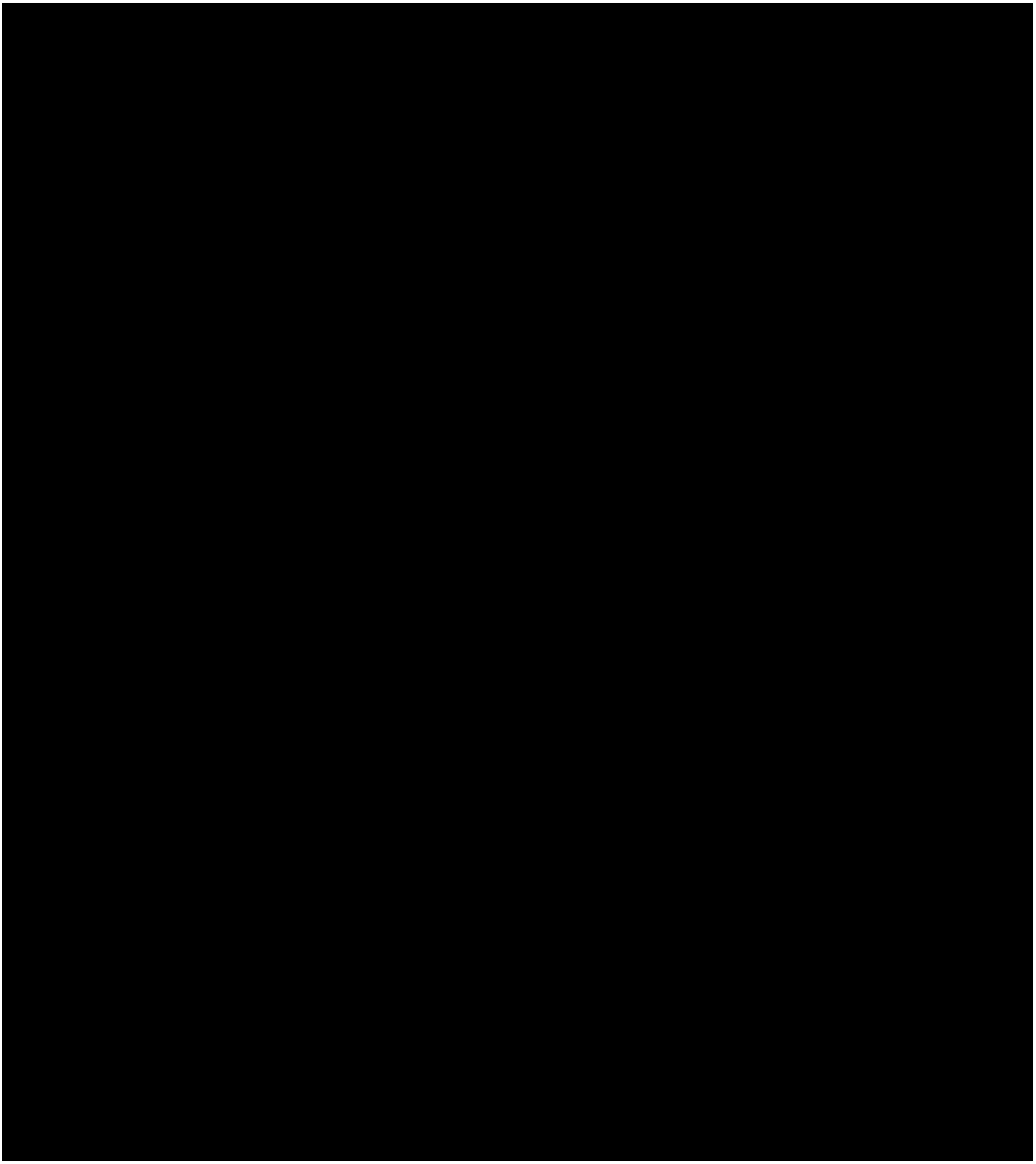
In a phase II study with traxoprodil administered intravenously, transient dissociative symptoms such as out-of-body experience, tunnel vision, derealisation, or depersonalization were reported. They were dose dependent, transient in nature and resolved completely within hours. Other side effects were mild and included abnormal feeling, dizziness, paraesthesia, somnolence, dry mouth, abnormal urine odour, and an increase in blood pressure [R17-3810]. The clinical development of traxoprodil was discontinued, probably for a CYP 2D6 liability or effects on the QT/ QTc interval [P06-11048], [R19-1029].

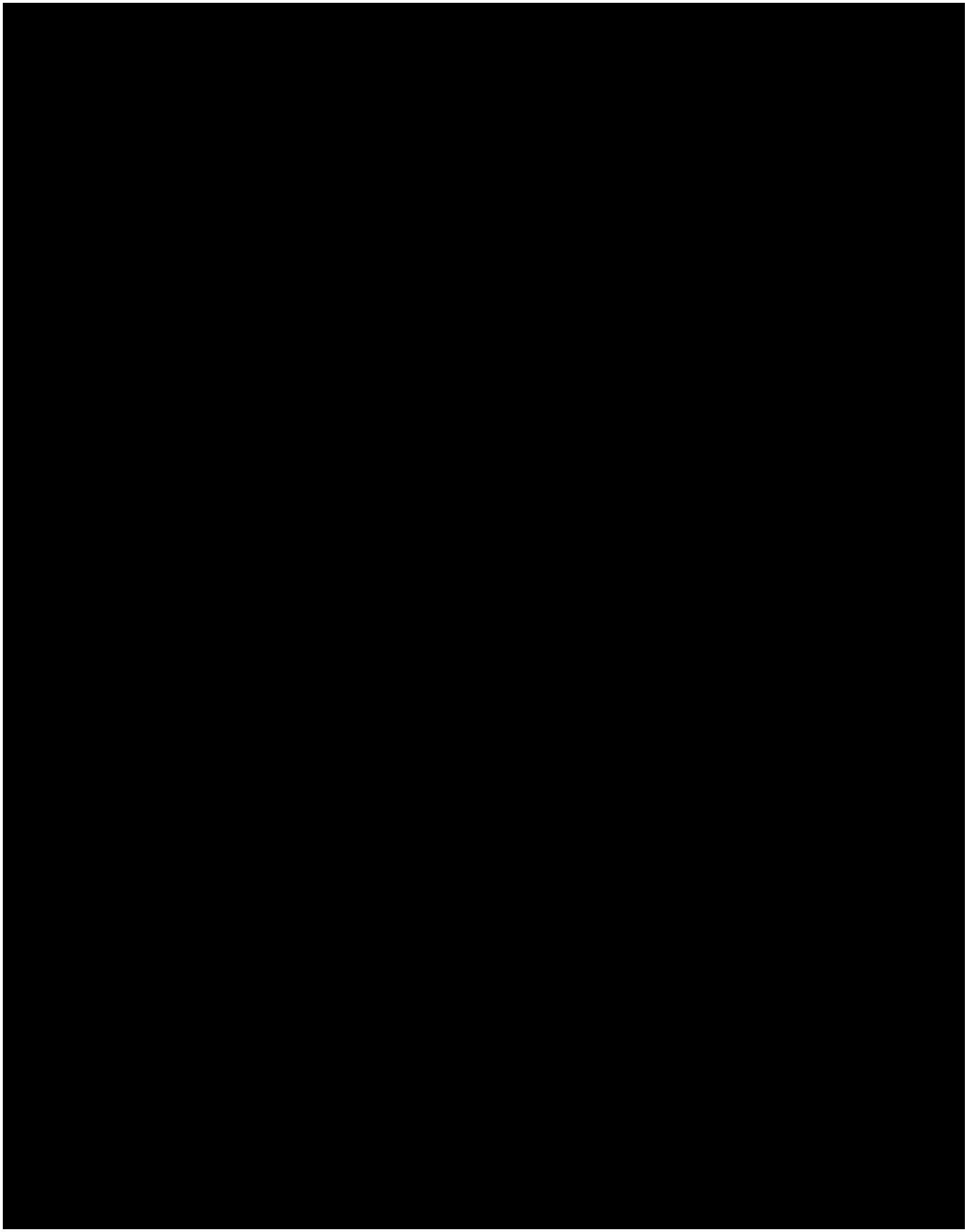
In phase 2 studies with rislenemdaz, dizziness, headache, diarrhoea, dry mouth, somnolence, paraesthesia, and blood pressure increase were reported. Rislenemdaz did not lead to dissociative symptoms [R19-0986].

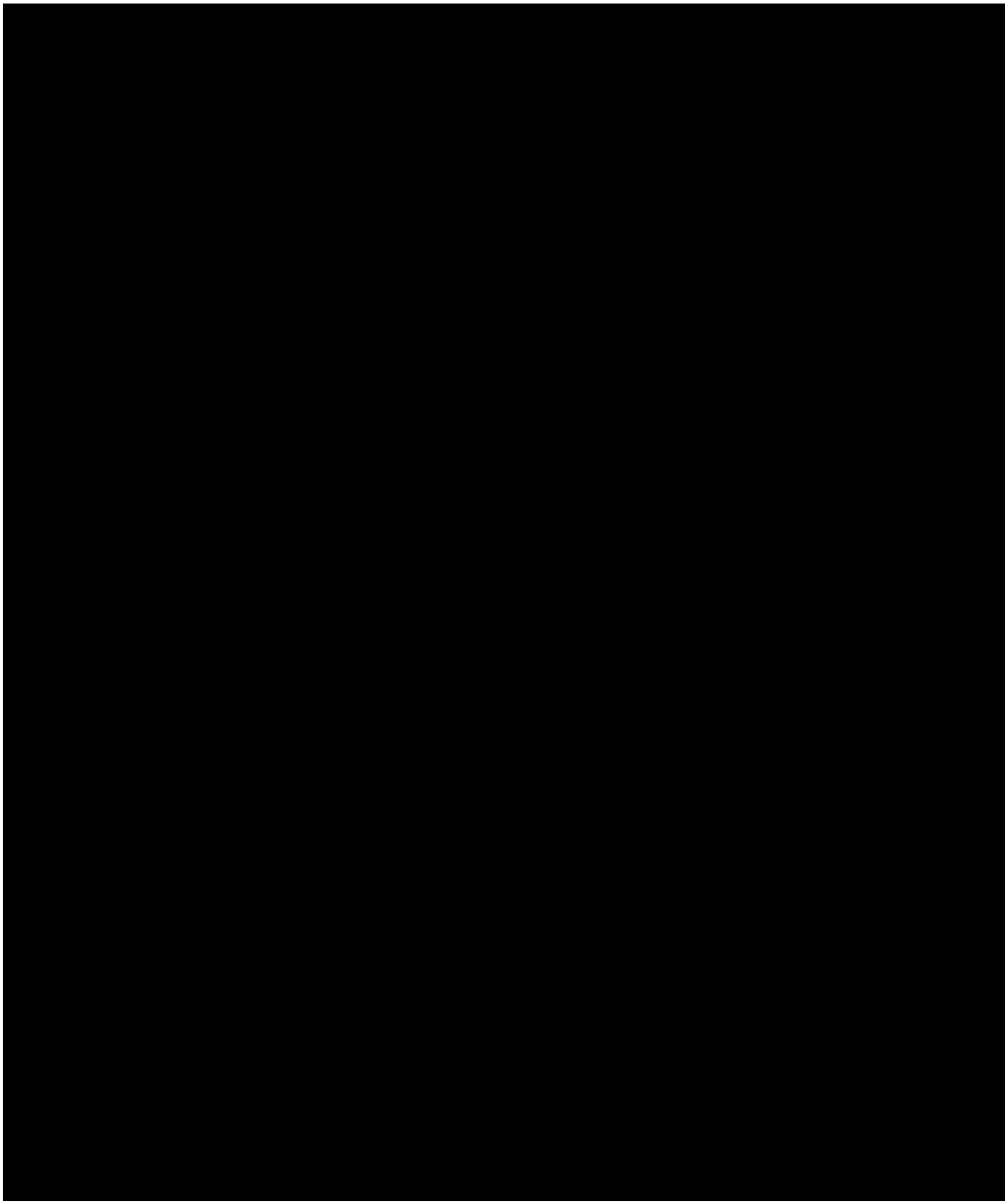
Overall, for these NR2B-specific negative allosteric modulators, no serious safety concerns have been identified.

For unspecific NMDA inhibitors, like esketamine, the most important side effects were transient CNS effects, like dissociation (altered time and sensory perception) and sedation as well as increases in blood pressure. In phase 3 studies with esketamine, transient dizziness, dissociative/ perceptual symptoms, nausea, vertigo, headache, dysgeusia, paraesthesia, hypaesthesia, sedation, and vomiting were reported. Dissociative and sedative side effects typically peaked within 2 h after drug administration and resolved within 6 h. Transient increases in blood pressure were also common.









1.2.7 Residual Effect Period

The Residual Effect Period (REP) of BI 1569912 in humans is not known to date. This is the period after the last dose with measurable drug levels and/ or pharmacodynamic effects still likely to be present. Based on the pharmacokinetic half-life of about 3 hours, it could be determined with about 15 hours. However, due to observed seizure in dogs at a dose of 5 mg/kg/day at 28 h post-dose, the REP is estimated up to 36 hours, until further information is available.

1.2.8 Drug product

For a more detailed description of the BI 1569912 profile, see current IB [c29289852].

BI 1569912 has a molecular weight of 360 g/mol and appears as a white to off white powder. It is very soluble at low pH values and freely soluble at neutral pH (solubility terms according to Ph. Eur.).

Two oral dosage forms have been developed for clinical use:

- Powder for oral Solution (PfoS) with a co-supplied solvent
- Tablets

The PfoS will be applied in the single rising dose (SRD)- and the relative bioavailability/ food effect (BA/ FE)-Part. Tablets will be applied in the BA/ FE-Part only.

Powder for Oral Solution/ Solvent for Oral Solution

An oral solution is prepared for dosing at the clinical site according to the instructions in Appendix 10.1, using a powder and solvent for oral solution. The powder for oral solution is the BI 1569912 drug substance in amber glass bottles with plastic, screw-cap closures. The solvent for oral solution is a tartaric acid solution 0.5%, provided in amber glass bottles with plastic, screw-cap closures. The solvent alone is used as a placebo.

The oral solution will be prepared with BI 1569912 drug substance to a concentration of 0.625 mg/mL in order to cover the dose range of 0.25 mg to 50 mg.

The powder for oral solution and solvent for oral solution should be stored in the containers provided, and handled according to the labelled storage instructions and shelf life. Based on the available data, there are no special storage precautions for the powder for oral solution.

The solvent for oral solution should not be frozen.

The prepared oral dosing solution should be used within the in-use period stated on the label other trial documents.

Tablets

BI 1569912 tablets have been developed in 3 dosage strengths: 0.5 mg (approx. 5.5 mm round), 2.5 mg (approx. 10 mm round) and 5 mg (approx. 17.8x8.6 mm oval). In addition to the drug substance, the tablets contain the following standard pharmaceutical excipients in common amounts: lactose monohydrate, microcrystalline cellulose, hydroxypropyl cellulose, croscarmellose sodium, and magnesium stearate.

The clinical trial supplies will be provided in polypropylene bottles with screw-cap closures.

The tablets should be stored in the containers provided and handled according to the labelled storage instructions and shelf life.

For further information, see Section 4.1 and current IB [c29289852].

1.3 RATIONALE FOR PERFORMING THE TRIAL

As a transition from preclinical investigations to clinical development, safety, tolerability, pharmacokinetics and pharmacodynamics of BI 1569912 will be assessed in healthy male subjects using single rising oral doses in order to provide a basis for the further clinical development in the indication of MDD.

Healthy male subjects will be recruited for this study. They provide a relatively stable physiological, biochemical and hormonal basis for studying drug effects, show no disease-related variations and are not taking regular concomitant medications.

In the SRD-Part, within each dose group, all actively treated individuals will receive the same BI 1569912 dose. The next higher dose will only be administered to the next group, if the treatment in the preceding dose group was safe and showed acceptable tolerability.

In the BA/ FE-Part, BI 1569912 will be administered to subjects in a randomized three-way crossover fashion to understand the effect of food on relative bioavailability of the tablet formulation and to support upcoming clinical studies in respect to trial designs.

1.3.1 Justification for starting dose

An estimation was made based on the US FDA Guidance for Industry 'Estimating the Maximum Recommended Safe Starting Dose in Initial Clinical Trials for Therapeutics in Healthy Volunteers' [R06-1037].

Step 1: NOAEL Determination

The NOAELs for BI 1569912 (see Table 1.2.3.2: 1) after daily oral treatment over 6 weeks are:
Rat: 20 mg/kg/day
Dog: 1 mg/kg/day

Step 2: Human Equivalent Dose (HED) Calculation:

Rat: NOAEL = 20 mg/kg/day

Conversion factor from rat dose to HED: 6.2
HED = 20 mg/kg/day, divided by 6.2 = 3.2 mg/kg/day
= 161 mg/subject (body weight 50 kg)

Dog: NOAEL = 1 mg/kg/day
Conversion factor from dog dose to HED: 1.8
HED = 1 mg/kg/day, divided by 1.8 = 0.56 mg/kg/day
= 27.8 mg/subject (body weight 50 kg)

Step 3: Most Appropriate Species Selection

Based on HED calculations, the dog is the most sensitive species.

Step 4: Application of Safety Factor

Due to observed convulsions in dogs in the 5 and 15 mg/kg dose groups (see Section 1.2.3.2), a safety factor of 30, instead of the conventional factor of 10, will be applied.
Calculation of the Maximum Recommended Starting Dose (MRSD):
MRSD = 27.8 mg/subject (HED)/ 30 (safety factor) = 0.93 mg/ subject

Step 5: Consideration of the Pharmacologically Active Dose (PAD)

The therapeutic dose of BI 1569912 is predicted to be about 2 mg/day (see Section 1.2.7). The application of a safety factor of 10 would result in a MRSD of 0.2 mg. Since this dose is lower than the 0.93 mg/day, 0.2 mg/ day is selected as the MRSD and rounded up to 0.25 mg which results in 0.4 mL of oral solution. This dose is not expected to trigger any relevant or unacceptable biological activities.

1.3.2 Justification for dose escalation scheme

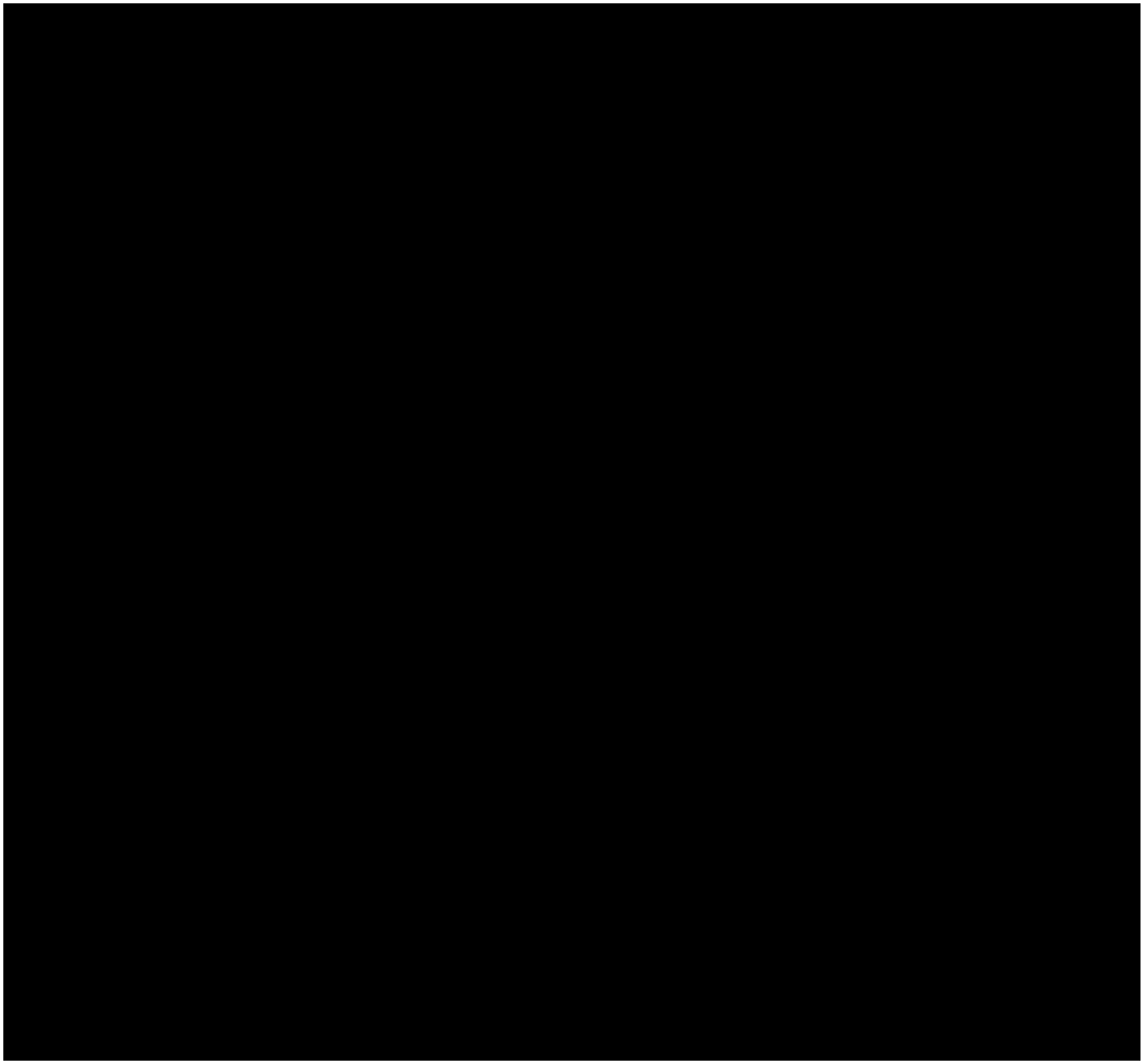
Dose escalation in the SRD-Part will be guided (besides by safety parameters) by preliminary (monitored and predicted) pharmacokinetic parameters, i.e., C_{max} and AUC_{0-24h} in healthy subjects.

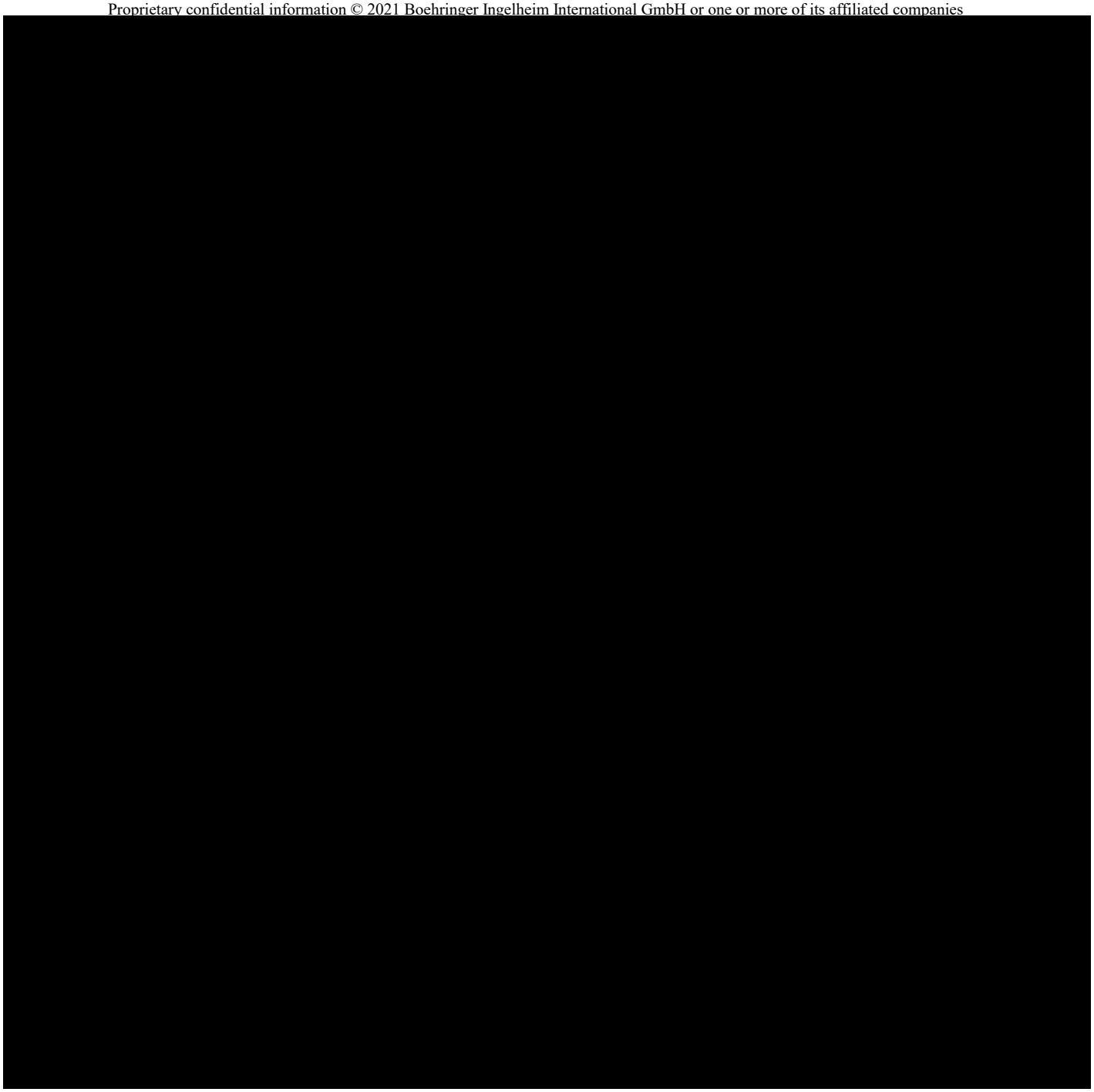
1.3.2.1 Maximum systemic exposure and safety margins

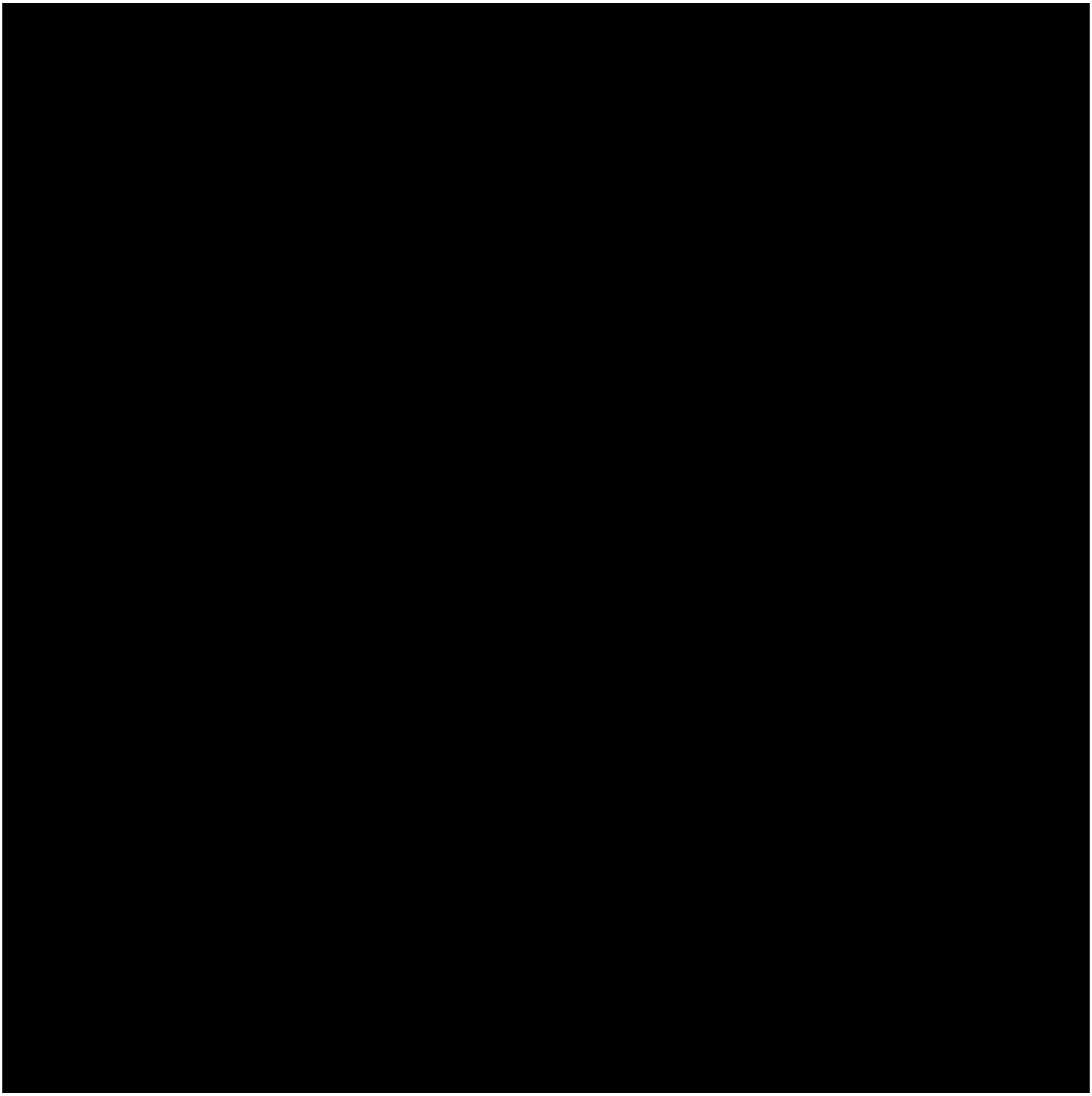
In the 6-week oral toxicity GLP dog study, the dose of 1 mg/ kg was free of behavioural signs and was defined as NOAEL at a mean C_{max} of 1560 nM and a mean AUC_{0-24h} of 2490 nM*h. In the same toxicity study, the dose of 3 mg/ kg induced behavioural signs but was free of focal seizures/ convulsions at a mean C_{max} of 4490 nM and a mean AUC_{0-24h} of 7850 nM*h. These observed behavioural signs could (at least in part) indicate exaggerated pharmacology of BI 1569912. In the dose range finding non-GLP dog study, the dose of 5 mg/ kg induced generalised convulsions at a mean C_{max} of 8230 nM and a mean AUC_{0-24h} of 14600 nM*h.

Therefore, the maximal acceptable systemic exposure is defined as gMean C_{max} of 1560 nM and a gMean AUC_{0-24h} of 2490 nM*h which should not be surpassed with the highest applied

dose in the SRD-Part of the study. This would be about 5-fold (C_{max}) and 6-fold (AUC_{0-24h}) lower than the exposures in dog at doses inducing generalised convulsions, and about 3-fold (C_{max} and AUC_{0-24h}) lower than the exposures in dog at doses inducing behavioural signs.







1.3.2.2 Dose escalation

Dose escalation will start with 0.25 mg. Decreasing escalation factors, from 3 to 1.25, between dose escalating steps will be applied up to the highest planned dose of 50 mg. As noted above, the doses will progress only as long as the maximal acceptable systemic exposure has not been reached.

As long as the pre-defined maximum systemic exposure has not been reached, higher doses than the expected therapeutic dose of 2 mg may be justified for the following reasons:

First, subsequent clinical trials in patients may indicate that the required therapeutic dose (exposure) is significantly higher than predicted. Also, higher doses (exposures) might be needed for obese patients (different volumes of distribution) or in patients with various concomitant diseases. While higher doses (exposures) may still be well tolerated, they provide a larger variety and magnitude of therapeutic effects.

Further, testing of doses higher than 2 mg is justified to account for uncertainties in translation from non-clinical to clinical information, i.e., actual pharmacokinetic or pharmacodynamics parameters may deviate from predicted values, like

- Bioavailability may be (significantly) less than predicted
- Clearance may be higher than predicted
- Half-life may be shorter than predicted
- Pharmacokinetics may be non-linear with a less than dose-proportional increase,
- Translation from pharmacodynamics data o to human

After each completed dose level, preliminary safety and pharmacokinetic data will be reviewed by the sponsor and Principal Investigator to decide on the escalation to the next planned dose level. Each dose escalation decision will be based on preliminary

- Safety and tolerability data of all available subjects on active treatment from at least 24 hours after the administration of study medication and
- Pharmacokinetic data collected in a minimum of 3 subjects on active treatment from each dose group from at least 24 hours after the administration of study medication (see Section 3.1).

1.3.3 Justification for relative bioavailability and food effect part

The BI 1569912 tablet formulation will only be available during the course of the study. Since the development program is planned to be continued with a tablet formulation, a relative bioavailability/ food effect part (BA/ FE-Part) will also be performed. To further characterize the tablet formulation, it will be examined with and without food (FE-Part).

Three dose strengths of BI 1569912 tablets will be provided: a 5 mg, 2.5 mg and 0.5 mg tablet. Only that tablet strength will be tested against the same dose of oral solution which was demonstrated to be safe and tolerable in the SRD-Part. Since the oral bioavailability was estimated to be about 60% (see Section 1.2.5) a safety factor of at least 2 will be applied between the safe and tolerable dose in the SRD-part and the applied doses in the BA/ FE-Part. This is to account for uncertainties of predictions and the theoretically possible increase of bioavailability by food intake. As such, the tablet with 5 mg will only be tested in the BA/ FE-Part, if at least 10 mg are safe and tolerated in the SRD-Part. Accordingly, the tablet with 2.5 mg will only be tested, if at least 5 mg are safe and tolerated and the tablet with 0.5 mg will only be tested, if at least 2 mg is safe and tolerated in the SRD-Part. Before starting the BA/ FE-Part, a summary of safety and tolerability data (including AEs, SAEs, ARs and

SARs) generated so fare during the course of the trial will be submitted via substantial amendment.

1.4 BENEFIT - RISK ASSESSMENT

Participation in this clinical trial is without any (therapeutic) benefit for healthy subjects. Their participation, however, is of major importance to the development of BI 1569912.

Subjects are exposed to risks of study procedures and risks related to the exposure to the trial medication.

Procedure-related risks

The use of an indwelling venous catheter or venepuncture for e.g., blood sampling may result in mild bruising and, in rare cases, in transient inflammation of the wall of the vein, or nerve injury, potentially resulting in paraesthesia, reduced sensibility, and/or pain for an indefinite period.

The total volume of blood withdrawn per subject during the entire study will not exceed the volume of a normal blood donation (500 mL). No health-related risk to healthy subjects is expected from withdrawal of this volume of blood.

Drug-related risks and safety measures

Factors of risk may derive from particular knowledge or the lack thereof, regarding (1) the mode of action, (2) the nature of the target, (3) the relevance of animal models and/ or (4) findings in non-clinical safety studies. Further aspects will be (5) risks resulting from trial medication auxiliaries, and (6) drug induced liver injury, resulting in (7) measures of risk minimization (including safety precautions and stopping rules – see Section 1.4.7).

1.4.1 Mode of action

BI 1569912 is a negative allosteric modulator of subunit 2B (NR2B) containing N-methyl-D-aspartate (NMDA) receptors which is a therapeutic concept for depression that has been well described [R17-3810]. Clinical information on compounds of the same pharmacological class (NR2B NAMs like traxoprodil and rislenemdaz) or a related pharmacological class (unselective NMDA inhibitors, like esketamine) are available. For NR2B-specific negative allosteric modulators, no serious safety concerns have been identified (see Section 1.2.6).

1.4.2 Nature of the target

BI 1569912 is a partial, reversible and time-dependent negative allosteric modulator of the human NR2B-subunit contained in NMDA receptors (sodium and calcium channels). As such, BI 1569912 is not considered to be a high risk compound.

1.4.3 Relevance of animal models

Rat and dog were selected as animal models for repeat-dose toxicity testing, as the NR2B receptor is highly conserved and the prodil-binding site has 100% sequence identity across these species. BI 1569912 is similarly potent at the human, rat and dog NR2B subunit with EC_{50} values of 267 nM, 147 nM, and 198 nM, respectively [n00272786]. This indicates that the pharmacological responsiveness in rats and dogs should be similar to that in humans (see Section 1.2.3.2). In addition, plasma protein binding is similar across the investigated species, with a BI 1569912 free plasma fraction (f_U) in humans of 27.8% and in dogs of 34.5% (see Section 1.2.4.1). Therefore, the safety assessments based on these non-clinical animal models are relevant and should be directly translatable to humans.

1.4.4 Findings in non-clinical safety studies

The pro-arrhythmic potential of BI 1569912 was considered to be low up to a C_{max} of 6500 nM in dogs. In rats, the compound was free of respiratory effects up to a C_{max} of 3420 nM. Gastrointestinal, renal, hepatic and metabolic functions were not affected at predicted relevant clinical exposures (see Section 1.2.2). In a dedicated neuropathology study, BI 1569912 did not induce 'Olney lesions' (vacuolation of neurons) or neuronal death (see Section 1.2.3.7).

BI 1569912 was negative in an Ames test. Although it showed aneugenic activity in a chromosome aberration test and a micronucleus test with human lymphocytes in vitro, BI 1569912 is considered to be safe with regard to genotoxicity (see Section 1.2.3.3). No direct phototoxicity is anticipated in humans (see Section 1.2.3.6). Preclinical data reveal only weak and inconclusive evidence for an abuse potential of BI 1569912 and its overall risk to induce abuse or dependency is considered low (see Section 1.2.8).

The dog was chosen as the most appropriate toxicological species (see Section 1.3.1). The dose of 1 mg/kg/day in the 6-week oral toxicity GLP dog study was free of behavioural signs and defined as the NOAEL with a C_{max} of 1560 nM and an AUC_{0-24h} of 2490 nM*h. The dose group of 3 mg/kg/day in the same toxicity study was free of focal seizures or convulsions but induced behavioural signs at systemic exposures with a mean C_{max} of 4490 nM and a mean AUC_{0-24h} of 7850 nM*h. The dose of 5 mg/kg/day in the dose range finding non-GLP dog study induced a self-limiting generalised convulsion at systemic exposures with a mean C_{max} of 8230 nM and a mean AUC_{0-24h} of 14600 nM*h. (see Section 1.2.3.2).

1.4.5 Risks resulting from trial medication auxiliaries

Tablets contain lactose.

1.4.6 Drug induced liver injury

Although rare, a potential for drug-induced liver injury (DILI) is under constant surveillance by sponsors and regulators. Therefore, this trial requires timely detection, evaluation, and follow-up of laboratory alterations in selected liver laboratory parameters to ensure subjects´ safety (see Section 5.2.7.1.4).

1.4.7 Measures of risk minimization (including safety precautions and stopping rules)

The following precautionary measures will be taken in this study in order to minimize the risk for healthy subjects:

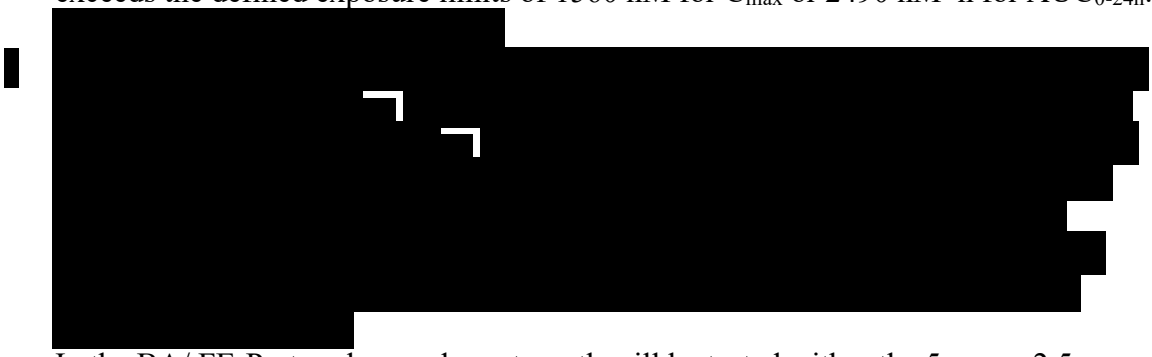
General Risk Minimization Measures:

- Careful selection of study subjects according to in- and exclusion criteria (see Section 3.3.2 and Section 3.3.3).
- Careful starting dose selection (see Section 1.3.1).
- Decreasing escalation factors with increasing doses (see Section 1.3.3).
- In the SRD-Part, for safety reasons, each dose group of 8 subjects (6 on active, 2 on placebo) will be divided into three cohorts of 2 (sentinel cohort), 3 and 3 subjects which will be separated by a time interval of at least 48 hours. The first 4 subjects of each dose group (2 subjects of the sentinel cohort and the first 2 subjects of the second cohort, i.e., random block 1) will be dosed in a fixed sequence manner (active – placebo – active – active). The remaining 4 subjects of each dose group (the last subject of the second cohort and 3 subjects of the third cohort, i.e., random block 2) will be randomised in a 3:1 ratio (active drug to placebo). While drug administration in the first cohort can be performed simultaneously (active - placebo), drug administration in the second and third cohort will be separated by at least 60 minutes. For orally administered drugs, this is usually a sufficient time interval to observe acute effects and to achieve a relevant proportion of systemic exposure.
- Extensive standard safety laboratory measurements will be performed before and after drug administration (see Flow Chart and Section 5.2.3).
- Close monitoring of blood pressure (see Flow Charts).
- A thorough ECG and heart rate monitoring will be performed, including continuous ECG measurements, over 4 hours post dose to cover the anticipated period of highest drug exposure, and additional repeated single 12-lead ECGs (see Flow Charts).
- Prior to each dose escalation, a documented safety review will be performed by the Principal Investigator or an authorized deputy and the Clinical Trial Leader or an authorized deputy (see Section 3.1).

BI 1569912 Specific Risk Minimization Measures:

- In each dose group, the three cohorts (see above) will be separated by at least 48 hours (between first subjects of each cohort) which is expected to cover the period of highest risk/ peak effect.
- Interim measurements of BI 1569912 plasma levels will be performed. The expected geometric mean systemic exposure (C_{max}, AUC_{0-24h}) in the next higher dose group will be estimated based on current and preceding doses using a dose proportionality or model-based approach, as applicable. The next higher dose level will only be administered, if the predicted geometric mean values of C_{max} or AUC_{0-24h} do not exceed the defined exposure limits of 1560 nM for C_{max} or 2490 nM*h for AUC_{0-24h}

(see Section 1.3.2.1) or, if no individually observed value of the current dose group exceeds the defined exposure limits of 1560 nM for C_{max} or 2490 nM*h for AUC_{0-24h}.



- In the BA/ FE-Part, only one dose strength will be tested, either the 5 mg or 2.5 mg or 0.5 mg tablet. Only the highest dose strength, as demonstrated in the SRD-Part to be safe and tolerable, will be tested, reduced by a safety factor of at least 2. Before starting the BA/ FE-Part, a summary of safety and tolerability data (including AEs, SAEs, ARs and SARs) generated so fare during the course of the trial will be submitted via substantial amendment.
- Subjects will be confined to the study site under close observation for at least 72 hours after drug administration at the trial site and will be discharged only after a formal assessment and confirmation of fitness by an investigator or qualified designee. During in-house confinement, subjects will be under medical observation and thoroughly monitored for both, expected and unexpected adverse events.
- Subjects will be evaluated with electrophysiological and functional assessments to identify any possible adverse event. Specifically, any signs of pro-convulsive activity of the brain will be assessed by repeated EEG measurements in each dose group. Close neurological/ psychiatric evaluation using a standardized clinical assessments for the detection of neurological symptoms, the Columbia-Suicide Severity Rating Scale for the assessment of suicidality and the CADSS for the assessment of suspected dissociative symptoms. The occurrence of anticipated dissociative symptoms will be reduced by quiet ambient conditions during and after drug administration.
- The preliminary determination of the REP is up to 36 hours.

Stopping Rules Related to Individual Subjects:

- Occurrence of one adverse event of severe intensity (including but not limited to QT prolongation, dissociative symptoms or a decrease in vigilance) assessed as related to the study drug by the investigator

- Occurrence of one serious adverse event assessed as related to the study drug by the investigator

Stopping Rules Related to Dose Groups:

- In case of any safety concerns, as listed below but not limited to them, the trial will be mandatorily discontinued. Occurrence of severe non-serious adverse events

considered as drug-related by the investigator in 2 subjects of the same dose group (8 subjects) in the SRD-Part, or 3 subjects of the BA/ FE group (12 subjects), or occurrence of at least one drug-related serious adverse event (SAE). Moreover, dose escalation in the SRD-Part will be terminated, if more than 3 of the actively dosed subjects at one dose level show drug-related and clinically relevant adverse events of at least moderate intensity.

- Clinical apparent seizures/ convulsions in one or more subjects in the same dose group.
- EEG changes consistent with pro-convulsive activities (e.g., spike and wave) that is considered to be drug-related in two or more subjects of the same dose group.
- Clinically relevant changes in the neurological assessment indicating dose-limiting intolerance that is considered to be drug-related in one or more subjects of the same dose group.
- Dose escalation in the SRD-Part or treatment in the BA/ FE-Part will be stopped, if at least 2 subjects on active treatment at one dose level (SRD-Part) or 2 subjects in the BA/ FE-Part have relevant individual QT prolongations, i.e., a QTc increase of greater than 60 ms from baseline and/ or an absolute QT or QTc greater than 500 ms, as confirmed by a repeat ECG recording.
- Dose escalation in the SRD-Part will be stopped, if the C_{max} or AUC_{0-24h} of at least 1 subject of one dose group increases above the following exposure thresholds or if the estimated geometric mean exposure is expected to exceed a C_{max} of 1560 nM or an AUC_{0-24h} of 2490 nM*h. In this case, one or two additional dose levels lower than the planned next dose level may be given, as long as the expected geometric mean exposure values of the interim dose do not exceed these exposure thresholds. Estimation will be done based on preliminary pharmacokinetic results of preceding dose groups (see Section 7.4)

Applicable for Amendment No. 7

[redacted]

1.4.8 Overall assessment and conclusion

Based on the well-characterized mode of action, nature of target, high relevance of used animal models and the comprehensive non-clinical safety package of BI 1569912, and taking into account specific safety measures, a participation in this single dose regimen does not represent an undue risk to healthy subjects. Considering the medical need for a better MDD treatment and taking into account the potential advantage of a highly selective NR2B negative allosteric modulator, the expected benefit of this trial is likely to outweigh the potential risks and justifies the exposure of healthy subjects to BI 1569912.

2. TRIAL OBJECTIVES AND ENDPOINTS

2.1 MAIN OBJECTIVES, PRIMARY AND SECONDARY ENDPOINTS

2.1.1 Main objectives

Main objectives of this trial are to investigate for

SRD-Part: Safety, tolerability, pharmacokinetics and pharmacodynamics following single rising doses of BI 1569912

BA/FE-Part: (a) the relative bioavailability of BI 1569912 PfoS and tablet, and

(b) the influence of food on the relative bioavailability of the BI 1569912 tablet

2.1.2 Primary endpoint

SRD-Part: The primary endpoint for assessment of safety and tolerability of BI 1569912 is the percentage of subjects with drug-related adverse events.

BA/FE-Part: The following pharmacokinetic parameters will be determined for BI 1569912:

- 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)

2.1.3 Secondary endpoint

SRD-Part: The following pharmacokinetic parameters will be determined, if feasible:

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

BA/FE-Part: The following pharmacokinetic parameter will be determined for BI 1569912:

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

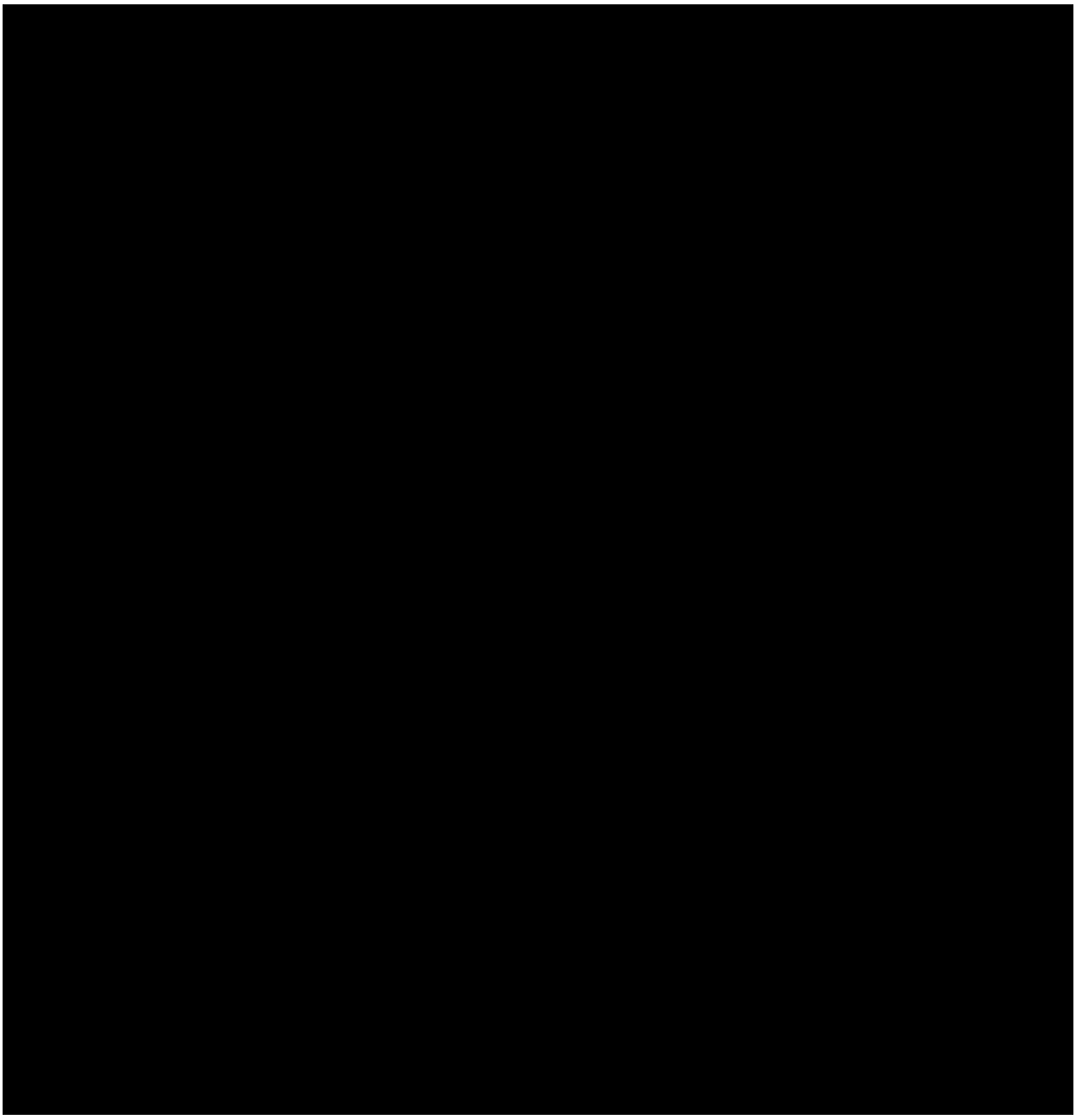
[redacted]

[redacted]

2.2.2.2 Further BI 1569912 specific endpoints of safety and tolerability

- Columbia-Suicide Severity Rating Scale (C-SSRS)
- Standardized physical/ neurological assessment
- Assessment of dissociative symptoms (e.g., CADSS)
- Electroencephalogram (EEG)

[redacted]



3. DESCRIPTION OF DESIGN AND TRIAL POPULATION

3.1 OVERALL TRIAL DESIGN AND PLAN

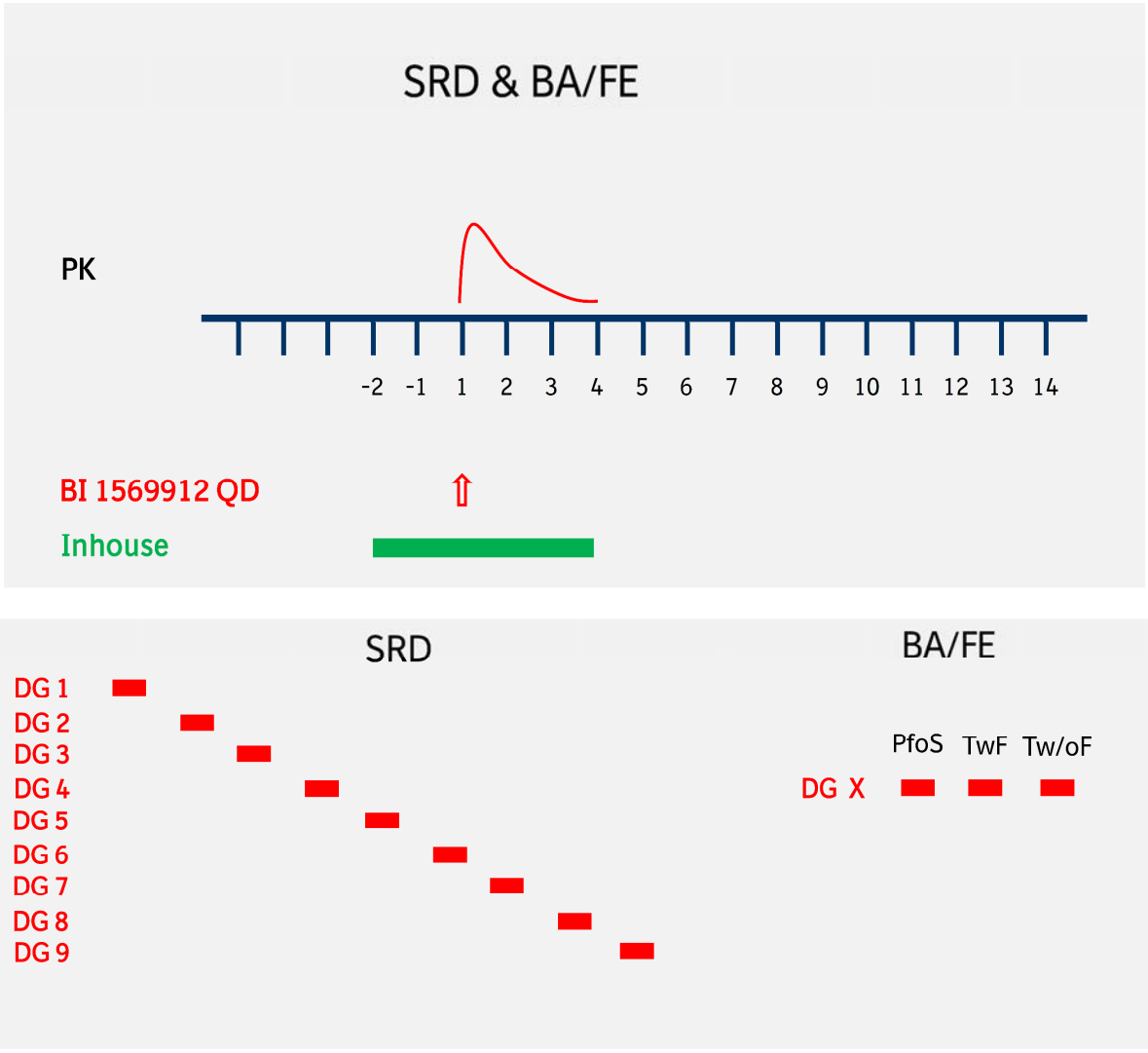


Figure 3.1: 1 Overall study design

SRD-Part

This single-rising dose part will be performed as a single-blind, partially randomised within dose group, placebo-controlled parallel-group trial.

It is planned to include a total of 72 healthy male subjects in the trial. The subjects will be assigned to 9 groups consisting of 8 subjects per group. These groups will be dosed sequentially (see Table 3.1: 1). The investigator (after consultation with the sponsor) is allowed to alter the scheduled dose groups (e.g., add low and/or intermediate dose groups) on

the basis of experience gained during the study (for instance, based on preliminary pharmacokinetic data), under the condition that the planned and approved highest dose is not exceeded. Thus, the actual number of subjects entered may be more than 72 but is not to exceed 88. Such changes may be implemented via non-substantial CTP amendments.

However, the addition of further dose groups exceeding the already tested dose levels for the evaluation of safety findings will be subject to a substantial CTP amendment requiring approval.

Within each dose group, 6 subjects will receive BI 1569912 and 2 will receive placebo. Only one dose is tested within each dose group.

In the SRD-Part, for safety reasons, each dose group of 8 subjects (6 on active, 2 on placebo) will be divided into three cohorts (for details, see Section 1.4.7, 'General Risk Minimization Measures).

The dose groups to be evaluated are outlined in Table 3.1: 1 below.

Table 3.1: 1 Dose groups

Dose Group	1	2	3	4	5	6	7	8	9
Daily dose (mg)	0.25	0.75	2	5	10	20	30	40	50
Number of subjects	8	8	8	8	8	8	8	8	8
Subjects receiving placebo	2	2	2	2	2	2	2	2	2
Subjects receiving BI 1569912	6	6	6	6	6	6	6	6	6

The groups will be dosed consecutively in ascending order, and a time interval of at least 7 days will be maintained between the last drug administration to subjects in the previous dose group and the first drug administration to subjects in the subsequent dose group. The decision to treat the next dose group will be based upon safety, tolerability and pharmacokinetic data of all the preceding dose groups. The next dose group will only be applied, if, in the opinion of the Principal Investigator (or an authorised deputy) and the Clinical Trial Leader (or an authorised deputy), no safety concerns have arisen in the preceding dose groups (i.e., no dose-limiting events occurred) and, if none of the pre-specified trial-specific stopping criteria have been met (refer to Section 3.3.4.2).

A documented safety review must take place prior to each dose escalation. Furthermore, an unscheduled safety review meeting can be requested anytime by the Principal Investigator (or an authorised deputy) or the sponsor of the study (for instance, due to the occurrence of any unforeseen adverse events).

Although no formal safety review meeting will take place within a given dose group, safety will be continuously monitored during this trial, and an individual will only be dosed in the absence of any safety concern (i.e., no dose-limiting events occurred) and if none of the pre-specified trial-specific stopping criteria have been met (refer to Section 3.3.4.2).

At a minimum, the following data need to be available for review to escalate to a higher dose.

- AEs in the current and preceding dose groups up to at least 48 h post dosing, including clinically relevant findings from ancillary safety testing listed below (Note: AEs may be ongoing at the time of Safety Reviews and AE information may be subject to change prior to Database Lock)
- Results from 12-lead ECG and continuous ECG monitoring in the current and preceding dose groups up to at least 48 h post dosing
- Vital signs in the current and preceding dose groups up to at least 48 h post dosing
- Clinical laboratory tests in the current and preceding dose groups up to at least 24 h post dosing
- EEG reports up to at least 24 h post dosing
- Preliminary pharmacokinetic data of at least 3 subjects on active treatment for up to at least 24 h post dosing as per Section 7.4
- Check of criteria for stopping subject treatment as per Section 3.3.4.1

The decision to escalate the dose will be made jointly by the Principal Investigator (or an authorised deputy) and the Clinical Trial Leader (or an authorised deputy) after in-depth analysis of all available safety data, especially SAEs (if occurred), AEs, and out-of-range laboratory results (if considered clinically relevant). In addition and depending on the results and findings, suitable experts from the sponsor or external institutions (e.g. [redacted]) may be consulted on an 'as needed' basis. In these cases, expert recommendations will be documented in the minutes of the safety review and considered for the decision making. Dose escalation will only be permitted, if no safety concerns exist neither in the opinion of the Principal Investigator (or an authorised deputy) nor the Clinical Trial Leader (or an authorised deputy).

Safety Reviews can be conducted face-to-face or by video/telephone conference. The Clinical Trial Leader is responsible for the organisation and minutes of the reviews. Minutes will be signed off by the Principal Investigator (or an authorised deputy) and Clinical Trial Leader (or an authorised deputy), and will be filed in the ISF and TMF.

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

BA/ FE-Part

Before starting the BA/ FE-Part, a summary of safety and tolerability data (including AEs, SAEs, ARs and SARs) generated so fare during the course of the trial will be submitted via a substantial amendment.
The BA/ FE part will be performed as an open-label, randomised, single-dose, intra-individual, six-sequence, three-way crossover trial in healthy male subjects in order to assess (1) the relative bioavailability of the tablet relative to PfoS (both administered under fasted conditions) and (2) the relative bioavailability of the tablet administered under fed and fasted conditions. For details, refer to Section 4.1.4. Subjects will be randomly allocated to the 6 treatment sequences. There will be a washout period of at least 5 days between the

treatments, i.e., the last dose in the first treatment period and the first dose in the following treatment period are separated by at least 5 days.

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

3.2 DISCUSSION OF TRIAL DESIGN, INCLUDING THE CHOICE OF CONTROL GROUPS

SRD-Part

For single-rising dose trials, the sequential rising dose design described in Section 3.1 is viewed favourably under the provision not to expose the subjects involved to undue risks.

Single-blind conditions regarding the subject's treatment (active or placebo) are maintained within each dose group. However, subjects and investigators will be aware of the dose of drug administered. The disadvantage of the trial design is a possible observer bias with regard to the dose-dependent effects; in addition, the sequential dosing of groups could potentially result in time-related effects. However, as such effects are expected to be small relative to the differences between the doses in the broad range investigated, unbiased comparisons between treatments can still be expected.

It is standard in single rising dose trials involving healthy volunteers to include a placebo group to control for safety, tolerability, and pharmacodynamic effects of the trial medication. Each dose group consists of 8 subjects, with 6 on active treatment, and 2 on placebo. For data analysis purposes, the placebo control group will include all subjects of all dose groups treated with placebo. Six subjects per active treatment group are generally considered to be sufficient for the exploratory evaluation of pharmacokinetics.

BA/ FE-Part

For relative bioavailability/ bioequivalence trials, the crossover design is preferred because of its efficiency: since each subject serves as hisown control, the comparison between formulations/ treatments is based on an intra-subject comparison, thus removing inter-subject variability from the comparison between formulations/treatments [R94-1529].

The open-label treatment is not expected to bias results, since the study endpoints are derived from measurement of plasma and urine concentrations of the analyte.

3.3 SELECTION OF TRIAL POPULATION

SRD-Part & BA/ FE-Part

It is planned that 84 healthy male will enter the study. The actual number of subjects entered may exceed the total of 84, if additional intermediate doses are tested (see Section 3.1). Subjects will be recruited from the volunteers' pool of the trial site.

Only male subjects will be included in the trial because no data on reproductive toxicology are available at this time.

A log of all subjects enrolled into the trial (i.e. who have signed informed consent) will be maintained in the ISF irrespective of whether they have been treated with investigational drug or not.

3.3.1 Main diagnosis for trial entry

The study will be performed in healthy male subjects.

3.3.2 Inclusion criteria

Subjects will only be included in the trial if they meet the following criteria:

1. Healthy male subjects according to the assessment of the investigator, as based on a complete medical history including a physical examination, vital signs (BP, PR, RR, T), 12-lead ECG, and clinical laboratory tests
2. Age of 18 to 45 years (inclusive)
3. BMI of 18.5 to 29.9 kg/m2 (inclusive)
4. Signed and dated written informed consent prior to admission to the study, in accordance with GCP and local legislation
5. Male subjects who meet any of the following criteria from at least 30 days before the first administration of trial medication until 30 days after trial completion:
    - Use of adequate contraception, e.g., any of the following methods (of female partners) *plus* condom: implants, injectables, combined oral or vaginal contraceptives, intrauterine device
    - Sexually abstinent
    - Surgically sterilised (including hysterectomy of female partner)
    - Postmenopausal female partner, defined as at least 1 year of spontaneous amenorrhea (in questionable cases a blood sample with FSH above 40 U/L and estradiol below 30 ng/L is confirmatory)

3.3.3 Exclusion criteria

Subjects will not be allowed to participate, if any of the following general criteria apply:

1. Any finding in the medical examination (including BP, PR or ECG) deviating from normal and assessed as clinically relevant by the investigator
2. Repeated measurement of systolic blood pressure outside the range of 90 to 140 mmHg, diastolic blood pressure outside the range of 50 to 90 mmHg, or pulse rate outside the range of 50 to 90 bpm
3. Any laboratory value outside the reference range that the investigator considers to be of clinical relevance, in particular, hepatic parameters (ALT, AST, total bilirubin) or renal parameters (creatinine) exceeding the ULN after repeated measurements
4. Any evidence of a concomitant disease assessed as clinically relevant by the investigator
5. Gastrointestinal, hepatic, renal, respiratory, cardiovascular, metabolic, immunological or hormonal disorders

6. Cholecystectomy or other surgery of the gastrointestinal tract that could interfere with the pharmacokinetics of the trial medication (except appendectomy or simple hernia repair)
7. Diseases of the central nervous system (including but not limited to any kind of seizures/ convulsions or stroke), and other relevant neurological or psychiatric disorders
8. History of relevant orthostatic hypotension, fainting spells, or any unexplained blackouts
9. Chronic or relevant acute infections
10. History of relevant allergy or hypersensitivity (including allergy to the trial medication or its excipients)
11. Use of drugs within 30 days (or within five times of their respective elimination half-lives – whatever is longer) of planned administration of trial medication that might reasonably influence the results of the trial (including drugs that cause QT/ QTc interval prolongation)
12. Intake of an investigational drug in another clinical trial within 60 days (or within five times of the respective elimination half-live – whatever is longer) of planned administration of investigational drug in the current trial, or concurrent participation in another clinical trial in which investigational drug is administered
13. Smoker (more than 10 cigarettes or 3 cigars or 3 pipes per day)
14. Inability to refrain from smoking on specified trial days
15. Alcohol abuse (consumption of more than 24 g per day)
16. Drug abuse or positive drug screening
17. Blood donation of more than 100 mL within 30 days of planned administration of trial medication or intended blood donation during the trial
18. Sperm donation from the time of drug administration until 30 days thereafter
19. Intention to perform excessive physical activities within one week prior to the administration of trial medication or during the trial
20. Inability to comply with the dietary regimen of the trial site
21. A marked baseline prolongation of QT/ QTc interval (such as QTc intervals that are repeatedly greater than 450 ms) or any other relevant ECG finding at screening
22. A history of additional risk factors for Torsade de Pointes (such as heart failure, hypokalaemia, or family history of Long QT Syndrome)
23. Subject is assessed as unsuitable for inclusion by the investigator, for instance, because the subject is not considered able to understand and comply with study requirements, or has a condition that would not allow safe participation in the study, or is in custody by order of an authority or court of law

In addition, the following trial-specific exclusion criteria apply:

24. Any lifetime history of suicidal behaviour (i.e. actual attempt, interrupted attempt, aborted attempt, or preparatory acts or behaviour)
25. Any suicidal ideation of type 2 to 5 on the C-SSRS (i.e. active suicidal thought, active suicidal thought with method, active suicidal thought with intent but without specific plan, or active suicidal thought with plan and intent) in the past year prior to randomization

26. History or presence of epilepsy, history of more than one febrile seizure in childhood or a family history of seizures/ convulsions
27. Epileptiform abnormalities in EEG at Screening
28. History of clinically relevant head injury or trauma (e.g., associated with loss of consciousness)

In addition, the following SARS-CoV-2/COVID-19-specific exclusion criteria apply:

29. A positive PCR test for SARS-CoV-2/COVID-19 and/ or any clinical symptom suggestive for this disease at screening and on Day -3.

For study restrictions, refer to Section 4.2.2.

3.3.4 Withdrawal of subjects from treatment or assessments

Subjects may discontinue trial treatment or withdraw consent to trial participation as a whole ('withdrawal of consent') with very different implications; see Sections 3.3.4.1 and 3.3.4.2 below.

If a subject is removed or withdraws from the trial prior to the first administration of trial medication, the data of this subject will not be entered in the case report form (CRF) and will not be reported in the clinical trial report (CTR). If a subject is removed or withdraws from the trial after the first administration of trial medication, this will be documented and the reason for discontinuation must be recorded in the CRF. In addition, the data will be included in the CRF and will be reported in the CTR.

At the time of discontinuation, a complete end of trial (EoT) examination will be performed, if possible, and the information will be recorded in the CRF. If the discontinuation occurs before the end of the REP (see Section 1.2.6), the discontinued subject should, if possible, be questioned for AEs and concomitant therapies at or after the end of the REP in order to ensure collection of AEs and concomitant therapies throughout the REP, if not contrary to any consent withdrawal of the subject.

3.3.4.1 Discontinuation of trial treatment

An individual subject will discontinue trial treatment if:

1. The subject wants to discontinue trial treatment, without the need to justify the decision
2. The subject has repeatedly shown to be non-compliant with important trial procedures and, in the opinion of both, the investigator and sponsor representative, is not willing or able to adhere to the trial requirements in the future
3. The subject needs to take concomitant medication that interferes with the investigational medicinal product or other trial treatment
4. The subject can no longer receive trial treatment for medical reasons (such as surgery, adverse events (AEs), or diseases)
5. An AE or clinically significant laboratory change or abnormality occurs that the investigator assesses as warranting discontinuation of treatment. This may include cases of sustained symptomatic hypotension (BP <90/50 mmHg) or hypertension (BP

>180/100 mmHg), clinically relevant changes in ECG requiring intervention, unexplained hepatic enzyme elevations at any time during the trial, or development of suicidal ideation

6. The subject has an elevation of AST and/or ALT ≥3-fold ULN and an elevation of total bilirubin ≥2-fold ULN (measured in the same blood sample) and/or needs to be followed up according to the DILI checklist provided in the ISF

In addition to these criteria, the investigator may discontinue subjects at any time based on his or her clinical judgment.

Even if the trial treatment is discontinued, the subject remains in the trial and, given his agreement, will undergo the procedures for early treatment discontinuation and follow up as outlined in the Flow Chart and Section 6.2.3.

3.3.4.2 Withdrawal of consent to trial participation

Subjects may withdraw their consent to trial participation at any time without the need to justify the decision. If a subject wants to withdraw consent, the investigator should be involved in the discussion with the subject and explain the difference between trial treatment discontinuation and withdrawal of consent to trial participation, as well as explain the options for continued follow up after trial treatment discontinuation. See also Section 3.3.4.1 above.

3.3.4.3 Discontinuation of the trial by the sponsor

Boehringer Ingelheim reserves the right to discontinue the trial at any time for any of the following reasons:

1. Failure to meet expected enrolment goals overall or at a particular trial site
2. New toxicological findings, serious adverse events, or any safety information invalidating the earlier positive benefit-risk assessment.
3. Violation of GCP, or the CTP impairing the appropriate conduct of the trial.
4. The sponsor decides to discontinue the further development of the investigational product.

The investigator/ trial site will be reimbursed for reasonable expenses incurred in case of trial termination (except if item 3 applies).

3.3.5 Replacement of subjects

SRD & BA/ FE-Part

A maximum of 4 subjects can replace those who dropped-out during the course the trial for safety reasons related or not related to the study medication. A replacement subject will be assigned a unique study subject number, and will be assigned to the same treatment as the subject he replaces.

4. TREATMENTS

4.1 INVESTIGATIONAL TREATMENTS

The investigational products have been manufactured by BI Pharma GmbH & Co. KG.

See also Section 1.2.7 and current IB [c29289852].

4.1.1 Identity of the Investigational Medicinal Products

SRD-Part

The characteristics of the test product are given below:

Substance:	BI 1569912
Pharmaceutical formulation:	Powder for oral solution
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	0.625 mg/ mL; 0.25 mg, 0.75 mg, 2 mg, 5 mg, 10 mg, 20 mg, 30 mg, 40 mg, 50 mg QD
Posology:	1-0-0
Route of administration:	Oral
Duration of use:	Single dose

The characteristics of the reference product (placebo) are given below:

Substance:	Not applicable, solvent only
Pharmaceutical formulation:	Solvent only
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	N/A
Posology:	1-0-0
Route of administration:	Oral
Duration of use:	Single dose

The matching placebo is the solvent for oral solution only (Tartaric acid 5 mg/mL)

BA/ FE-Part

Only the highest dose strength, reduced by a factor of at least 2, will be tested, as demonstrated in the SRD-Part to be safe and tolerable (refer also to Section 1.3.3 and Section 1.4.7.

The characteristics of the test product are given below:

Substance:	BI 1569912
Pharmaceutical formulation:	Tablet
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	5 mg
Posology:	1-0-0
Route of administration:	Oral
Duration of use:	Single dose in each treatment period

The characteristics of the test product are given below:

Substance:	BI 1569912
Pharmaceutical formulation:	Tablet
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	2.5 mg
Posology:	1-0-0
Route of administration:	Oral
Duration of use:	Single dose in each treatment period

The characteristics of the test product are given below:

Substance:	BI 1569912
Pharmaceutical formulation:	Tablet
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	0.5 mg
Posology:	1-0-0
Route of administration:	Oral
Duration of use:	Single dose in each treatment period

4.1.2 Selection of doses in the trial

SRD-Part

Doses selected for this trial part cover the sub-therapeutic as well as the estimated therapeutic range and include a safety margin (see Section 1.2).

BA/ FE-Part

The dose selected for this trial part is the highest tablet dose strength that was demonstrated to be safe and tolerable in the SRD-Part reduced by a safety factor of 2. (see Section 1.3.3 and Section 4.1.1).

4.1.3 Method of assigning subjects to treatment groups

Prior to the screening visit, subjects will be contacted in writing and informed about the planned visit dates. Subjects willing to participate in the SRD-Part will be recruited to dose groups (consisting of 3 cohorts) according to their temporal availability.. Therefore, the allocation of subjects to dose groups is not influenced by trial personnel but only by the subjects' temporal availability. Because the study includes healthy subjects from a homogenous population, relevant imbalances between the dose groups are not expected.

Subjects will be assigned to treatments (active treatment or placebo) in the SRD part or to treatment sequences in the BA/ FE-Part prior to the first administration of trial medication. For this purpose, the respective randomisation lists will be provided to the trial site in advance. Numbers of the randomization lists will be allocated by the investigator in ascending, sequential order to eligible subjects. Subjects are then assigned to treatment (SRD part) or treatment sequences (BA/ FE part) according to the randomisation list.

The randomisation procedure is described in Section 7.6.

Once a subject number has been assigned, it cannot be reassigned to any other subject.

4.1.4 Drug assignment and administration of doses for each subject

SRD-Part

The treatments to be evaluated are outlined in Table 4.1.4: 1 below. The number of units/dose volume for placebo corresponds to the number of units/ dose volume of the corresponding dose level.

Table 4.1.4: 1 BI 1569912 and placebo treatments, oral administration

Dose group	Substance	Pharmaceutical form	Unit strength	Dose volume per administration	Total daily dose
1	BI 1569912	oral solution	0.625 mg/mL	0.4 mL (0.25 mg) single dose	0.25 mg
2	BI 1569912	oral solution	0.625 mg/mL	1.2 mL (0.75 mg) single dose	0.75 mg
3	BI 1569912	oral solution	0.625 mg/mL	3.2 mL (2 mg) single dose	2 mg
4	BI 1569912	oral solution	0.625 mg/mL	8 mL (5 mg) single dose	5 mg
5	BI 1569912	oral solution	0.625 mg/mL	16 mL (10 mg) single dose	10 mg
6	BI 1569912	oral solution	0.625 mg/mL	32 mL (20 mg) single dose	20 mg
7	BI 1569912	oral solution	0.625 mg/mL	48 mL (30 mg) single dose	30 mg
8	BI 1569912	oral solution	0.625 mg/mL	64 mL (40 mg) single dose	40 mg
9	BI 1569912	oral solution	0.625 mg/mL	80 mL (50 mg) single dose	50 mg
1-9	Placebo*	oral solution	--	identical to active treatment	--

* Subjects receiving placebo are equally distributed across dose groups

The oral solutions for dosing (active treatment and placebo) will be prepared by pharmacists or qualified pharmacy staff members or qualified medical study personnel at the trial site under the responsibility of the investigator according to the instructions provided in Appendix 10.1.

Administration of trial medication will be performed after subjects have fasted overnight; fasting is to start no later than 10 h before the scheduled dosing. The investigator (or authorised designee) will administer the trial medication as an oral dose together with about 240 mL of water to subjects who are in a standing position. For drug administration, the so-called four-eye principle (two-person rule) should be applied. For this, 1 authorised employee of the trial site should witness the administration of trial medication, and – if applicable – its preparation (e.g., reconstitution), if correct dosage cannot be ensured otherwise.

Subjects will be kept under close medical surveillance until 72 h after drug administration. During the first 4 h after drug administration, subjects are not allowed to lie down (i.e. no declination of the upper body of more than 45 degrees from upright posture except for medical examination) or to sleep.

BA/ FE-Part

This trial is a 3-way crossover study. All subjects will receive 3 treatments in randomised order. **Only one** of the treatment schemes will be evaluated as outlined in Tables 4.1.4: 2 to 4.1.4: 4 below.

Table 4.1.4: 2 Dosage strength of 5 mg BI 1569912 and treatment schedule

Treatment	Substance	Formulation	Unit strength	Dosage	Total dose
T1 (Test 1)	BI 1569912	Tablet (fed)	5 mg	1 tablet (5 mg) single dose	5 mg
T2 (Test 2)	BI 1569912	Tablet (fasted)	5 mg	1 tablet (5 mg) single dose	5 mg
R (Reference)	BI 1569912	Oral solution (fasted)	0.625 mg/mL	8 mL (5 mg) single dose	5 mg

Table 4.1.4: 3 Dosage strength of 2.5 mg BI 1569912 and treatment schedule

Treatment	Substance	Formulation	Unit strength	Dosage	Total dose
T1 (Test 1)	BI 1569912	Tablet (fed)	2.5 mg	1 tablet (2.5 mg) single dose	2.5 mg
T2 (Test 2)	BI 1569912	Tablet (fasted)	2.5 mg	1 tablet (2.5 mg) single dose	2.5 mg
R (Reference)	BI 1569912	Oral solution (fasted)	0.625 mg/mL	4 mL (2.5 mg) single dose	2.5 mg

Table 4.1.4: 4 Dosage strength of 0.5 mg BI 1569912 and treatment schedule

Treatment	Substance	Formulation	Unit strength	Dosage	Total dose
T1 (Test 1)	BI 1569912	Tablet (fed)	0.5 mg	1 tablet (0.5 mg) single dose	0.5 mg
T2 (Test 2)	BI 1569912	Tablet (fasted)	0.5 mg	1 tablet (0.5 mg) single dose	0.5 mg
R (Reference)	BI 1569912	Oral solution (fasted)	0.625 mg/mL	0.8 mL (0.5 mg) single dose	0.5 mg

Administration of trial medication will be performed after subjects have fasted overnight; fasting is to start no later than 10 h before the scheduled dosing. The investigator (or authorised designee) will administer the trial medication as an oral dose together with about 240 mL of water to subjects who are in a standing position. For drug administration, the so-called four-eye principle (two-person rule) should be applied. For this, one authorised employee of the trial site should witness the administration of trial medication, and – if applicable – its preparation (e.g., reconstitution), if correct dosage cannot be ensured otherwise.

In T1 treatment, a high-fat, high-calorie meal will be served 30 min before drug administration. The subjects must completely consume the meal prior to drug intake. The composition of the standard high-fat, high-calorie meal is detailed in Table 4.1.4: 5; this meal is in compliance with the FDA guidance 'Food-Effect Bioavailability and Fed Bioequivalence Studies' [R03-2269]. For restrictions with regard to diet, see Section 4.2.2.2.

Table 4.1.4: 5 Composition of the high-fat, high-calorie meal

Ingredients	kcal
2 chicken eggs (whole content) for scrambled eggs	192
10 g butter for frying scrambled eggs	75
35 g fried bacon	186
2 toasted slices of wheat bread	130
15 g butter for buttering toast slices	113
115 g hash brown potatoes	132
240 mL whole milk (3.5% fat)	156
Sum[1]	984

[1] The total caloric content was supplied approximately as following: 150 kcal as protein, 250 kcal as carbohydrate, and 500 to 600 kcal as fat.

Subjects will be kept under close medical surveillance until 72 h after drug administration. During the first 4 h after drug administration, subjects are not allowed to lie down (i.e. no

declination of the upper body of more than 45 degrees from upright posture, except for medical examination).

The treatments will be separated by a wash-out phase of at least 5 days/weeks.

4.1.5 Blinding and procedures for unblinding

4.1.5.1 Blinding

SRD-Part

The SRD part of the trial is designed single-blind. The treatments administered (active or placebo) will be blinded to subjects, but will be known to the investigators (outcome assessors). Only the current dose level will be known to the subjects due to the rising dose design.

A single-blind design is considered acceptable because the potential bias in this type of study seems to be low and according to study procedures it is assured that the investigator's knowledge of the next treatment does not influence the decision to enter a subject.

All trial data will be handled open label. This means that trial functions of the sponsor are unblinded (including Clinical Trial Leader, data manager, statistician, bioanalyst, pharmacokineticist, pharmacometrician, drug metabolism scientist as well as dedicated personnel of the trial site).

Access to the randomisation schedule will be controlled and documented by a signed confidentiality statement, which will be stored in the TMF.

Applicable for Amendment No. 7

Within the central ECG lab, the staff involved with interval measurements will be blinded with respect to subject, treatment , recording date and time as well as planned time points of the ECGs. For quality control of the measurements, certain members of the ECG evaluation team will be unblinded to subject.

If a preliminary analysis of ECG data is required during the trial, a part of the staff of the central ECG lab (different from the ECG evaluation team) may be unblinded. This part of the staff will receive the necessary information which will be stored with no access to the ECG evaluation team.

Access to the randomisation schedule will be controlled and documented by a signed confidentiality statement, which will be stored in the TMF.

BA/ FE-Part

This Phase I trial will be handled in an open fashion throughout (that is, during the conduct, including data cleaning and preparation of the analysis). This is considered acceptable because the potential for bias seems to be low and does not outweigh practical considerations. Emergency envelopes will not be provided, because the dose of trial medication is known to investigators and subjects.

Pharmacokinetic samples will be labelled in such a way that treatment allocation cannot be derived by the analytical site.

4.1.5.2 Unblinding and breaking the code

SRD & BA/ FE-Part

As the SRD- part will be conducted 'single-blind', and the BA/ FE-part will be 'open', subjects' treatment assignments will be known to investigators. Therefore, no emergency envelopes will be provided.

4.1.6 Packaging, labelling, and re-supply

SRD-Part & BA/ FE-Part

The investigational medicinal products will be provided by BI. They will be packaged and labelled in accordance with local law and the principles of Good Manufacturing Practice.

For details of packing and the description of the label, refer to the ISF.

The telephone number of the sponsor and the name, address and telephone number of the trial site are provided in the subject information form. The EudraCT number is indicated on the title page of this protocol as well as on the subject information and informed consent forms. Examples of the labels will be available in the ISF.

No re-supply is planned.

4.1.7 Storage conditions

SRD-Part & BA/ FE-Part

Drug supplies will be kept in their original packaging and in a secure limited access storage area in accordance with the recommended (labelled) storage conditions. If necessary, a temperature log must be maintained to make certain that the drug supplies are stored at the correct temperature. If the storage conditions are found to be outside the specified range, the local clinical monitor (as provided in the list of contacts) is to be contacted immediately.

4.1.8 Drug accountability

SRD-Part & BA/ FE-Part

The investigator or designee will receive the investigational drugs from the sponsor when the following requirements are fulfilled:

- Approval of the clinical trial protocol by the IRB/ ethics committee
- Availability of a signed and dated clinical trial contract between the sponsor and the investigational site
- Approval/ notification of the regulatory authority, e.g., competent authority
- Availability of the *curriculum vitae* of the Principal Investigator
- Availability of a signed and dated clinical trial protocol

Only authorised personnel documented in the 'Staff Record' may dispense medication to trial subjects. The trial medication must be administered in the manner specified in the CTP.

The investigator or designee must maintain records of the product's delivery to the trial site, the inventory at the site, the use by each subject, and the disposal of unused products.These records will include dates, quantities, batch/ serial numbers, expiry ('use-by') dates, and the unique code numbers assigned to the investigational medicinal product and trial subjects. The investigator or designee will maintain records that document adequately that the subjects were provided the doses specified by the CTP and reconcile all investigational medicinal products received from the sponsor. At the time of disposal of remaining trial medication, the investigator or designee must verify that no remaining supplies are in the investigator's possession.

All unused trial medication will be disposed of locally by the trial site upon written authorisation of the Clinical Trial Leader. Receipt, usage, and disposal of trial medication must be documented on the appropriate forms. Account must be given for any discrepancies.

4.2 OTHER TREATMENTS, EMERGENCY PROCEDURES, RESTRICTIONS

4.2.1 Other treatments and emergency procedures

There are no special emergency procedures to be followed. No additional treatment is planned. However, if adverse events require treatment, the investigator can authorise symptomatic therapy. In those cases, site staff will follow standard operating procedures and subjects will be treated as necessary and, if required, kept under supervision at the trial site or transferred to a hospital until all results of medical evaluations are acceptable.

4.2.2 Restrictions

4.2.2.1 Restrictions regarding concomitant treatment

In principle, no concomitant therapy is allowed. All concomitant or rescue therapies will be recorded (including time of intake on study days) on the appropriate pages of the CRF.

4.2.2.2 Restrictions on diet and life style

While admitted to the trial site, subjects will be instructed not to consume any foods or drinks other than those provided by the staff. Standardised meals will be served at the times indicated in the Flow Chart. In the SRD-part, a light breakfast will be served about 2h after drug intake. In the BA/ FE-part, no food is allowed for at least 4 h after drug intake.

From 1 h before drug intake until lunch, fluid intake is restricted to the milk served with breakfast (see Table 4.1.4: 5), the water administered with the drug, and an additional 240 mL of water served on Day 1at 2 h and 4 h post-dose (mandatory for all subjects). During the days of urine collection, total fluid intake should be at least 1.5 litres and should not exceed 3.5 litres.

Alcoholic beverages, grapefruits, Seville oranges (sour or bitter oranges) and their juices, and dietary supplements and products containing St. John's wort (*Hypericum perforatum*) are not permitted from 7 days before the first administration of trial medication until after the last pharmacokinetic sample of each study period is collected.

Methylxanthine-containing drinks or foods (such as coffee, tea, cola, energy drinks, or chocolate) are not allowed from 4 h before until 4 h after each administration of trial medication.

Smoking is not allowed during in-house confinement while admitted to the trial site.

Excessive physical activity (such as competitive sport) should be avoided from 7 days before the first administration of trial medication until the EoT examination.

Direct exposure to the sun or exposure to solarium radiation should be avoided during the entire study.

Applicable for Amendment No. 7

In the SRD-part, no food is allowed for at least 4 h after drug intake.

From 1 h before drug intake until lunch, fluid intake is restricted to the water administered with the drug, and an additional 240 mL of water served on Day 1at 2 h and 4 h post-dose (mandatory for all subjects).

4.3 TREATMENT COMPLIANCE

Compliance will be assured by administration of all trial medication in the study centre under supervision of the investigating physician or a designee. The measured plasma concentrations and/ or urinary excretion of trial medication will provide additional confirmation of compliance.

Subjects who are non-compliant (for instance, who do not appear for scheduled visits or violate trial restrictions) may be removed from the trial and the CRF will be completed accordingly (for further procedures, see Section 3.3.4.1).

5. ASSESSMENTS

5.1 ASSESSMENT OF EFFICACY

Not applicable. No efficacy endpoints will be evaluated in this trial.

5.2 ASSESSMENT OF SAFETY

5.2.1 Physical examination

At screening, the medical examination will include demographics, height and body weight, body temperature, smoking and alcohol history (results not mandatory to be entered into CRF or to be reported), relevant medical history and concomitant therapy, review of inclusion and exclusion criteria, review of vital signs (BP, PR, RR, T), 12-lead ECG (including rhythm strip of at least 15 minutes, SRD-Part only), laboratory tests, and a physical/ neurological examination. At the EoT examination, it will include review of vital signs, 12-lead ECG, laboratory tests, and a physical/neurological examination and determination of weight.

5.2.2 Vital signs

Systolic and diastolic blood pressures (BP) as well as pulse rate (PR) or heart rate (heart rate is considered to be equal to pulse rate) will be measured by a blood pressure monitor (e.g., Dinamap V 100 or Dash 4000, [redacted]) at the times indicated in the Flow Chart, after subjects have rested for at least 5 min in a supine position. All recordings should be made using the same type of blood pressure recording instrument on the same arm, if possible.

5.2.3 Safety laboratory parameters

For details on SARS-CoV-2/ COVID-19 specific tests at screening and Day -3, refer to Appendix 10.5.

For the assessment of laboratory parameters, blood and urine samples will be collected by the trial site at the times indicated in the Flow Chart after the subjects have fasted for at least 10 h. For retests, at the discretion of the investigator or designee, overnight fasting is not required.

The parameters that will be determined are listed in Tables 5.2.3: 1 and 5.2.3: 2. Reference ranges will be provided in the ISF, Section 10.

Manual differential white blood cell count or urine sediment examinations will only be performed, if there is an abnormality in the automatic blood cell count or urinalysis, respectively.

Table 5.2.3: 1 Routine laboratory tests

Functional lab group	**BI test name [comments/ abbreviations]**	**A**	**B**
Haematology	Haematocrit	X	X
	Haemoglobin	X	X
	Red Blood Cell Count/ Erythrocytes	X	X
	Reticulocytes, absol.	X	X
	Reticulocytes/ Erythrocyte	X	X
	White Blood Cells/ Leucocytes	X	X
	Platelet Count/ Thrombocytes (quant)	X	X
Automatic WBC differential, relative	Neutrophils/ Leukocytes; Eosinophils/ Leukocytes; Basophils/ Leukocytes; Monocytes/ Leukocytes; Lymphocytes/ Leukocytes	X	X
Automatic WBC differential, absolute	Neutrophil, absol.; Eosinophils, absol.; Basophils, absol.; Monocytes, absol.; Lymphocytes, absol.	X	X
Manual differential WBC (*if automatic differential WBC is abnormal*)	Neut. Poly (segs)/ Leukocytes; Neutrophils Bands/ Leukocytes; Eosinophils/ Leukocytes; Basophils/ Leukocytes; Monocytes/ Leukocytes; Lymphocytes/ Leukocytes	X	X
Coagulation	Activated Partial Thromboplastin Time	X	X
	Prothrombin time – INR (International Normalization Ratio)	X	X
	Fibrinogen	X	X
Enzymes	AST [Aspartate transaminase]/ GOT, SGOT	X	X
	ALT [Alanine transaminase]/ GPT, SGPT	X	X
	Alkaline Phosphatase	X	X
	Gamma-Glutamyl Transferase	X	X
	Glutamate Dehydrogenase (GLDH)	X	X
	Creatine Kinase [CK]	X	X
	Creatine Kinase Isoenzyme MB [only if CK is elevated]	X	X
	Lactic Dehydrogenase	X	X
	Lipase	X	X
	Amylase	X	X
Hormones	Thyroid Stimulating Hormone	X	--

A: parameters to be determined at Visit 1 (screening examination) and Visit 3 (EoT examination)
B: parameters to be determined at Visit 1 and 2 on Days -3 to 4 (for time points refer to Flow Chart)

Table 5.2.3: 1 Routine laboratory tests (cont).

Functional lab group	BI test name [comments/ abbreviations]	A	B
Substrates	Glucose (Plasma)	X	X
	Creatinine	X	X
	Bilirubin, Total	X	X
	Bilirubin, Direct	X	X
	Protein, Total	X	X
	Albumin	X	X
	Albumin (Protein Electrophoresis)	X	--
	Alpha-1-Globulin (Protein Electrophoresis)	X	--
	Alpha-2-Globulin (Protein Electrophoresis)	X	--
	Beta-Globulin (Protein Electrophoresis)	X	--
	Gamma-Globulin (Protein Electrophoresis)	X	--
	C-Reactive Protein (Quant)	X	X
	Uric Acid	X	X
	Cholesterol, total	X	X
	Triglyceride	X	X
Electrolytes	Sodium	X	X
	Potassium	X	X
	Chloride	X	X
	Calcium	X	X
	Phosphate (as Phosphorus, Inorganic)	X	X
Urinalysis (Stix)	Urine Nitrite (qual)	X	X
	Urine Protein (qual)	X	X
	Urine Glucose (qual)	X	X
	Urine Ketone (qual)	X	X
	Urobilinogen (qual)	X	X
	Urine Bilirubin (qual)	X	X
	Urine RBC/ Erythrocytes (qual)	X	X
	Urine WBC/ Leucocytes (qual)	X	X
	Urine pH	X	X
Urine sediment (microscopic examination *if erythrocytes, leukocytes, nitrite or protein are abnormal in urine*)	Only positive findings will be reported (for instance, the presence of sediment bacteria, casts in sediment, squamous epithelial cells, erythrocytes, leukocytes)	X	X

A: parameters to be determined at Visit 1 (screening examination) and Visit 3 (EoT examination)
B: parameters to be determined at Visit 1 and 2 on Days -3 to 4 (for time points refer to Flow Chart)

The tests listed in Table 5.2.3: 2 are exclusionary laboratory tests that may be repeated as required. The results will not be entered in the CRF/ database and will not be reported in the CTR. It is planned to perform these tests at screening only. Drug screening will be performed at Screening and prior to each treatment period (BA/ FE-Part only).

Table 5.2.3: 2 Exclusionary laboratory tests

Functional lab group	Test name
Drug screening (urine)	Amphetamine/MDA
	Barbiturates
	Benzodiazepine
	Cannabis
	Cocaine
	Methadone
	Methamphetamines/MDMA/XTC
	Opiates
	Phencyclidine
	Tricyclic antidepressants
Infectious serology (blood)	Hepatitis B surface antigen (qualitative)
	Hepatitis B core antibody (qualitative)
	Hepatitis C antibodies (qualitative)
	HIV-1 and HIV-2 antibody (qualitative)

To encourage compliance with alcoholic restrictions, a breath alcohol test (e.g., Alcotest® 6510 or 6820, [redacted]) will be performed prior to each treatment period, and may be repeated at any time during the study at the discretion of an investigator or designee. The results will not be included in the CTR.

The laboratory tests listed in Tables 5.2.3: 1 and 5.2.3: 2 will be performed at [redacted] with the exception of drug screening tests which can also be done at the trial site using the test systems from [redacted] or an equivalent test system.

Laboratory data will be transmitted electronically from the laboratory to the trial site.

5.2.4 Electrocardiogram (SRD-Part)

5.2.4.1 12-lead resting ECG

Recording

Twelve-lead resting ECGs (I, II, III, aVR, aVL, aVF, V1 - V6) will be recorded using a computerised electrocardiograph (e.g., MAC VU360, [redacted]) at the time points given in the Flow Chart. Electrode placement will be performed according to the method of Wilson, Goldberger and Einthoven.

To achieve a stable heart rate at rest and to assure high quality recordings, the site personnel will be instructed to assure a relaxed and quiet environment, so that all subjects are at complete rest.

All ECGs will be recorded for a 10 second duration after subjects have rested for at least 5 minutes in a supine position. ECG recording will always precede all other study procedures scheduled for the same time (except for blood drawing from an intravenous cannula that is already in place) to avoid compromising ECG quality.

ECGs will be recorded as single and triplicate ECGs, as indicated in the Flow Chart.

ECGs may be repeated for quality reasons, for instance due to alternating current artefacts, muscle movements, or electrode dislocation. The repeat ECGs are assigned to the respective scheduled time point. Additional (unscheduled) ECGs may be recorded for safety reasons.

Applicable for Amendment No. 7

ECGs will be recorded as single ECGs or as triplicate ECGs (i.e. three single ECGs recorded within 180 sec) as indicated in the Flow Chart.

ECGs may be repeated for quality reasons for instance due to alternating current artefacts, muscle movements, or electrode dislocation. For repetition within triplicate ECGs the time window of 180 sec applies as well. The repeat ECGs are assigned to the respective scheduled time point.

Storing

All ECGs will be stored electronically on the Muse Cardiology Information System ([redacted]).

Data transfer

For time points specified in the Flow Chart, ECGs will be transferred electronically to the [redacted] for evaluation and/ or storage except for ECGs from screening and EoT visits which will not be transferred.

Unscheduled ECGs (for safety reasons) will be transferred to the central ECG lab but will not be included into the statistical analysis of interval lengths.

Data transfer from the central ECG lab to the sponsor is described in the ECG data transfer agreement (see TMF).

If needed, ECGs may be evaluated at a later time.

Evaluation

Trial site

All local ECGs will be evaluated by the investigator or a designee.

For the inclusion or exclusion (see Section 3.3) of a subject and for the assessment of cardiac safety during the study, the QT and QTcF values generated by the computerised ECG system or their manual corrections by the investigators will be used.

Abnormal findings will be reported as AEs (during the trial) or baseline conditions (at screening), if judged clinically relevant by the investigator.

Any ECG abnormalities will be monitored carefully and, if necessary, the subject will be removed from the trial and will receive the appropriate medical treatment.

Applicable for Amendment No. 7

Central ECG lab

Central ECG lab evaluation will be performed for the first of three replicate ECGs at each time point on Days -1 to 4. The remaining second and third ECGs of the triplicate ECGs will be stored for additional analysis, if required.

RR and QT intervals will be determined semi-automatically, whereas PR interval, QRS duration and QRS-axis are measured automatically by a validated GE 12-SL-algorithm or equivalent.

For the statistical analyses, heart rate (HR) and the QT interval corrected for HR (QTc e.g., QTcF and QTcB) will be determined by the sponsor (refer to TSAP for details).
All semi-automatic interval measurements in one subject will be performed on the same lead. The intervals will be measured from four cardiac cycles (beats) in lead II. If lead II shows a flat T wave or is not measurable for any reason, lead V5 will be used, or if that lead is not measurable, then lead I will be used. The lead actually used will be reported in the CTR.

For automatic interval measurements no lead will be provided.

For blinding arrangements refer to Section 4.1.5. No more than two blinded readers will evaluate all ECGs of the study. ECGs from a particular subject should be evaluated by a single reader. For quality assurance and control of the measurements, all ECGs of a subject will be subsequently reviewed by the ECG technician supervisor or his/ her designee to assess the overall variance of the measured intervals and, to detect accidental switching of leads and/ or false subject assignments of the ECGs. After quality control, the fiducial point markings will be reviewed by the cardiologist assigned to the study.

Evaluation of ECGs will comply with the ICH E14 guidance document and supplements [R07-4722, R16-0366] as well as the FDA requirements for annotated digital ECGs [R09-4830].

5.2.4.2 Continuous ECG monitoring

Cardiac rhythm (including heart rate) will be monitored by means of continuous ECG recording using the Telemetry System Central Station V2 with Apex Pro Transmitters, [redacted], for at least 15 min before drug administration (for baseline assessment) and for 4 h following drug administration). This continuous ECG monitoring supports the early detection of adverse events such as clinically relevant bradycardia, tachycardia, or arrhythmia at the trial site. Beyond this clinical evaluation at the trial site, no further data collection or analyses are performed based on continuous ECG monitoring.

ECG data from continuous ECG recording will not be transferred to the clinical trial database. Abnormal findings during continuous ECG recording will be recorded as AEs, if judged clinically relevant by the Investigator.

5.2.5 Electrocardiogram (BA/ FE-Part)

Twelve-lead resting ECGs (I, II, III, aVR, aVL, aVF, V1 - V6) will be recorded using a computerised electrocardiograph (e.g., MAC VU360, [redacted]) at the time points given in the Flow Chart. Electrode placement will be performed according to the method of Wilson, Goldberger and Einthoven.

To achieve a stable heart rate at rest and to assure high quality recordings, the site personnel will be instructed to assure a relaxed and quiet environment, so that all subjects are at complete rest.

All ECGs will be recorded for a 10 second duration after subjects have rested for at least 5 minutes in a supine position. ECG recording will always precede all other study procedures scheduled for the same time (except for blood drawing from an intravenous cannula that is already in place) to avoid compromising ECG quality.

ECGs will be recorded as single and triplicate ECGs, as indicated in the ECGs will be recorded as single ECGs as indicated in the Flow Chart.

ECGs may be repeated for quality reasons, for instance due to alternating current artefacts, muscle movements, or electrode dislocation. The repeat ECGs are assigned to the respective scheduled time point. Additional (unscheduled) ECGs may be recorded for safety reasons.

Storing

All ECGs will be stored electronically on the Muse Cardiology Information System ([redacted]).

Data transfer

For time points specified in the Flow Chart, ECGs will be transferred electronically to the [redacted] for evaluation and/ or storage except for ECGs from screening and EoT visits which will not be transferred.

Unscheduled ECGs (for safety reasons) will be transferred to the central ECG lab but will not be included into the statistical analysis of interval lengths.

Data transfer from the central ECG lab to the sponsor is described in the ECG data transfer agreement (see TMF).

If needed, ECGs may be evaluated at a later time.

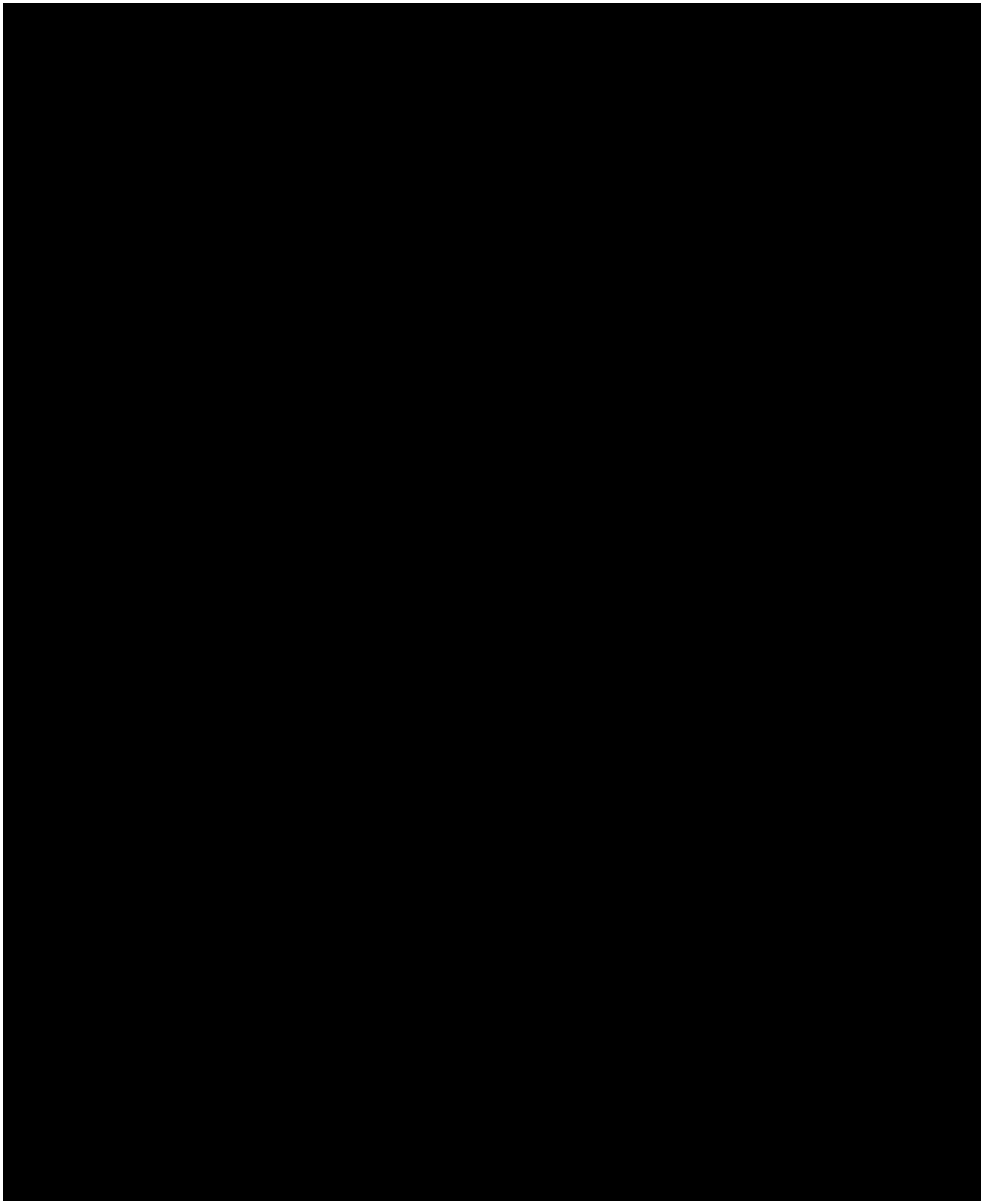
Trial site

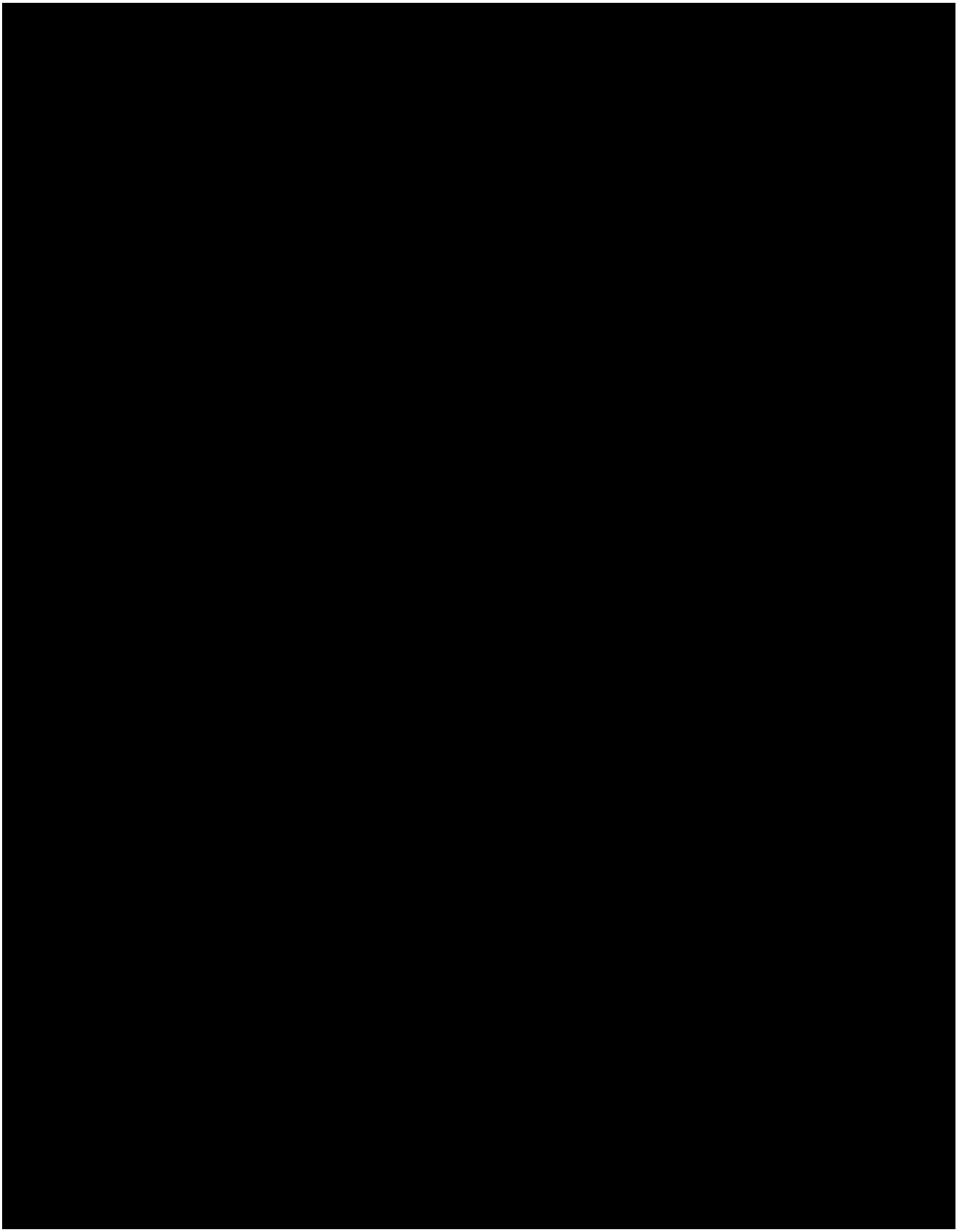
All local ECGs will be evaluated by the investigator or a designee.

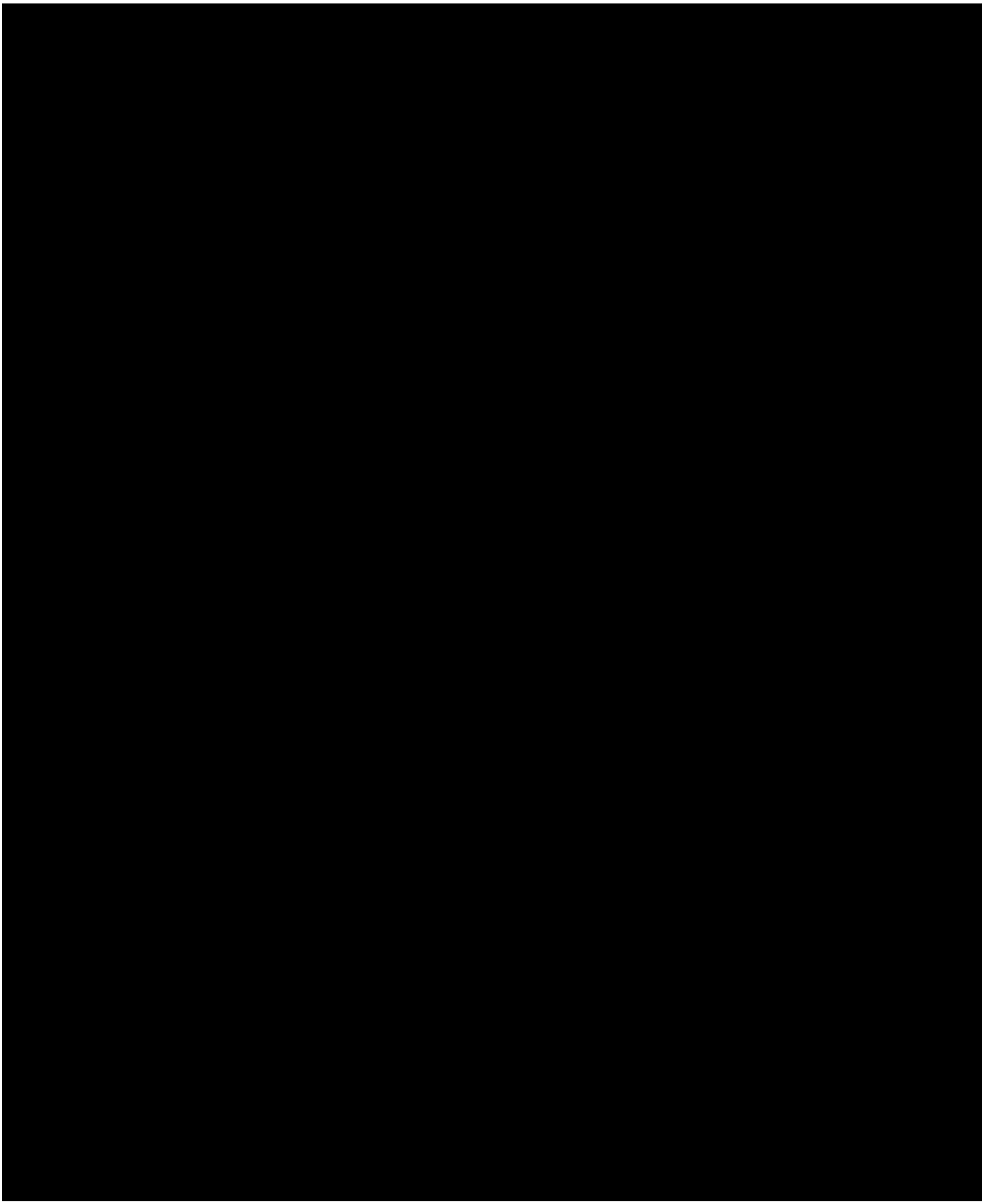
For the inclusion or exclusion (see Section 3.3) of a subject and for the assessment of cardiac safety during the study, the QT and QTcF values generated by the computerised ECG system or their manual corrections by the investigators will be used.

Abnormal findings will be reported as AEs (during the trial) or baseline conditions (at screening), if judged clinically relevant by the investigator.

Any ECG abnormalities will be monitored carefully and, if necessary, the subject will be removed from the trial and will receive the appropriate medical treatment.







5.2.7 Assessment of adverse events

For the documentation of adverse events, related to SARS-CoV-2/COVID-19, refer to Appendix 10.5.

5.2.7.1 Definitions of adverse events

5.2.7.1.1 Adverse event

An adverse event (AE) is defined as any untoward medical occurrence in a patient or clinical investigation subject administered a medicinal product and which does not necessarily have to have a causal relationship with this treatment.

An AE can therefore be any unfavourable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

The following should also be recorded as an AE in the CRF and BI SAE form (if applicable):

- Worsening of the underlying disease or of other pre-existing conditions
- Changes in vital signs, ECG, physical examination, and laboratory test results, if they are judged clinically relevant by the investigator

If such abnormalities already pre-exist prior to trial inclusion, they will be considered as baseline conditions and should be collected in the eCRF only.

5.2.7.1.2 Serious adverse event

A serious adverse event (SAE) is defined as any AE which fulfils at least one of the following criteria:

- Results in death
- Is life-threatening, which refers to an event in which the patient was at risk of death at the time of the event; it does not refer to an event that hypothetically might have caused death if more severe

- Requires inpatient hospitalisation
- Requires prolongation of existing hospitalisation
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly/birth defect
- Is deemed serious for any other reason if it is an important medical event when based upon appropriate medical judgment which may jeopardise the patient and may require medical or surgical intervention to prevent one of the other outcomes listed in the above definitions. Examples of such events are intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalisation or development of dependency or abuse

5.2.7.1.3 AEs considered 'Always Serious'

Cancers of new histology and exacerbations of existing cancer must be classified as a serious event regardless of the time since discontinuation of the trial medication and must be reported as described in Section 5.2.6.2, subsections 'AE Collection' and '**AE reporting to sponsor and timelines'**.

In accordance with the European Medicines Agency initiative on Important Medical Events, Boehringer Ingelheim has set up a list of further AEs, which, by their nature, can always be considered to be 'serious' even though they may not have met the criteria of an SAE as defined above.

The latest list of 'Always Serious AEs' can be found in the eDC system, an electronic data capture system which allows the entry of trial data at the trial site. These events should always be reported as SAEs as described above.

5.2.7.1.4 Adverse events of special interest

The term adverse events of special interest (AESI) relates to any specific AE that has been identified at the project level as being of particular concern for prospective safety monitoring and safety assessment within this trial, e.g., the potential for AEs based on knowledge from other compounds in the same class. AESIs need to be reported to the sponsor's Pharmacovigilance Department within the same timeframe that applies to SAEs; see Section 5.2.7.2.2.

The following are considered as AESIs:

- Hepatic injury
  A hepatic injury is defined by the following alterations of hepatic laboratory parameters:
  - An elevation of AST (aspartate transaminase) and/or ALT (alanine transaminase) ≥3-fold ULN combined with an elevation of total bilirubin ≥2-fold ULN measured in the same blood sample, or
  - Aminotransferase (ALT, and/or AST) elevations ≥10 fold ULN

These lab findings constitute a hepatic injury alert and the subjects showing these lab abnormalities need to be followed up according to the 'DILI checklist' provided in the ISF. In case of clinical symptoms of hepatic injury (icterus, unexplained encephalopathy, unexplained coagulopathy, right upper quadrant abdominal pain, etc.) without lab results (ALT, AST, total bilirubin) available, the Investigator should make sure that these parameters are analysed, if necessary in an unscheduled blood test. Should the results meet the criteria of hepatic injury alert, the procedures described in the DILI checklist should be followed.

No further AESIs have been defined for this trial.

5.2.7.1.5 Intensity (severity) of AEs

The intensity (severity) of the AE should be judged based on the following:

Mild:	Awareness of sign(s) or symptom(s) that is/are easily tolerated
Moderate:	Sufficient discomfort to cause interference with usual activity
Severe:	Incapacitating or causing inability to work or to perform usual activities

5.2.7.1.6 Causal relationship of AEs

Medical judgment should be used to determine the relationship, considering all relevant factors, including pattern of reaction, temporal relationship, de-challenge or re-challenge, confounding factors such as concomitant medication, concomitant diseases and relevant history.

Arguments that may suggest that there is a reasonable possibility of a causal relationship could be:

- The event is consistent with the known pharmacology of the drug
- The event is known to be caused by or attributed to the drug class
- A plausible time to onset of the event relative to the time of drug exposure
- Evidence that the event is reproducible when the drug is re-introduced
- No medically sound alternative aetiologies that could explain the event (e.g., pre-existing or concomitant diseases, or co-medications)
- The event is typically drug-related and infrequent in the general population not exposed to drugs (e.g., Stevens-Johnson syndrome)
- An indication of dose-response (i.e. greater effect size if the dose is increased, smaller effect size if dose is reduced)

Arguments that may suggest that there is no reasonable possibility of a causal relationship could be:

- No plausible time to onset of the event relative to the time of drug exposure is evident (e.g., pre-treatment cases, diagnosis of cancer or chronic disease within days / weeks of drug administration; an allergic reaction weeks after discontinuation of the drug concerned)

- Continuation of the event despite the withdrawal of the medication, taking into account the pharmacological properties of the compound (e.g., after 5 half-lives). Of note, this criterion may not be applicable to events whose time course is prolonged despite removing the original trigger
- Additional arguments amongst those stated before, like alternative explanation (e.g., situations where other drugs or underlying diseases appear to provide a more likely explanation for the observed event than the drug concerned)
- Disappearance of the event even though the trial drug treatment continues or remains unchanged

5.2.7.2 Adverse event collection and reporting

5.2.7.2.1 AE collection

Upon enrolment into a trial, the subject's baseline condition is assessed (for instance, by documentation of medical history/ concomitant diagnoses), and relevant changes from baseline are noted subsequently.

Subjects will be required to report spontaneously any AEs as well as the time of onset, end time, and intensity of these events. In addition, each subject will be regularly assessed by the medical staff throughout the clinical trial and whenever the investigator deems necessary. As a minimum, subjects will be questioned for AEs (and concomitant therapies) at the time points indicated in the Flow Chart. Assessment will be made using non-specific questions such as 'How do you feel?'. Specific questions will be asked wherever necessary in order to more precisely describe an AE.

A carefully written record of all AEs shall be kept by the investigator in charge of the trial. Records of AEs shall include data on the time of onset, end time, intensity of the event, and any treatment or action required for the event and its outcome.

The following must be collected and documented on the appropriate CRF(s) by the investigator:

- From signing the informed consent onwards until an individual subject's end of trial:
    - All AEs (serious and non-serious) and all AESIs
    - The only exception to this rule are AEs (serious and non-serious) and AESIs in Phase I trials in healthy volunteers, when subjects discontinue from the trial due to screening failures prior to administration of any trial medication. In these cases, the subjects' data must be collected at trial site but will not be entered in the CRF or trial database and will not be reported in the CTR.
- After the individual subject's end of trial:
    - The investigator does not need to actively monitor the subject for new AEs but should only report any occurrence of cancer and related SAEs and related AESIs of which the investigator may become aware of by any means of communication, e.g., phone call. Those AEs should, however, not be reported in the CRF

5.2.7.2.2 AE reporting to the sponsor and timelines

The Investigator must report SAEs, AESIs, and non-serious AEs which are relevant for the reported SAE or AESI, on the BI SAE form via fax immediately (within 24 hours) to the sponsor's unique entry point (country specific contact details will be provided in the ISF). The same timeline applies, if follow-up information becomes available. At specific occasions, the Investigator could inform the sponsor upfront via telephone. This does not replace the requirement to complete and fax the BI SAE form.

With receipt of any further information to these events, a follow-up SAE form has to be provided. For follow-up information, the same rules and timeline apply as for initial information.

5.2.7.2.3 Information required

All (S)AEs, including those persisting after the individual subject's end of trial, must be followed up until they have resolved, have been assessed as 'chronic' or 'stable', or no further information can be obtained.

5.2.7.2.4 Pregnancy

Once the male subject has been enrolled in the clinical trial and has taken trial medication, and, if a partner of the male trial participant becomes pregnant, the investigator must report any drug exposure during pregnancy in a partner of the male trial participant immediately (within 24 hours) by means of Part A of the Pregnancy Monitoring Form to the sponsor's unique entry point, after a written consent of the pregnant partner was obtained.

The outcome of the pregnancy, associated with the drug exposure during pregnancy, must be followed up and reported to the sponsor's unique entry point on the Pregnancy Monitoring Form for Clinical Trials (Part B).

The ISF will contain the Pregnancy Monitoring Form for Clinical Trials (Part A and Part B) as well as non-trial specific information and consent form for the pregnant partner.

As pregnancy itself is not to be reported as an AE, in the absence of an accompanying SAE and/ or AESI, only the Pregnancy Monitoring Form for Clinical Trials and not the SAE form is to be completed. If there is an SAE and/ or AESI associated with the pregnancy, an SAE form must be completed in addition.

5.3 DRUG CONCENTRATION MEASUREMENTS AND PHARMACOKINETICS

5.3.1 Assessment of pharmacokinetics

SRD-Part & BA/ FE-Part

For the assessment of pharmacokinetics, blood and urine samples will be collected at time points/ time intervals indicated in the Flow Chart. Date and clock times of drug

administration and pharmacokinetic sampling will be recorded in the CRFs. The actual sampling times will be used for determination of pharmacokinetic parameters.

Pharmacokinetic sampling times and periods may be adapted during the trial based on information obtained during trial conduct (e.g., as a result of preliminary pharmacokinetic data), including addition of samples and visits, as long as the total blood volume taken per subject does not exceed 500 mL. Such changes would be implemented via non-substantial CTP-amendments.

5.3.2 Methods of sample collection

5.3.2.1 Blood sampling for pharmacokinetic analysis (SRD-Part & BA/ FE-Part)

For quantification of BI 1569912 concentrations in plasma, 3.0 mL of blood will be drawn from an antecubital or forearm vein into an K_2-EDTA (dipotassium ethylenediaminetetraacetic acid)-anticoagulant blood drawing tube at the times indicated in the Flow Chart. Blood will be withdrawn by means of either an indwelling venous catheter or by venepuncture with a metal needle.

EDTA-anticoagulated blood samples will be centrifuged for approximately 10 minutes at approximately 2000 g to 4000 g and at 4 to 8 °C. Two plasma aliquots will be obtained and stored in polypropylene tubes. The first aliquot should contain at least 0.6 mL of plasma. The process from blood collection until transfer of plasma aliquots into the freezer should be completed in less than 60 min with interim storage of blood samples and aliquots in ice water or on ice. The time at which each aliquot was placed in the freezer will be documented. Until transfer on dry ice to the analytical laboratory, the aliquots will be stored upright at approximately -20 °C or below at the trial site. The second aliquot will be transferred to the analytical laboratory after the bioanalyst has acknowledged safe arrival of the first one. At the analytical laboratory, plasma samples will be stored at approximately -20 °C or below until analysis.

At a minimum, sample tube labels should list BI trial number, subject number, visit, and planned sampling time. Further information such as matrix, aliquot number and analyte may also be provided.

Plasma samples, dedicated to pharmacokinetic analysis, are transferred to:

After completion of the trial, plasma samples may be used for further methodological investigations (e.g., for stability testing or assessment of metabolites). However, only data related to the analyte and/ or its metabolite(s), including anti-drug antibodies (if applicable) will be generated by these additional investigations. The study samples will be discarded after completion of the additional investigations, but not later than 5 years after the CTR is archived.

5.3.2.2 Blood sampling for metabolism analysis (SRD-Part only)

Additional K_2-EDTA plasma samples for the optional identification of drug metabolites will be investigated in the 5 mg-dose group. Based on the knowledge gained during the trial conduct, e.g., from preliminary pharmacokinetic results, the dose group may be modified to a different one. The change will be implemented via a non-substantial CTP amendment.

The blood samples will be drawn at the same time points as pharmacokinetic samples on Days 1 to 4 (see Flow Chart). At each of these times, 3.0 mL blood will be needed for metabolite analysis. Blood samples will be processed in the same way as the phamacokinetic samples (see Section 5.3.2.1). However, the plasma obtained (approximately 1 mL) will be transferred into a single polypropylene tube. Samples will be stored at approximately -20 °C or below until transfer to the metabolism laboratory.

At a minimum, sample tube labels should list BI trial number, subject number, visit, planned sampling time and 'MetID'.

Plasma samples, dedicated to metabolism investigation, are transferred to:

[redacted]

Only data related to the parent compound and its metabolites will be acquired. Evaluation of drug metabolism will be reported separately and will not be included in the CTR. The study samples will be discarded after completion of the experiments but not later than 5 years after the CTR has been archived.

5.3.2.3 Urine sampling for pharmacokinetic analysis

A blank urine sample will be collected before the administration of trial medication (within 3h before drug dosing) and two 0.5 mL aliquots will be retained to check for analytical interference by concomitant or rescue medication.

All urine voided during the sampling intervals, indicated in the Flow Chart, will be collected in 2 L polyethylene (PE) containers and stored in the refrigerator during the collection interval. Subjects are told to empty their bladders at the end of each sampling interval. In order to facilitate urine sampling, subjects will be advised to drink at least 100 mL water before the end of each urine sampling interval.

The urine weight/ volume for each collection interval will be documented. However, no correction for the specific gravity of urine is done; i.e., 1 L is defined to be equal to 1 kg. Two 0.5 mL aliquots will be stored in polypropylene (PP) tubes for bioanalytical measurements. If more than one collection container is used in an interval, the content of all containers are to be mixed before aliquots are prepared. Mixing should be done by transferring the entire content of all collection containers into a single polyethylene (PE)/ PP or glass container, and by stirring the mixed fractions for about 1 min (manually or using a stir bar or other stirring device made of PE, PP, Teflon, or glass).

At a minimum, sample tube labels should list BI trial number, subject number, visit, and planned collection time. Further information, such as matrix and analyte may also be provided.

Until transfer on dry ice to the analytical laboratory, urine samples will be stored at approximately -20 °C or below at the trial site. The second aliquot will be transferred after the bioanalyst has acknowledged safe arrival of the first one. At the analytical laboratory, urine samples will be stored at approximately -20 °C or below until analysis.

Urine samples, dedicated to pharmacokinetic analysis, are transferred to:

[redacted]

After completion of the trial, urine samples may be used for further methodological investigations (e.g., for stability testing or assessment of metabolites). However, only data related to the analyte and/ or its metabolite(s) will be generated by these additional investigations. The study samples will be discarded after completion of the additional investigations but not later than 5 years after the CTR has been archived.

5.3.2.4 Additional blood sample for stability-testing (SRD-Part only)

In order to assess the stability of the analyte in whole blood, one additional blood sample will be obtained from all subjects of the 2 mg-dose group. Based on the knowledge gained during the trial conduct, e.g., from preliminary pharmacokinetic results, the chosen timing or dose group may be changed to a different one. The change will be implemented via a non-substantial CTP amendment.

Approximately 4 mL of blood will be drawn from an antecubital or forearm vein into two 2.0 mL K_2-EDTA-blood drawing tubes at the time point of 3 h post-dose (immediately after the drawing of a regular blood pharmacokinetic sample which means that no additional venous puncture will be necessary).

From each K_2-EDTA tube, aliquots will be generated:

- One aliquot (‘stability reference’) will be centrifuged within 30 min after collection. Centrifugation will last for approximately 10 min (at approximately 2000 g to 4000 g and 4 to 8 °C). Then plasma will be separated and transferred into a freezer.
- The second aliquot (‘stability test’) will be stored for about 4 h at room temperature and ambient light conditions (storage time must be documented) and will then be centrifuged and stored like the first aliquot.

At a minimum, aliquots should be labelled with BI trial number, administered drug, subject number, planned sampling time, and whether the sample is the ‘stability reference’ or ‘stability test’.

Until transfer to the analytical laboratory, both aliquots will be stored at approximately -20 °C or below at the trial site. They will be provided to the responsible

bioanalyst together with the information about sample handling (i.e., storage time of stability test sample at room temperature). After receipt, the aliquots will be stored at the bioanalytical laboratory at approximately -20 °C or below until analysis.

Plasma samples dedicated to stability testing are transferred to:

[redacted]

The results of the analysis of these samples will not be reported in the CTR but will be used for bioanalytical assay validation and therefore included in the corresponding method validation report. The remaining sample volume will be discarded at latest upon completion of the method validation report.

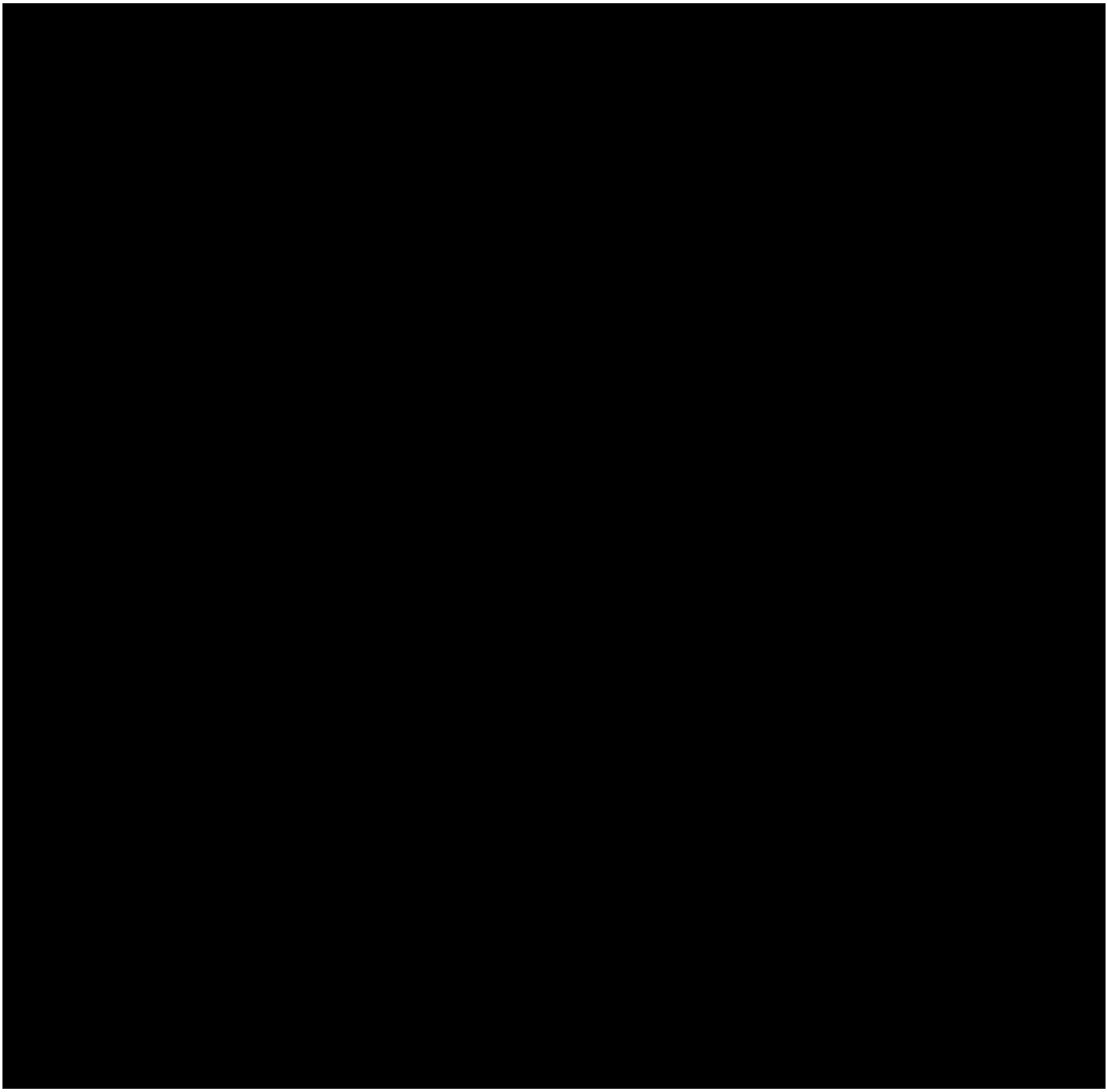
[redacted]

5.3.4 Pharmacokinetic - pharmacodynamic relationship

If data allows, an exploratory and descriptive PK/PD analysis of plasma concentrations/ PK parameters of BI 1569912 will be plotted against pharmacodynamics readouts, such as qEEG parameters.

5.4 ASSESSMENT OF BIOMARKERS (SRD-PART& BA/ FE-PART)

[redacted]



5.4.2 Quantitative EEG, event-related potentials

EEG is a non-invasive method to measure the electrical activity of large, synchronously firing, populations of neurons in the brain with electrodes placed on the scalp. EEG read-outs will be assessed as potential markers for indirect target engagement and physiological response. The following EEG parameters will be measured prior to drug administration and at selected time points after drug administration (see Flow Chart): Quantitative EEG (qEEG) and Event-Related Potential (ERP). The timing of qEEG/ ERPs may be adjusted in consultation with the Principal Investigator and sponsor, based on emerging pharmacokinetics or other data.

When qEEG/ ERP is scheduled for the same time points as blood draws, blood draws will take priority, and the qEEG/ ERP will be obtained shortly thereafter.

qEEG and ERP testing will occur in an environmentally controlled room. Tests will be completed with subjects seated comfortably in front of a computer monitor. EEG electrodes will be affixed to the scalp according to the International 10-20 electrode placement system. qEEG and ERP records will be collected using a Comet AS40 Amplifier from [redacted]. The COMET AS40 has CE Class IIa medical certification and FDA approval. The sampling rate will be fixed at 400 Hz within a standard range of 0.1-100 Hz. A 50 Hz notch filter will be applied. Tests will occur in series, with qEEG preceding ERP testing, if applicable.

Prior to start of the test, some time should be planned to hook up EEG electrodes. After the test, 5 to 10 min are required to unhook electrodes.

5.4.2.1 Quantitative EEG (qEEG)

Eyes Closed Segment (5 Minutes)

A resting eyes closed recording segment is necessary to assess variations in vigilance over time and acquire pertinent EEG information such as posterior dominant rhythm, standard EEG spectral bands (Delta, Alpha, Beta, etc.), and a number of derived spectral measures (Alpha Slow-Wave Index, Theta-Beta Ratio, etc.). Utmost care should be taken by the technologist to minimize subject eye fluttering, muscle tension, and monitor for drowsiness. A minimum of 90-120 seconds artefact-free signal is desired.

Eyes Open Segment (5 Minutes)

Eyes open recording segment is necessary as a counterpart to eyes closed segment. Care should be taken by the technologist to minimize subject eye movement, excessive blink frequency, eye fluttering, or muscle tension. Short external interventions for vigilance stimulation (talking to subject, tapping a pen etc.) are permissible, if a decrease in vigilance is observed, for instance in case of intrusion of slow-wave activity in the EEG. A minimum of 90-120 seconds artefact-free signal is desired.

5.4.2.2 Event-Related Potential (ERP)

In the ERP task, one standard tone (standard tone, probability 0.82, 1000 Hz, 50 ms duration, 5 ms rise/ fall time, 85 dB SPL) will be presented binaurally through headphones with a constant SOA (stimulus onset asynchrony) of 0.5 s, randomly interrupted by three types of deviant tone bursts (deviant tone, probability 0.06 each): a duration deviant (1000 Hz, 125 ms duration, 5 ms rise/ fall time, 85 dB SPL), a pitch deviant (1500 Hz, 50 ms duration, 5 ms rise/ fall time, 85 dB SPL) and an double deviant (1500 Hz, 125 ms duration, 5 ms rise/ fall time, 85 dB SPL). A total of 1800 tones will be presented in three blocks separated by 20-second rest periods. This will produce 108 deviant stimuli of each type and a total of 1476 standards.

All stimuli will be presented binaurally over headphones. Subjects will be seated comfortably facing a monitor and viewing a slowly changing slide show of nature scenes (landscapes, animals). Subjects will be instructed to ignore the tones and silently count the number of

different nature scenes shown. No responses are required during the ERP task. However, subjects will be asked to report the number of slide show scenes they count and this number will be recorded on the transmittal form. No judgement of accuracy will be made concerning the scene count, as the task serves merely to keep the subjects alert and attentive. An additional electrode on the nose (Nz) is required for accurate identification of the ERP topography. The total task duration will be 12.67 min (3 * 4 min + 2 * 0.33 min).

5.5 BIOBANKING

Not applicable.

5.6 OTHER ASSESSMENTS

5.6.1 Pharmacogenomic evaluation

Pharmacogenomic investigations explore the role of genetic variations in determining an individual's response to drugs. Therefore, one mandatory blood sample of at most 3 mL for pharmacogenomic testing will be taken from each subject. In case of unexplainable variability in pharmacokinetic or pharmacodynamic parameters, DNA might be extracted from these samples and used for exploratory analysis of variants of genes related to these parameters, such as genes involved in Absorption, Distribution, Metabolism and Excretion (ADME). It is not intended to include these data in the final report. However, the data may be part of the report, if necessary. All remaining samples will be destroyed no later than three years after the end of the trial.

5.6.1.1 Methods and timing of sample collection

One blood sample of at most 3 mL will be taken from an arm vein into a PAXgene Blood DNA tube at Visit 2 (see Flow Chart). If not feasible at Visit 2, the sample may also be drawn at any later visit. Directly after blood collection, gently invert the PAXgene Blood DNA tube at least 8 times and then store the blood sample at a temperature of approximately -20°C or below. Once frozen, thawing of the samples should be avoided. Frozen blood samples should be shipped on dry ice to:

5.7 APPROPRIATENESS OF MEASUREMENTS

All measurements performed during this trial are standard measurements and will be performed in order to monitor subjects' safety and to determine pharmacokinetic and pharmacodynamic parameters in an appropriate way. The scheduled measurements will allow monitoring of changes in vital signs, standard laboratory values, and ECG parameters that might occur as a result of administration of trial medication. The safety assessments are standard, are accepted for evaluation of safety and tolerability of an orally administered drug, and are widely used in clinical trials.

Since, in pre-clinical studies, seizures/ convulsions were detected in dogs at higher exposure levels, a routine EEG is included in order to detect suspect EEG signals as early as possible. The pharmacokinetic parameters and measurements outlined in Section 5.4 are generally used assessments of drug exposure. The biomarkers and pharmacodynamic parameters and measurements outlined in Section 5.4 are of exploratory nature.

6. INVESTIGATIONAL PLAN

6.1 VISIT SCHEDULE

Exact times of measurements outside the permitted time windows will be documented. The acceptable time windows for screening and the EoT examination are provided in the Flow Chart.

Study measurements and assessments scheduled to occur 'before' trial medication administration on Day 1 are to be performed and completed within a 3 hour-period prior to the trial drug administration (including blank values for pharmacokinetics and biomarkers).

The acceptable deviation from the scheduled time for vital signs, ECG, and laboratory tests will be ± 5 minutes for the first 4 h, 10 minutes thereafter up to 24 h after trial drug administration and ± 30 minutes thereafter.

The tolerance for drug administration will be ± 1 minute on Days 1.

If several activities are scheduled at the same time point in the Flow Chart, PK sample collection has priority, i.e. that venepuncture will be performed at the exact time point and only exceptionally the order of assessments will be changed. For planned individual plasma concentration sampling times and urine collection intervals, refer to the Flow Chart. While these nominal times should be adhered to as closely as possible, the actual sampling times will be recorded and used for the determination of pharmacokinetic parameters.

If a subject misses an appointment, it will be rescheduled, if possible. The relevance of measurements outside the permitted time windows will be assessed no later than at the Report Planning Meeting.

Applicable for Amendment No. 7
If several activities are scheduled at the same time point in the Flow Chart, ECG should be the first and meal the last activity. Furthermore, if several measurements including venipuncture are scheduled for the same time, venipuncture should be the last of the measurements due to its inconvenience to the subject and possible influence on physiological parameters. However, the PK sample collection should have priority in the sense that it should be performed at the exact planned clock time which means that other measurements scheduled at the same time point, e.g., ECG, have to be taken a bit earlier.

6.2 DETAILS OF TRIAL PROCEDURES AT SELECTED VISITS

6.2.1 Screening periods

After having been informed about the trial, all subjects will provide written informed consent in accordance with GCP and local legislation prior to enrolment in the study.

For information regarding laboratory tests (including drug and virus screening), ECG, vital signs, and physical examination, refer to Sections 5.2.3 to 5.2.5.

For details on SARS-CoV-2/ COVID-19 specific measures, refer to Appendix 10.5.

6.2.2 Treatment period

SRD-Part

Each subject will receive one dose of trial medication (BI 1569912 or placebo) at Visit 2.

Trial medication will be taken orally by each subject under direct supervision of the investigator or his/ her designee. Details on treatments and procedures of administration are described in Section 4.1.4.

Study participants will be admitted to the trial site in the evening of Day -2 and kept under close medical surveillance for at least 72 h following drug administration.

BA/ FE-Part

Each subject is expected to participate in 3 treatment periods (Days 1 in each period). At least 5 days will separate drug administrations between treatment periods.

On Day -2 of each treatment period, study participants will be admitted to the trial site and kept under close medical surveillance for at least 72 h following drug administration. Subjects will then be allowed to leave the trial site after formal assessment and confirmation of their fitness.

SRD-Part & BA/ FE-Part

For details on time points and procedures for collection of plasma and urine samples for pharmacokinetic analysis, refer to Flow Chart and Section 5.3.2.

The safety measurements performed during the treatment period are specified in Section 5.3 and in the Flow Chart. For details on times of all other trial procedures, refer to the Flow Chart. AEs and concomitant therapy will be assessed continuously from screening until the EoT examination.

6.2.3 Follow-up period and trial completion

For AE assessment, laboratory tests, recording of ECG and vital signs, and physical examination during the follow-up period, see Sections 5.2.2 to 5.2.5.

Subjects who discontinue treatment before the end of the planned treatment period should undergo the end-of-trial (EoT) Visit.

All abnormal values (including laboratory parameters) that are assessed as clinically relevant by the investigator will be monitored using the appropriate tests until a return to a medically acceptable level is achieved. (S)AEs persisting after a subject's EoT Visit must be followed until they have resolved, have been sufficiently characterised, or no further information can be obtained.

7. STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

7.1 STATISTICAL DESIGN – MODEL

SRD-Part

The main objectives of this trial will be assessed by calculating descriptive statistics for safety as well as for pharmacokinetic and pharmacodynamic parameters, which will be compared between the treatment groups. Further analyses of these endpoints comprise the power model for assessment of dose proportionality.

BA/ FE-Part

The main objectives of this part of the trial are to investigate the relative bioavailability of BI 1569912 given as tablet under fed condition (Test 1, T1) compared with BI 1569912 given as tablet under fasted condition (Test 2, T2) as well as the relative bioavailability of BI 1569912 given as tablet under fasted condition (T2) compared with BI 1569912 given as oral solution under fasted condition (Reference, R) on the basis of the primary and secondary pharmacokinetic endpoints, as listed in Section 2.1.2 and Section 2.1.3.
This part of the trial is designed to allow intra-subject comparisons and will be evaluated statistically by use of a linear model for logarithmically transformed pharmacokinetic endpoints.

7.2 NULL AND ALTERNATIVE HYPOTHESES

SRD-Part

It is not planned to test any statistical hypotheses in this study.

Any confidence intervals computed are to be interpreted in the perspective of the exploratory character of the study; i.e., confidence intervals are considered as interval estimates for effects.

BA/ FE-Part

The relative bioavailability of BI 1569912 will be estimated by the ratios of the geometric means (T1/ T2, T2/ R), and their corresponding 2-sided 90% confidence intervals (CIs) will be provided. This method corresponds to the two one-sided t-test procedure, each at the 5% significance level. Since the main focus is on estimation and not testing, a formal hypothesis test and associated acceptance range is not specified.

7.3 PLANNED ANALYSES

Analysis sets

For both parts, statistical analyses will be based on the following analysis sets:

- Treated set (TS): The treated set includes all subjects who were randomized and treated with at least one dose of study drug. The treatment assignment will be determined based on the first treatment the subjects received. 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 primary or secondary pharmacokinetic endpoint that was not excluded due to a protocol deviation relevant to the evaluation of pharmacokinetics or due to pharmacokinetic non-evaluability (as specified in the following subsection 'Pharmacokinetics'). Thus, a subject will be included in the PKS, even if he contributes only one pharmacokinetic parameter value for one period to the statistical assessment. Descriptive and model based analyses of pharmacokinetic parameters will be based on the PKS.

Adherence to the protocol will be assessed by the trial team. Important protocol deviation (iPD) categories will be specified in the Integrated Quality and Risk Management Plan, iPDs will be identified no later than in the Report Planning Meeting, and the iPD categories will be updated as needed.

Pharmacokinetics

The pharmacokinetic parameters listed in Section 2.1 for drug BI 1569912 will be calculated according to relevant BI internal procedures.
.

Plasma and urine (in the SRD part of the trial) concentration data and parameters of a subject will be included in the statistical pharmacokinetic analyses, if they are not flagged for exclusion due to a protocol deviation relevant to the evaluation of pharmacokinetics (to be decided no later than in the Report Planning Meeting) or due to pharmacokinetic non-evaluability (as revealed during data analysis, based on the criteria specified below). Exclusion of a subject's data will be documented in the CTR.

Relevant protocol violations may be

- Incorrect trial medication taken, i.e. the subject received at least one dose of trial medication the subject was not assigned to
- Incorrect dose of trial medication taken
- Use of restricted medications

SRD-Part

Plasma and urine 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 t_{max} of the respective treatment (median t_{max} is to be determined excluding the subjects experiencing emesis)
- Missing samples/concentration data at important phases of pharmacokinetic disposition curve

BA/ FE-Part

Plasma and urine 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 t_{max} of the respective treatment (Median t_{max} is to be determined excluding the subjects experiencing emesis),
- The subject experiences emesis at any time during the labelled dosing interval,
- A predose concentration is >5% C_{max} value of that subject,
- Missing samples/ concentration data at important phases of pharmacokinetic disposition curve.

SRD-Part & BA/ FE-Part

Plasma/ urine concentration data and parameters of a subject which is flagged for exclusion will be reported with its individual values but will not be included in the statistical analyses.

Only concentration values within the validated concentration range and actual sampling times will be used for the calculation of pharmacokinetic parameters. Concentrations used in the pharmacokinetic calculations will be in the same format as in the bioanalytical report (that is to the same number of decimal places provided in the bioanalytical report).

7.3.1 Primary endpoint analyses

SRD-Part

The primary endpoint as specified in Section 2.1.2 will be derived according to BI standards. The analysis will be based on the treated set (TS) and will be descriptive in nature.

BA/ FE-Part

Primary analysis

The primary endpoints (refer to Section 2.1.2) will be calculated according to the relevant internal BI procedures..

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, phamacokinetic 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, … , 6

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, 3
τ_k = the k_{th} treatment effect, k = 1, 2, 3
eijkm = the random error associated with the mth subject in sequence i who received treatment k in period j.

where sim ~ N(0,σB2) i.i.d., eijkm ~ N(0,σW2) i.i.d. and sim, eijkm are independent random variables.

Point estimates for the ratios of the geometric means (T1/ T2 and T2/ R) for the primary endpoints (see Section 2.1) 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.

[redacted]

In addition to the model based approach all parameters will be calculated and analysed descriptively.

7.3.2 Secondary endpoint analyses

SRD-Part

The secondary endpoints (refer to Section 2.1.3) will be analysed descriptively. Analyses will be performed for the parent drug and for metabolites.

BA/ FE-Part

The secondary endpoints (refer to Section 2.1.3) will be calculated according to the relevant internal BI procedures and will be assessed statistically using the same methods as described for the primary endpoints.
Further exploratory analyses

SRD-Part (assessment of dose proportionality):

Dose proportionality will be explored via graphical checks and if applicable via the power model stated below. The analysis will be performed for the pharmacokinetic endpoints $AUC_{0-\infty}$ and C_{max} specified in Section 2.1.3.

The power model describes the functional relationship between the dose level and pharmacokinetic endpoint on the log scale via

$$y_{km} = \log(x_{km}) = \mu + \beta \cdot \log(D_k) + e_{km},$$

where

y_{km} logarithm of response (pharmacokinetic parameter) measured on subject m receiving dose k,
μ the overall mean,
β slope parameter of linear regression line,
D_k level of dose k, k=1,…,K,
e_{km} the random error associated with the m^{th} subject who was administered dose k ($e_{km} \sim N(0, \sigma^2)$ iid).

The slope parameter β together with its two-sided 90% confidence interval will be estimated. Additionally, the r-fold change $r^{\beta-1}$ together with its 90% CI will be derived.

As some small doses at the beginning and/or some doses at the upper end might not contribute to the linear relationship between dose and pharmacokinetics, dose proportionality over the entire dose range investigated might not be shown. In that case an attempt will be made to identify a subrange of at least 3 consecutive doses where dose proportionality can be concluded.

[redacted]

7.3.4 Safety analyses

Safety will be assessed as defined by the endpoints listed in Section 2.1.2 and 2.2.2 based on the treated set (TS). Safety analyses will be descriptive in nature and will be based on BI standards.

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).

Treatments will be compared in a descriptive way. The placebo group in the safety evaluation will consist of all subjects treated with placebo, regardless of the dose group in which they were treated. The test treatment groups will be compared to the placebo group in a descriptive way. Tabulations of frequencies/proportions will be used for the evaluation of categorical (qualitative) data, and tabulations of descriptive statistics will be used to analyse continuous (quantitative) data.

Measurements (such as ECGs, 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 intake of trial medication will be assigned to the screening period, those between the trial medication intake and end of REP (see Section 1.2.6) or next intake of study medication (BA/ FE part) will be assigned to the treatment period. Events occurring after the REP but prior to trial termination date will be assigned to 'follow-up'. 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 database lock will be reported to Pharmacovigilance only and will not be captured in the trial database.

Additionally, further treatment intervals (called analysing treatments) may be defined in the TSAP in order to provide summary statistics for other than above periods, such as combined treatments, on-treatment totals, or periods without treatment effects (such as screening and post-study intervals).

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). Frequency, severity, and causal relationship of AEs will be tabulated by treatment, system organ class and preferred term. SAEs, AESIs (see Section 5.2.6.1) and other significant AEs (according to ICH E3) will be listed separately.

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

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

Vital signs or other safety-relevant data will be assessed with regard to possible on-treatment changes from baseline.

SRD-Part

The ECG variables QT, PR, QRS, and RR obtained from the centralised evaluation of 12-lead ECG recordings (see Section 5.2.4) will be the basis for the derivation of quantitative

and categorical ECG endpoints with regard to QT/QTc interval, HR, PR interval and QRS duration. These endpoints and their analyses will be described in the TSAP.

BA/ FE-Part

Relevant ECG findings will be reported as AEs.

7.3.5 Pharmacokinetic - pharmacodynamic analysis (SRD-Part only)

Not applicable

Applicable for Amendment No. 7

[redacted]

7.4 INTERIM ANALYSES

SRD-Part

No interim analysis is planned.

A preliminary analysis of safety and non-compartmental pharmacokinetic parameters (AUC_{0-24} and C_{max} of BI 1569912) provided as individual values and geometric means will be performed.

In contrast to the final PK/ PD calculations, the preliminary pharmacokinetic analysis will be based on planned sampling times rather than on actual times, regardless of whether actual times were within the time windows. Therefore, minor deviations may occur between preliminary and final results. The preliminary analysis will provide individual and mean concentration/ effect-time profiles and summary statistics of individual values without subject identification information. The preliminary results will be distributed to the investigator and the trial team.

Depending on the results of available preliminary PK/ PD analyses and the tolerability and safety of the compound, changes to the dosing schedule (e.g., additional intermediate doses), and additional PK/ PD preliminary analysis may be performed if requested by the Clinical Trial Leader, the Investigator, or Trial Clinical Pharmacokineticist. Preliminary PK/ PD results will not be reported in the CTR.

No inferential statistical interim analysis is planned. However, after completion of each dose group the investigator (or his or her deputy) is allowed to postpone further dose progression until a preliminary analysis of the data has been performed.

BA/ FE-Part

No interim analysis is planned.

7.5 HANDLING OF MISSING DATA

7.5.1 Safety

It is not planned to impute missing values for safety parameters.

7.5.2 Pharmacokinetics

Handling of missing pharmacokinetic data will be performed according to the relevant Corporate Procedure (001-MCS-36-472).

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

7.6 RANDOMISATION

SRD-Part

Subjects will be partially randomised within each dose group in a 3:1 ratio (test treatment to placebo).

BA/ FE-Part

Subjects will be randomised to one of the 6 treatment sequences in a 1:1:1:1:1:1 ratio. The block size will be documented in the CTR.

SRD-Part & BA/ FE-Part

The sponsor will arrange for the randomisation as well as packaging and labelling of trial medication. The randomisation list will be generated using a validated system that uses a pseudo-random number generator and a supplied seed number so that the resulting allocation is both reproducible and non-predictable.

The randomisation list will contain additional blocks to allow for subject replacement (refer to Section 3.3.5).

7.7 DETERMINATION OF SAMPLE SIZE

SRD-Part

It is planned to include a total of 72 subjects in this part of the trial. The planned sample size is not based on a power calculation. The size of 8 subjects per dose group (6 on active treatment, and 2 on placebo) is commonly used in single-rising dose studies of the present type and is in general considered as sufficient for the exploratory evaluation of single dose safety and pharmacokinetics.

Additional subjects may be entered to allow testing of additional doses on the basis of experience gained during the trial conduct (e.g., preliminary pharmacokinetic data), provided

the planned and approved highest dose will not be exceeded. Thus, the actual number of subjects entered in the SRD part may exceed 72 but will not exceed 88 subjects entered.

BA/ FE-Part

It is planned to enter a total of 12 subjects in this part of the trial because this sample size is considered sufficient to achieve the aims of this exploratory trial part. With this sample size, the following precision in estimating the ratio of geometric means (test/ reference) can be expected with 95% probability. Precision is defined as the ratio of upper CI limit to the relative BA estimate. Note that the precision is independent of the actual ratio of geometric means.

For this First-in-Man trial, no information on intra-subject variability is available. Therefore, Table 7.7: 1 provides an overview on the achievable precision for estimating the ratio of geometric means (test/ reference) for three different gCV. For illustrative purposes, the expected 90% confidence intervals with 95% coverage probability are displayed for different values of geometric mean ratios T/R in the three-period six-sequence crossover design.

Table 7.7: 1 Precision that can be expected with 95% probability and illustrative two-sided 90% confidence intervals around the ratios of geometric means (T/R) for different gCVs in a 3x3 crossover trial (N=12)

gCV[%]	**Precision (upper CI limit/ relative BA estimate)**	**Ratio1 [%]**	**90% CI [%]**
15	1.141	80	(70.13, 91.25)
		100	(87.67, 114.07)
		125	(109.58, 142.58)
20	1.191	80	(67.17, 95.28)
		100	(83.97, 119.10)
		125	(104.96, 148.87)
25	1.243	80	(64.38, 99.41)
		100	(80.47, 124.27)
		125	(100.59, 155.33)

[1]Ratio of geometric means (test/ reference) for a pharmacokinetic endpoint is defined by $\exp(\mu T)/\exp(\mu R)$.

The calculation was performed as described by Julious [R11-5230] using R Version 3.5.1.

8. INFORMED CONSENT, TRIAL RECORDS, DATA PROTECTION, PUBLICATION POLICY, AND ADMINISTRATIVE STRUCTURE

For details on SARS-CoV-2/ COVID-19 specific measures, refer to Appendix 10.5.

The trial will be carried out in compliance with the protocol, the ethical principles laid down in the Declaration of Helsinki, in accordance with the ICH Harmonized Guideline for Good Clinical Practice (GCP), relevant BI Standard Operating Procedures (SOPs), the EU regulation 536/ 2014, and other relevant regulations. Investigators and site staff must adhere to these principles.

Standard medical care (prophylactic, diagnostic, and therapeutic procedures) remains the responsibility of the subject's treating physician.

The investigator will inform the sponsor immediately of any urgent safety measures taken to protect the trial subjects against any immediate hazard, as well as of any serious breaches of the protocol or of ICH GCP.

The Boehringer Ingelheim transparency and publication policy can be found on the following web page: trials.boehringer-ingelheim.com. The rights of the investigator and of the sponsor with regard to publication of the results of this trial are described in the investigator contract. As a general rule, no trial results should be published prior to archiving of the CTR.

The terms and conditions of the insurance coverage are made available to the investigator and the subjects, and are stored in the ISF.

8.1 TRIAL APPROVAL, SUBJECT INFORMATION, INFORMED CONSENT

This trial will be initiated only after all required legal documentation has been reviewed and approved by the responsible Institutional Review Board (IRB)/ Independent Ethics Committee (IEC) and competent authority (CA) according to national and international regulations. The same applies for the implementation of changes introduced by amendments.

Prior to a subject's participation in the trial, written informed consent must be obtained from each subject according to ICH-GCP and to the regulatory and legal requirements of the participating country. Each signature must be personally dated by each signatory and the informed consent and any additional subject-information form retained by the investigator as part of the trial records. A signed copy of the informed consent and any additional subject information must be given to each subject.

The subject must be given sufficient time to consider participation in the trial. The investigator or delegate obtains written consent of the subject's own free will with the informed consent form after confirming that the subject understands the contents. The investigator or [redacted] delegate must sign (or place a seal on) and date the informed consent form. If a trial collaborator has given a supplementary explanation, the trial collaborator also signs (or places a seal on) and dates the informed consent.

Re-consenting may become necessary when new relevant information becomes available and should be conducted according to the sponsor's instructions.

The consent and re-consenting process should be properly documented in the source documentation.

8.2 DATA QUALITY ASSURANCE

A risk-based approach is used for trial quality management. It is initiated by the assessment of critical data and processes for trial subject protection and reliability of the results as well as identification and assessment of associated risks. An Integrated Quality and Risk Management Plan documents the rationale and strategies for risk management during trial conduct including monitoring approaches, vendor management and other processes focusing on areas of greatest risk.

Continuous risk review and assessment may lead to adjustments in trial conduct, trial design or monitoring approaches.

A quality assurance audit/inspection of this trial may be conducted by the sponsor, sponsor's designees, or by IRB/ IEC or by regulatory authorities. The quality assurance auditor will have access to all medical records, the investigator's trial-related files and correspondence, and the informed consent documentation of this clinical trial.

8.3 RECORDS

CRFs for individual subjects will be provided by the sponsor. For drug accountability, refer to Section 4.1.8.

8.3.1 Source documents

In accordance with regulatory requirements, the investigator should prepare and maintain adequate and accurate source documents and trial records for each trial subject that include all observations and other data pertinent to the investigation. Source data as well as reported data should follow the 'ALCOA principles' and be attributable, legible, contemporaneous, original, and accurate. Changes to the data should be traceable (audit trail).

Data reported on the CRF must be consistent with the source data or the discrepancies must be explained.

Before providing any copy of subjects' source documents to the sponsor, the investigator must ensure that all subject identifiers (e.g., subject's name, initials, address, phone number, and social security number) have properly been removed or redacted to ensure subject confidentiality.

If the subject is not compliant with the protocol, any corrective action (e.g., re-training) must be documented in the subject file.

For the CRF, data must be derived from source documents, for example:

- Subject identification: sex, year of birth (in accordance with local laws and regulations)
- Subject participation in the trial (substance, trial number, subject number, date subject was informed)

- Dates of subject's visits, including dispensing of trial medication
- Medical history (including trial indication and concomitant diseases, if applicable)
- Medication history
- AEs and outcome events (onset date [mandatory], and end date [if available])
- SAEs (onset date [mandatory], and end date [if available])
- Concomitant therapy (start date, changes)
- Originals or copies of laboratory results and other imaging or testing results (e.g., EEG), with proper documented medical evaluation (in validated electronic format, if available)
- ECG results (original or copies of printouts)
- Completion of subject's participation in the trial (end date; in case of premature discontinuation, document the reason for it, if known)
- Prior to allocation of a subject to a treatment into the clinical trial, there must be documented evidence in the source data (e.g., medical records) that the trial participant meets all inclusion criteria and does not meet any exclusion criteria. The absence of records (either medical records, verbal documented feedback of the subject or testing conducted specific for a protocol) to support inclusion/ exclusion criteria does not make the subject eligible for the clinical trial.

8.3.2 Direct access to source data and documents

The investigator/ institution will allow site trial-related monitoring, audits, IRB/ IEC review and regulatory inspections. Direct access must be provided to the CRF and all source documents/ data, including progress notes, copies of laboratory and medical test results, which must be available at all times for review by the Clinical Research Associate, auditor and regulatory inspector (e.g., FDA). They may review all CRFs and informed consents. The accuracy of the data will be verified by direct comparison with the source documents described in Section 8.3.1. The sponsor will also monitor compliance with the protocol and GCP.

8.3.3 Storage period of records

Trial site:
The trial site must retain the source and essential documents (including ISF) according to contract or the local requirements valid at the time of the end of the trial (whatever is longer).

Sponsor:
The sponsor must retain the essential documents according to the sponsor's SOPs.

8.4 EXPEDITED REPORTING OF ADVERSE EVENTS

BI is responsible to fulfil their legal and regulatory reporting obligation in accordance with regulatory requirements.

8.5 STATEMENT OF CONFIDENTIALITY AND SUBJECT PRIVACY

Individual subject data obtained as a result of this trial is considered confidential and disclosure to third parties is prohibited with the exceptions noted in Section 8.7.

Data protection and data security measures are implemented for the collection, storage and processing of patient data in accordance with the principles 6 and 12 of the WHO GCP handbook.

Personalised treatment data may be given to the subject's personal physician or to other appropriate medical personnel responsible for the subject's welfare. Data generated at the site as a result of the trial need to be available for inspection on request by the participating physicians, the sponsor's representatives, by the IRB/ IEC and the regulatory authorities.

8.5.1 Collection, storage and future use of biological samples and corresponding data

Measures are in place to comply with the applicable rules for the collection, storage and future use of biological samples and clinical data, in particular

- Sample and data usage has to be in accordance with the informed consent
- The BI-internal facilities storing biological samples from clinical trial participants as well as the external banking facility are qualified for the storage of biological samples collected in clinical trials
- An appropriate sample and data management system, incl. audit trail for clinical data and samples to identify and destroy such samples according to ICF is in place
- A fit for the purpose documentation (biomarker proposal, analysis plan and report) ensures compliant usage
- If applicable, a fit for purpose approach will be used for assay/ equipment validation depending on the intended use of the biomarker data
- Samples and/ or data may be transferred to third parties and other countries as specified in the ICF

8.6 TRIAL MILESTONES

The **start of the trial** is defined as the date when the first subject in the whole trial signs informed consent.

The **end of the trial** is defined as the 'date of the last visit of the last subject in whole trial' ('Last Subject Completed') or 'end date of the last open AE' or 'date of the last follow-up test' or 'date of an AE has been decided as sufficiently followed-up', whichever is latest.

Early termination of the trial is defined as the premature termination of the trial for any reason before the end of the trial as specified in this protocol.

Temporary halt of the trial is defined as any unplanned interruption of the trial by the sponsor with the intention to resume it.

Suspension of the trial is defined as an interruption of the trial based on a Health Authority request.

The EC/ competent authority in each participating EU member state will be notified about the trial milestones according to the laws of each member state.

A final report of the clinical trial data will be written only after all subjects have completed the trial in all countries (EU or non-EU), so that all data can be incorporated and considered in the report.

8.7 ADMINISTRATIVE STRUCTURE OF THE TRIAL

The trial is sponsored by Boehringer Ingelheim (BI).

The trial will be conducted at [redacted], under the supervision of the Principal Investigator. Relevant documentation on the participating (Principal) Investigators (e.g., their curricula vitae) will be filed in the ISF.

BI has appointed a Clinical Trial Leader, responsible for coordinating all required trial activities, in order to

- Manage the trial in accordance with applicable regulations and internal SOPs
- Direct the clinical trial team in the preparation, conduct, and reporting of the trial
- Ensure appropriate training and information of local clinical monitors (CTM), Clinical Research Associates, and investigators of participating trial sites

The trial medication will be provided by the [redacted].

Safety laboratory tests will be performed by the local laboratory of the trial site ([redacted]).

Analyses of BI 1569912 concentrations in plasma will be performed at the [redacted] or a suitable contract research organisation.

The digitally recorded EEGs will be sent to a specialised contract research organisation ([redacted]) for evaluation.

The digitally recorded [redacted] measurements will be sent to a specialised contract research organisation ([redacted]) for evaluation.

The digitally recorded 12-lead ECGs will be sent to a specialised contract research organisation ([redacted]) for evaluation. On-site monitoring will be performed by BI or a contract research organisation appointed by BI.

Data management and statistical evaluation will be done by BI or a contract research organisation appointed by BI, according to BI SOPs.

Tasks and functions assigned in order to organise, manage, and evaluate the trial are defined according to BI SOPs. A list of responsible persons and relevant local information can be found in the ISF.

9. REFERENCES

9.1 PUBLISHED REFERENCES

P06-11048 Taylor TJ; Diringer K; Russell T; Venkatakrishnan K; Wilner K; Crownover PH, Benincosa LJ; Gibbs MA. Absolute oral bioavailability of traxoprodil in cytochrome P450 2D6 extensive and poor metabolisers. Clin Pharmacokinet 2006; 45(10); 989-1001.

P06-11895 Rush AJ, Trivedi MH, Wisniewski SR, Nierenberg AA, Stewart JW, Warden D, Niederehe G, Thase ME, Lavori PW, Lebowitz BD, McGrath PJ, Rosenbaum JF, Sackeim HA, Kupfer DJ, Luther J, Fava M. Acute and longer-term outcomes in depressed outpatients requiring one or several treatment steps: a STAR*D report. Am J Psychiatry 2006 ; 163(11) ; 1905-1917.

R03-2269 Guidance for industry: food-effect bioavailability and fed bioequivalence studies. In: U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER); 2002. p. 1-9.

R06-0086 Trivedi MH; Rush AJ; Wisniewski SR; Nierenberg AA; Warden D; Ritz L; Norquist G; Howland RH; Lebowitz B; McGrath PJ; Shores-Wilson K; Biggs MM; Balasubramani GK; Fava M; STAR*D Study Team. Evaluation of outcomes with citalopram for depression using measurement-based care in STAR*D: implications for clinical practice. Am J Psychiatry 2006 ; 163(1) ; 28-40.

R06-1037 Guidance for industry: estimating the maximum safe starting dose in initial clinical trials for therapeutics in adult healthy volunteers. U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER); 2005.

R07-4722 Guidance for industry: E14 clinical evaluation of QT/QTc interval prolongation and proarrhythmic potential for non-antiarrhythmic drugs. Rockville: U.S. Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research (CDER), Center for Biologics Evaluation and Research (CBER); 2005.

R09-4830 Brown BD, Badilini F. HL7 aECG implementation guide (March 21, 2005). 2005

R11-5230 Julious SA. Sample sizes for clinical trials. Boca Raton: Taylor & Francis Group (2010).

R14-3147 Ferrari AJ; Somerville AJ; Baxter AJ; Norman R; Patten SB; Vos T; Whiteford HA. Global variation in the prevalence and incidence of major depressive disorder: a systematic review of the epidemiological literature. Psychol Med (Lond) 2013 ; 43(3) ; 471-481.

R16-0366 E14 Implementation Working Group. ICH E14 guideline: the clinical evaluation of QT/QTc interval prolongation and proarrhythmic potential for non-antiarrhythmic drugs: questions & answers (R3) (current version dated 10 December 2015). Website: ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Efficacy/E14/E14_Q_As_R3__Step4.pdf (access date: 29 January 2015) ; Geneva: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2015.

R17-1266 Bolon B; Garman RH; Pardo ID; Jensen K; Sills RC; Roulois A; et al. STP position paper: recommended practices for sampling and processing the nervous system (brain, spinal cord, nerve, and eye) during nonclinical general toxicity studies. Toxicol Pathol 2013 ; 41(7) ; 1028-1048.

R17-3810 Preskorn SH; Baker B; Kolluri S; Menniti FS; Krams M; Landen JW. An innovative design to establish proof of concept of the antidepressant effects of the NR2B subunit selective N-methyl-D-aspartate antagonist, CP-101,606, in patients with treatment-refractory major depressive disorder. J Clin Psychopharmacol 2008 ; 28(6) ; 631-637.

R19-0549 Miller OH; Yang L; Wang CC; Hargroder EA; Zhang Y; Delpire E; et al. GluN2B-containing NMDA receptors regulate depression-like behavior and are critical for the rapid antidepressant actions of ketamine. eLife 2014 ; 3; e03581.

R19-0553 Caddy C; Giaroli G; White TP; Shergill SS; Tracy DK. Ketamine as the prototype glutamatergic antidepressant: pharmacodynamics actions, and a systematic review and meta-analysis of efficacy. Ther Adv Psychopharmacol 2014 ; 4(2) ; 75-99.

R19-0555 Hedegaard MK; Hansen KB; Andersen KT; Brauner-Osborne H; Traynelis SF. Molecular pharmacology of human NMDA receptors. Neurochem Int 2012 ; 61(4) ; 601-609.

R19-0681 Reynolds IJ; Miller RJ. Tricyclic antidepressants block N-methyl-D-aspartate receptors: similarities to the action of zinc. Br J Pharmacol 1988 ; 95; 95-102.

R19-0772 Berman RM; Cappiello A; Anand A; Oren DA; Heninger GR; Charney DS; et al. Antidepressant effects of ketamine in depressed patients. Biol Psychiatry 2000 ; 47(4) ; 351-354.

R19-0778 GBD 2016 Disease and Injury Incidence and Prevalence Collaborators. Global, regional, and national incidence, prevalence, and years lived with disability for 328 diseases and injuries for 195 countries, 1990 - 2016: a systematic analysis for the Global Burden of Disease study 2016. Lancet 2017 ; 390(10100) ; 1211-1259.

R19-0829 FDA approves new nasal spray medication for treatment-resistant depression; available only at a certified doctor's office or clinic (FDA news release, for immediate release, March 5, 2019). worldwideweb.fda.gov/NewsEvents/Newsroom/PressAnnouncements/ucm636327.htm (access date: 13 March 2019) ; U.S. Food and Drug Administration (FDA); 2019.

R19-0986 Paterson B; Fraser H; Wang C; Marcus R. A randomized, double-blind, placebo-controlled, sequential parallel study of CERC-301 in the adjunctive treatment of subjects with severe depression and recent active suicidal ideation despite antidepressant treatment. NNDC 2015, Ann Conf of the National Network of Depression Centers (NNDC), Ann Arbor, 5 - 6 Nov 2015 (Poster). 2015.

R19-1029 Nutt JG, Gunzler SA, Kirchhoff T, Hogarth P, Weaver JL, Krams M, et al. Effects of a NR2B selective NMDA glutamate antagonist, CP-101,606, on dyskinesia and parkinsonism. Mov Disord 2008. 23(13):1860-1866.

R20-0052 Bremner JD, Krystal JH, Putnam FW, Southwick SM, Marmar C, Charney DS, et al. Measurement of dissociative states with the Clinician-Administered Dissociative States Scale (CADSS). J Trauma Stress 1998; 11(1); 125-136.

R94-1529 Chow SC; Liu JP. Design and Analysis of Bioavailability and Bioequivalence Studies. New York: Marcel Dekker Inc; 1992.

R97-2207 Olney JW; Labruyere J; Price MT. Pathological changes induced in cerebrocortical neurons by phencyclidine and related drugs. Science 1989 ; 244; 1360-1362.

9.2 UNPUBLISHED REFERENCES

001-MCS-36-472 Standards and processes for analyses performed within Clinical Pharmacokinetics/Pharmacodynamics. Current version.

c29289852 [redacted] Investigator's brochure BI 1569912 in major depressive disorder (MDD). 03 Feb 2020.

c34977604 [redacted]. Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design). ctr-synoptic-interim-report BI 1569912. 23 Feb 2021

n00265114 [redacted] Core validation of an LC-MS/MS method for the quantification of BI 1569912 in dog plasma; calibration range 1.00 - 5000 nmol/L. 15 Nov 2019.

n00266469 [redacted] BI 1569912: 3-week oral (gavage) dose range finding study in Beagle dogs. 18B180. 08 Nov 2019.

n00267795 [redacted] Effects of BI 1569912 (0.15, 0.5 and 1.5 mg/kg p.o.) on gastrointestinal function in the conscious rat. GP2018/0048/PH4. 24 Oct 2019.

n00267798 [redacted] Effects of BI 1569912 (0.15, 0.5 and 1.5 mg/kg p.o.) on urine- and serum-derived parameters in conscious rats. GP2018/0049/Ph4. 25 Oct 2019.

n00269064 [redacted] BI 1569912: 6-week oral (gavage) toxicity study in rats. 19B058. 24 Feb 2020.

n00269216 [redacted] BI 1569912: 6-week oral (gavage) toxicity study in Beagle dogs. 19B059. 12 Dec 2019.

n00271186 [redacted] BI 1569912: Modified Irwin Study in Male and Female Rats (Single Oral Administration). QX04CK. 16 Mar 2020.

n00271550 [redacted] BI 1569912: Evaluation of Respiratory Parameters in the Conscious rat using Whole Body Bias Flow Plethysmography (Single Oral Administration). QS65JQ.

n00272730 [redacted] Core validation of an LC-MS/MS method for the quantification of BI 1569912 in diluted rat plasma (1+4 v/v with human plasma); calibration range 0.2 – 500 nmol/L. V748/19RM. 7 Nov 2019.

n00272731 [redacted] Core validation of an LC-MS/MS method for the quantification of BI 1569912 in diluted rat plasma (1+4 v/v with human plasma); calibration range 50.0 – 125000 nmol/L. V749/19RM. 08 Nov 2019.

n00272732 [redacted] Core validation of an LC-MS/MS method for the quantification of BI 1569912 in dog plasma; calibration range 1.00 - 5000 nmol/L. V750/19RM. 15 Nov 2019.

n00272786 [redacted] Inhibition of NR2B and NR2A by BI 1569912. BI1569912-Draheim-1909-B. 14 Oct 2019.

n00272820 [redacted] Quantitative whole-body autoradiography in male pigmented rats after single oral administration of [14C]BI 1569912. A128/19JS. 27 Sep 2019.

n00272895 [redacted] In vitro Pharmacology Study of Several Compounds. BI1569912-Just-1909-A. 13 Mar 2018.

n00272959 [redacted] Effects of BI 1569912 (0.3, 1 and 3 mg/kg, p.o.) on cardiovascular and respiratory functions in conscious rats. GP2018/0036/PH1. 25 Oct 2019.

n00272965 [redacted] Prediction of BI 1569912 Pharmacokinetics and Therapeutic Dose in Human. B6790. 11 Dec 2019.

n00272974 [redacted] Species comparison of in vitro binding of [14C]BI 1569912 to mouse, rat, dog and human plasma proteins and to the isolated proteins human serum albumin (HSA) and human alpha-1-acid glycoprotein (hAGP). A285/19RKU. 25 Oct 2019.

n00273070 [redacted] Data Report for Pharmacology Services (Panlabs Code 16707663 = BI 1569912). BI1569912-Just-1909-B. 03 May 2018.

n00273097 [redacted] The effect of the selective NMDA NR2B NAM BI 1569912 on EEG power spectra in conscious rats. BI1569912-Ferger-1910-D. 23 Oct 2019.

n00273364 [redacted] Effect of BI 1569912 in the mouse forced swimming test. BI1569912-Ceci-1910-U. 29 Nov 2019.

n00273820 [redacted] BI 1569912: Neurotoxicity Study by a Single Oral Gavage Administration to Han Wistar Rats with Histopathological Assessment of the Brain Following Euthanasia at 4 or 72 Hours Post-dose. PJ33CC.

n00273899 [redacted] BI 1569912: Effect on hERG inactivating tail currents recorded from stably transfected HEK 293 cells at near physiological temperature. A2434.

n00273900 [redacted] BI 1569912: Telemetric Evaluation of Cardiovascular Effects in the Conscious Dog (Single Oral Administration). NR26XB.

10. APPENDICES

10.1 RECONSTITUTION INSTRUCTIONS

TABLE OF CONTENTS

1. DRUG SUPPLIES OVERVIEW

a) BI 1569912 Powder for Oral Solution 50 mg (target solution concentration BI 1569912: 0.625 mg/mL), provided in 100 mL amber glass bottles with plastic screw cap

b) Solvent for Oral Solution 80 mL (Tartaric acid 5 mg/mL) provided in 100 mL amber glass bottle with plastic screw cap

2. REQUIRED EQUIPMENT AND DOSING AIDS – OVERVIEW

a) Mechanical (orbital) shaker for bottles (e.g., [redacted] Typ KL2)

b) Dosing dispensers/syringes and bottle adapters

For the withdrawal of respective volume aliquots from the final Oral Solution to be administered, amber [redacted] ExactaMed Syringes or [redacted] Exadoral oral dispensers should be used in a size as close as possible to the required dose volume. For this purpose, a range of syringe sizes from 1 mL up to 60 mL should be stocked in the trial site. For [redacted] Exadoral oral dispensers the largest dispenser size is 20 ml.

In order to ease the withdrawal of the oral solution from the glass bottles with the [redacted] ExactaMed Syringes or [redacted] Exadoral dispensers, [redacted] or [redacted] bottle adapters and dispenser tip caps should be used and stocked in the trial site:

Preferably [redacted] Adapta Cap Bottle Adapters (E-28 mm), [redacted] Press-In Bottle Adapters (PIBATM) or [redacted] Bottle adapter 28 mm.

Possible [redacted] Med Oral amber dispensers

- [redacted] ExactaMed amber oral dispenser 1 mL
- ExactaMed amber oral dispenser 3 mL
- ExactaMed amber oral dispenser 5 mL
- ExactaMed amber oral dispenser 10 mL
- ExactaMed amber oral dispenser 20 mL
- ExactaMed amber oral dispenser 60 mL

or alternatively:

- [redacted] Exadoral oral dispenser 1 mL
- Exadoral oral dispenser 2 mL
- Exadoral oral dispenser 5 mL
- Exadoral oral dispenser 10 mL
- Exadoral oral dispenser 20 mL

Only CE certified syringes and adapters are to be used!

3. RECONSTITUTION PROCEDURE

2 bottle concept, see also Chapter 4.

3.1 RECONSTITUTION PROCEDURE FOR THE PREPARATION OF THE ACTIVE BI 1569912 ORAL SOLUTION 0.625 MG/ML

3.1.1 Necessary materials

a) BI 1569912 Powder for Oral Solution 50 mg (target solution concentration BI 1569912: 0.625 mg/mL), provided in 100 mL amber glass bottles with plastic screw cap.

b) Solvent for Oral Solution 80 mL (Tartaric acid 5 mg/mL) provided in 100 mL amber glass bottles with plastic screw cap.

3.1.2 Reconstitution procedure

Step 1: Open the bottle containing the Solvent for Oral Solution 80 mL (Tartaric acid 5 mg/mL)

Step 2: Transfer the content of the Solvent for Oral Solution 80 mL (Tartaric acid 5 mg/mL) completely and carefully into the bottle containing the BI 1569912 Powder for Oral Solution 50 mg

Step 3: Close the bottle with the plastic screw cap and shake the bottle manually until the BI 1569912 powder is wetted. Mount the bottle in a horizontal recumbent position on a mechanical shaker (e.g., [redacted] Typ KL2)

Step 4: Let the bottle shake orbitally for 120 min. at 350 rpm in its horizontal recumbent position.

Step 5: Visually control that the powder is completely dissolved (clear to almost clear solution).

The final BI 1569912 Oral Solution concentration is 0.625 mg/mL.

The allowable dose range is from 0.25 mg – 50 mg.

3.2 SOLVENT FOR RAL SOLUTION FOR USE AS PLACEBO SOLUTION

3.2.1 Necessary materials

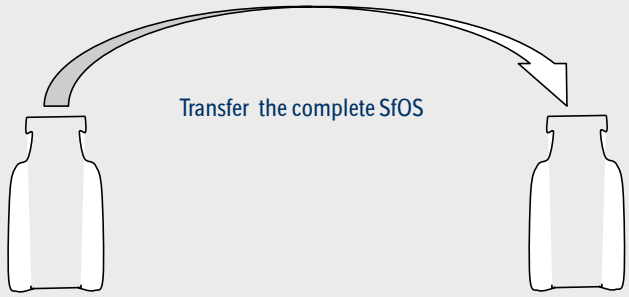
Solvent for Oral Solution 80 mL (Tartaric acid 5 mg/mL) provided in 100 mL amber glass bottles with plastic screw cap.

The Placebo Solution is the Solvent for Oral solution 80 mL (Tartaric acid 5 mg/mL).

4. ILLUSTRATION OF RECONSTITUTION PROCEDURE

The following scheme on the principle followed for the present PfoS formulation, 2-bottle concept, should serve as an additional illustration to clarify, how the reconstitution procedure for the preparation of the active oral solution has to be performed.

Active oral solution reconstitution procedure 2-bottle concept: SfOS + PfOS (API)



SfOS
Solvent for Oral Solution

PfOS
API powder in bottle
Powder for Oral Solution

Final solution for administration

0.625 mg/mL

Component	g/bottle	Function
Tartaric acid	0.400	Acid
Water	97.648	Solvent

Component	g/bottle	Function
BI 1569912	0,050	Active ingredient

The following picture below shows the bottles needed to prepare the PfoS formulation to clarify the procedure in additional.

5. IN-USE STABILITY

The in-use stability of the reconstituted solution is 24 h after its preparation, including storage in [redacted] dispensers or [redacted] Exadoral dispensers until administration.

6. MODE OF APPLICATION

Withdraw the required volume aliquot to obtain the required doses.

Use amber [redacted] ExactaMed syringes or [redacted] Exadoral dispensers for dose withdrawal/ administration.

Use [redacted] ExactaMed syringes or [redacted] Exadoral dispensers at a volume size as close as possible to the volume to be withdrawn.

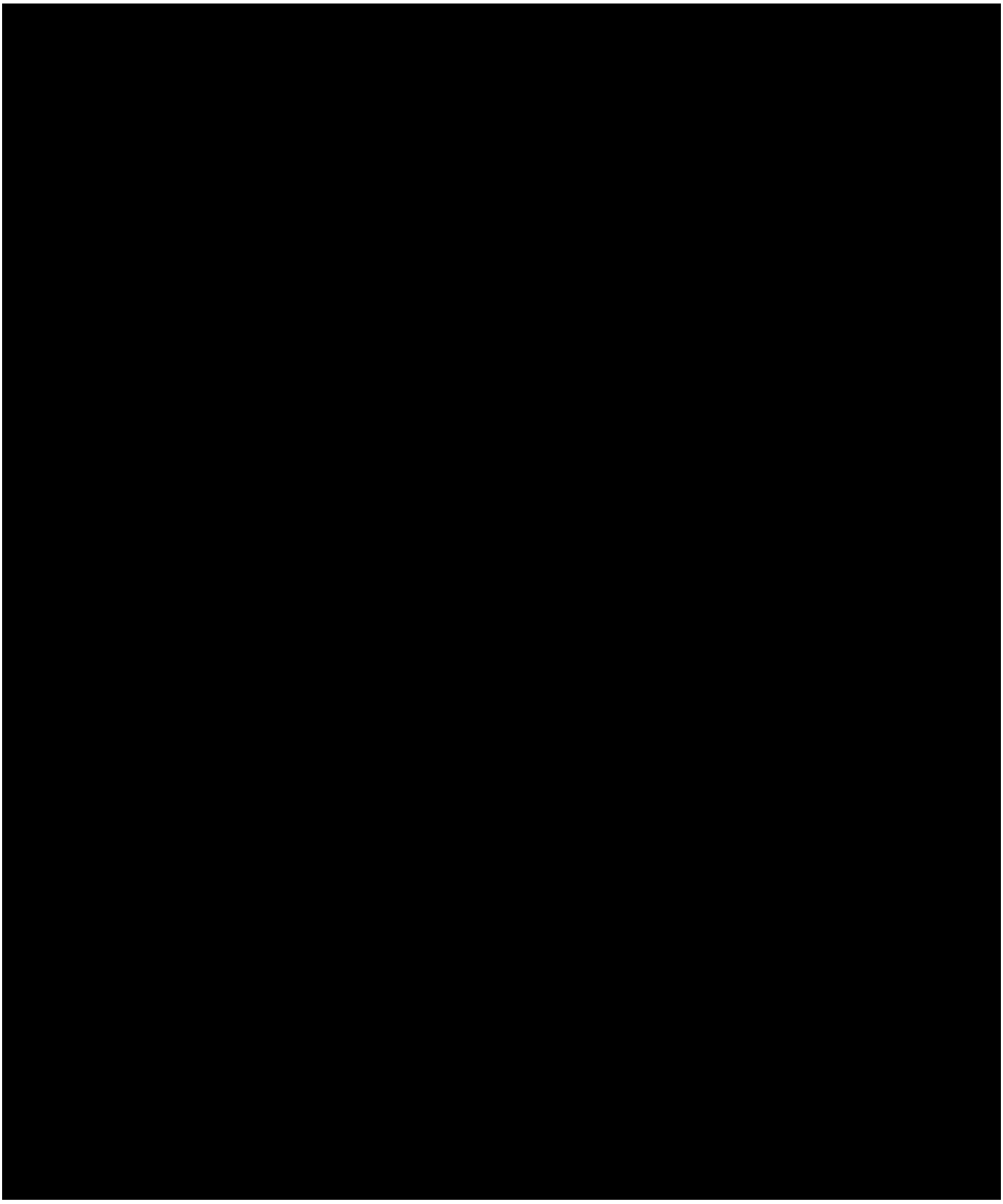
Please note that it is the responsibility of the CTL to assure that appropriate supplies are used for administration of a dose, based on guidance in the clinical trial protocol, and dosing is limited to the allowed dosing range for a specific dose formulation as stated in this Reconstitution Instruction.

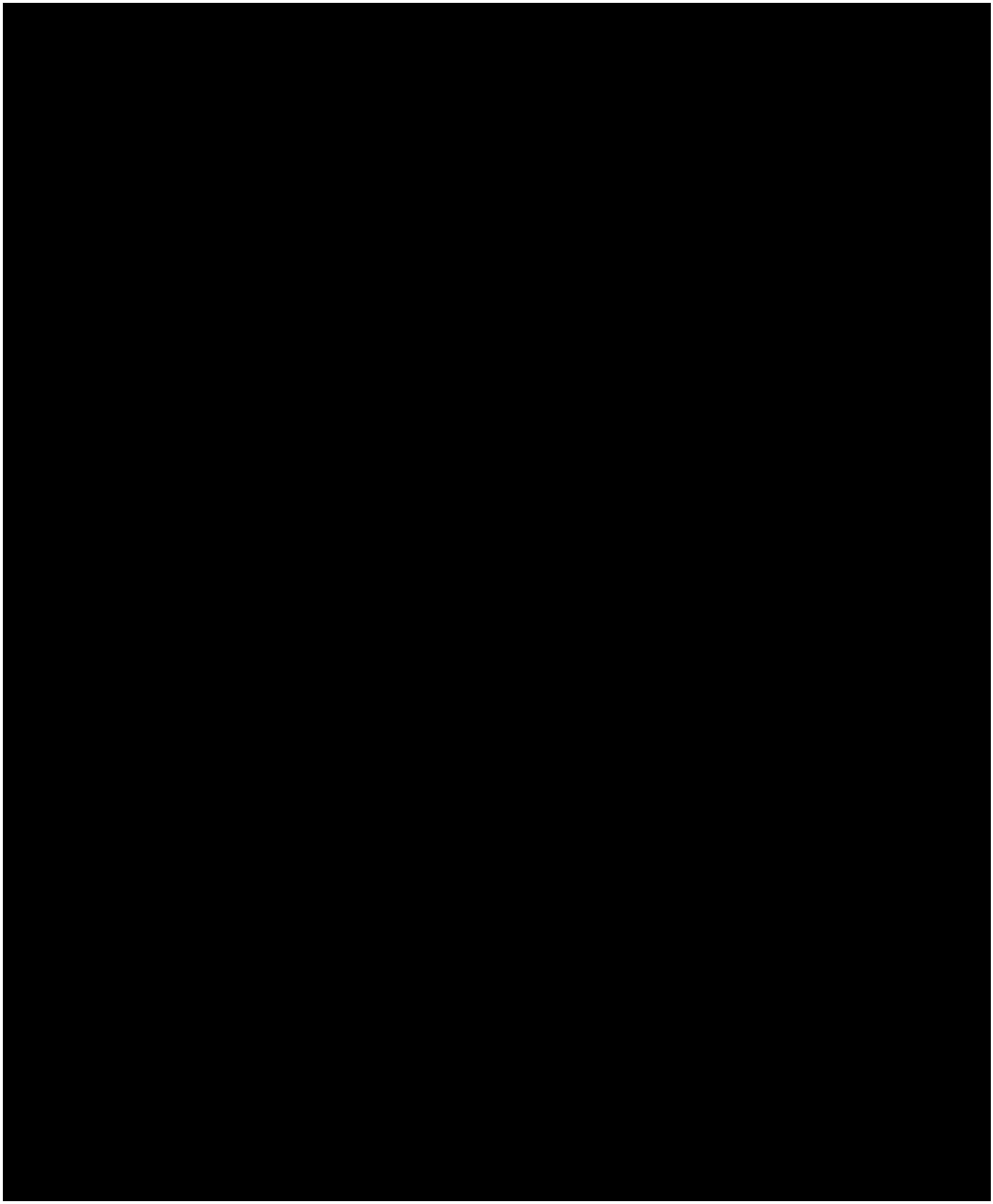
7. GENERAL REMARKS – IMPORTANT

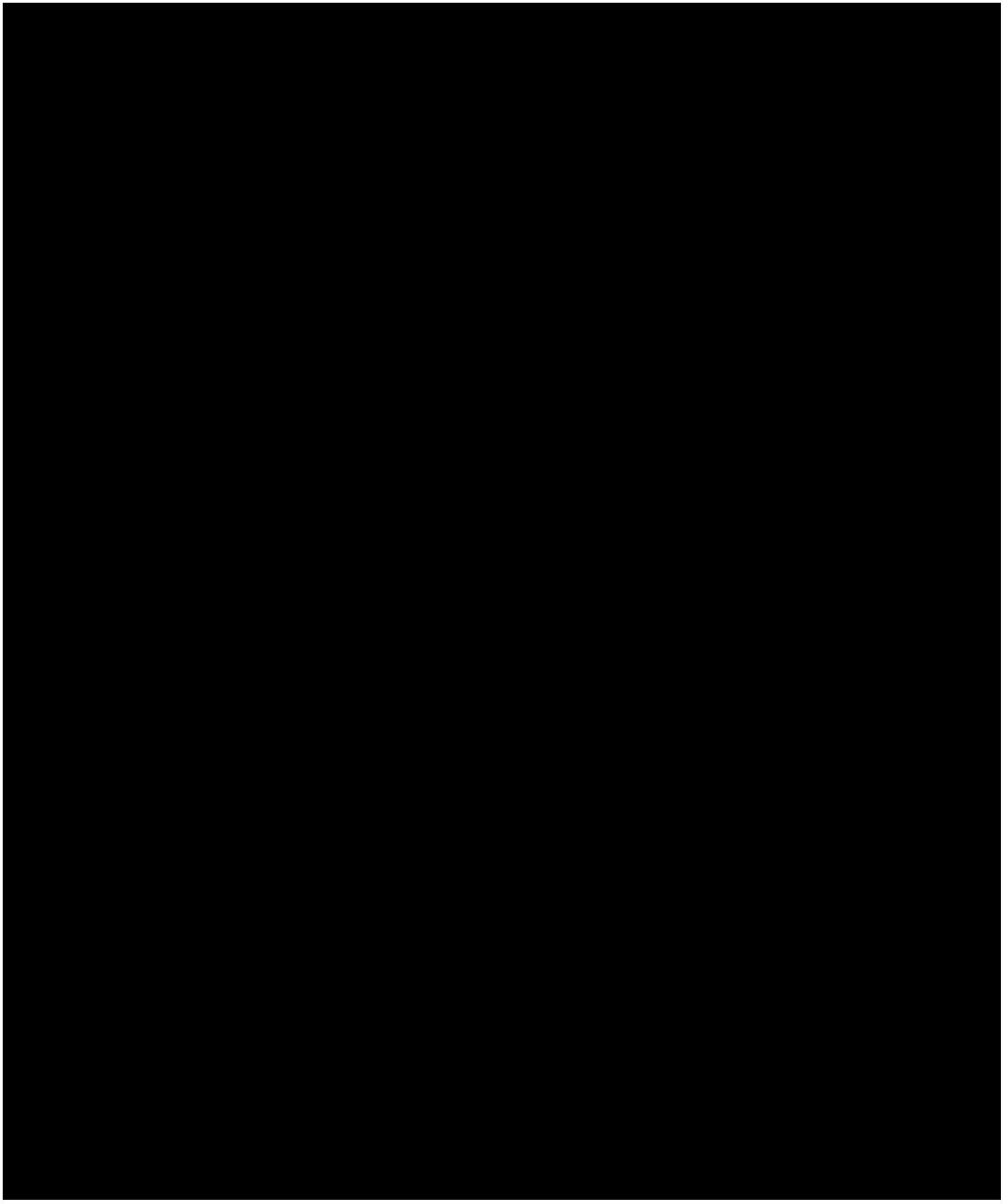
Because of lacking analytical coverage beyond the instructed preparation procedure of the different dose formulations, no further (external) dilutions of the reconstituted solutions are allowed.

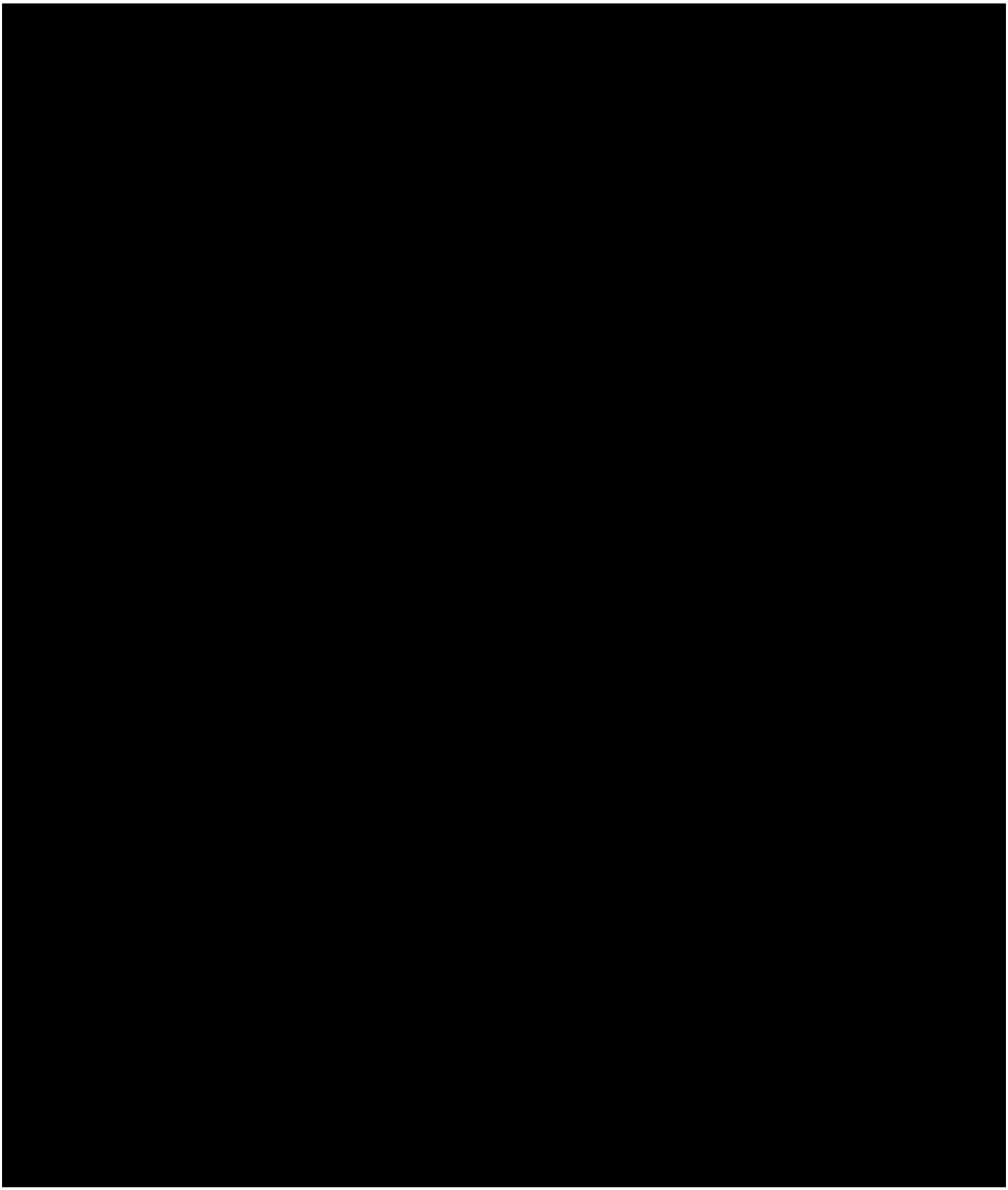
The present reconstitution instruction does not contain any advice how to withdraw a specific dose from the reconstituted solutions. The specific dose volumes to be withdrawn from the described dose formulations in order to obtain a required dose will be calculated and documented in the clinical trial protocol and subsequent documents (e.g., work sheets).

[redacted]

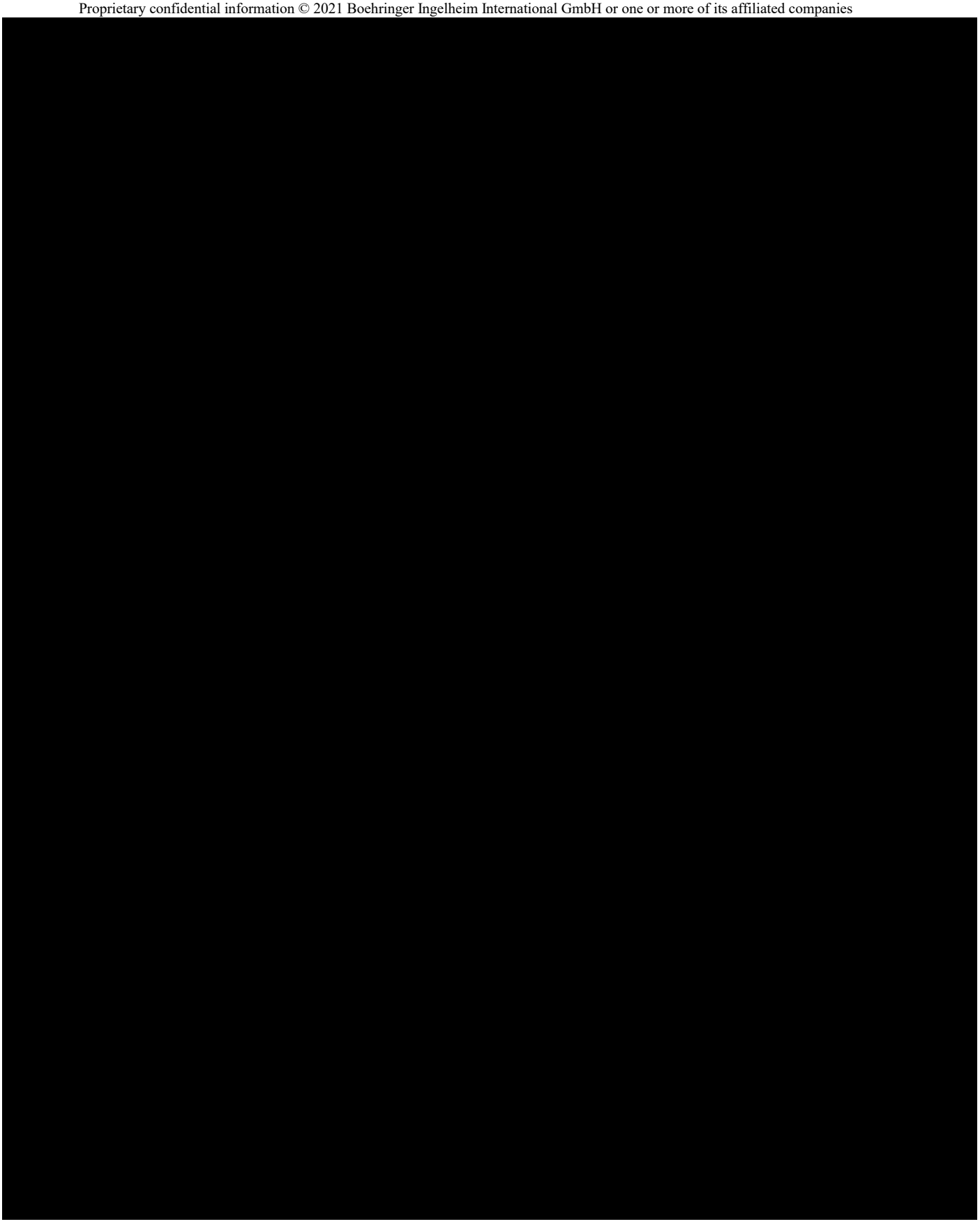


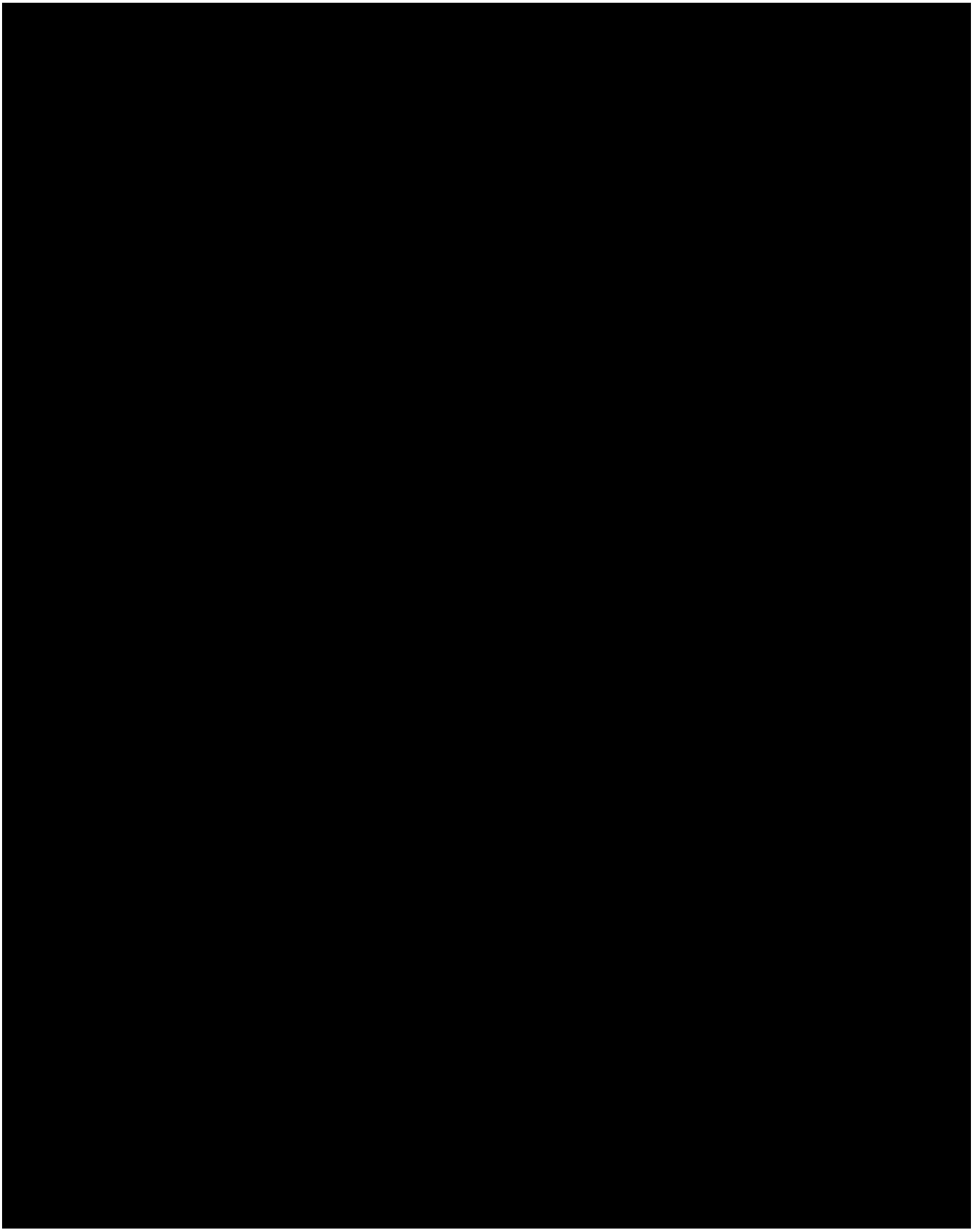


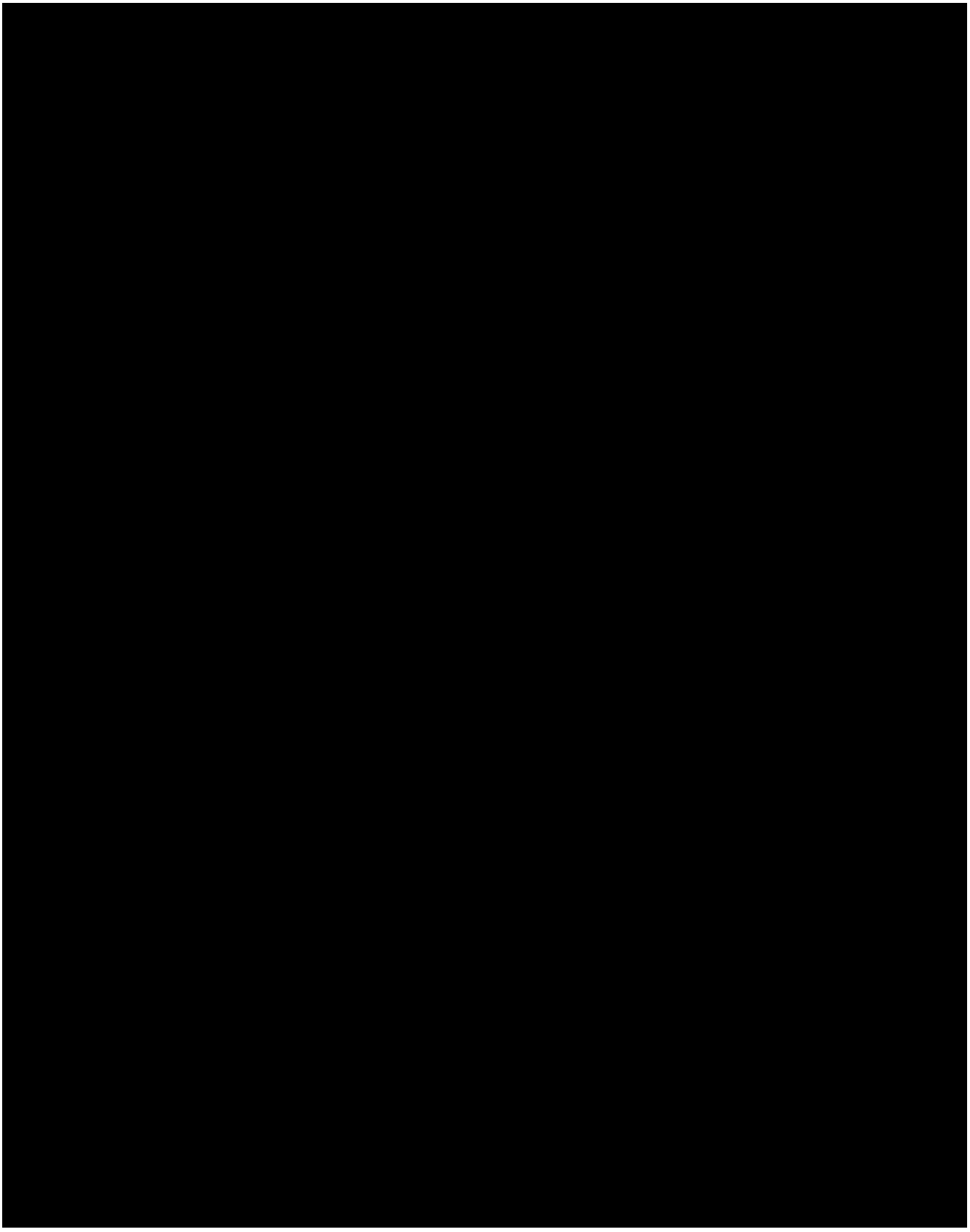


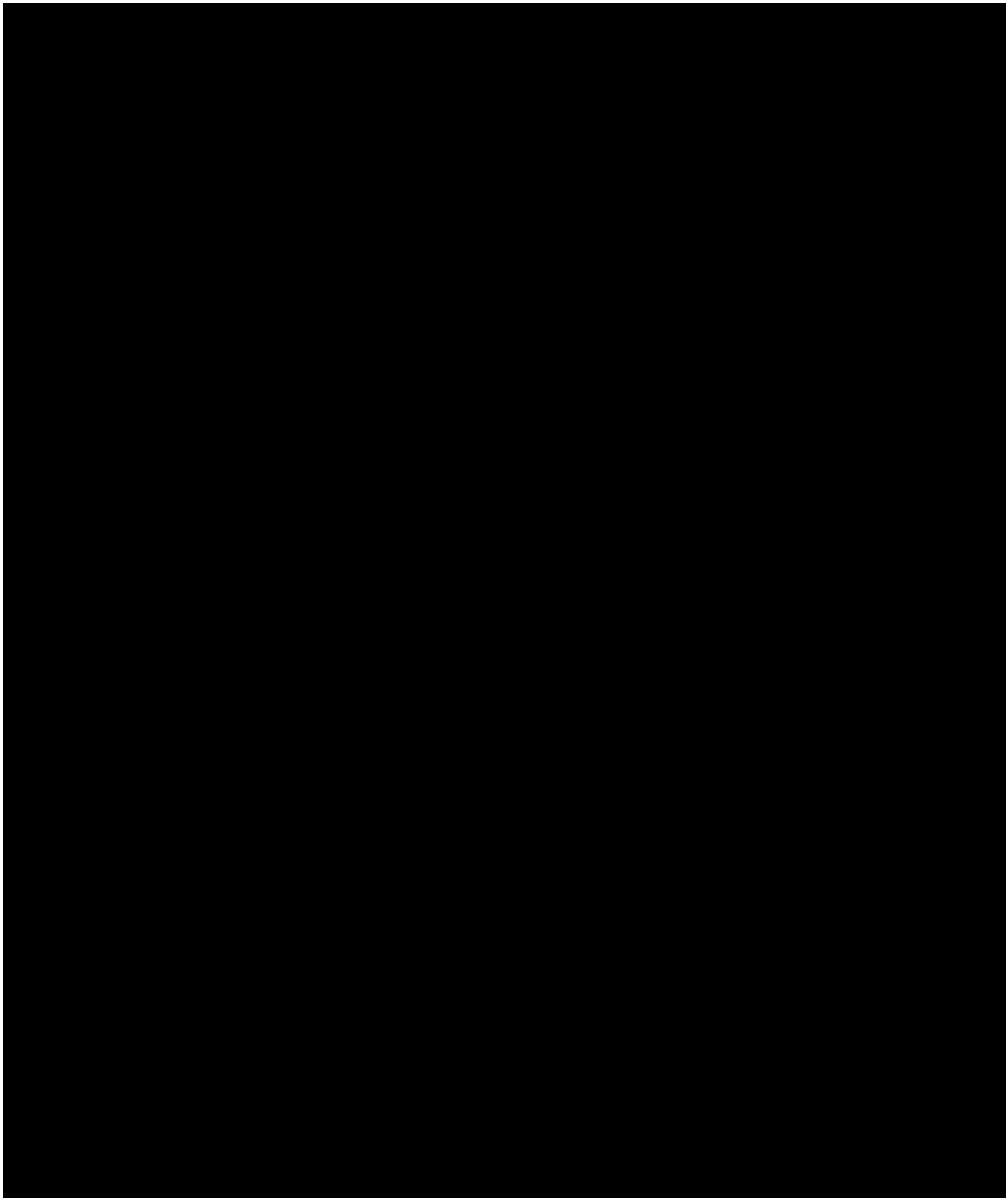


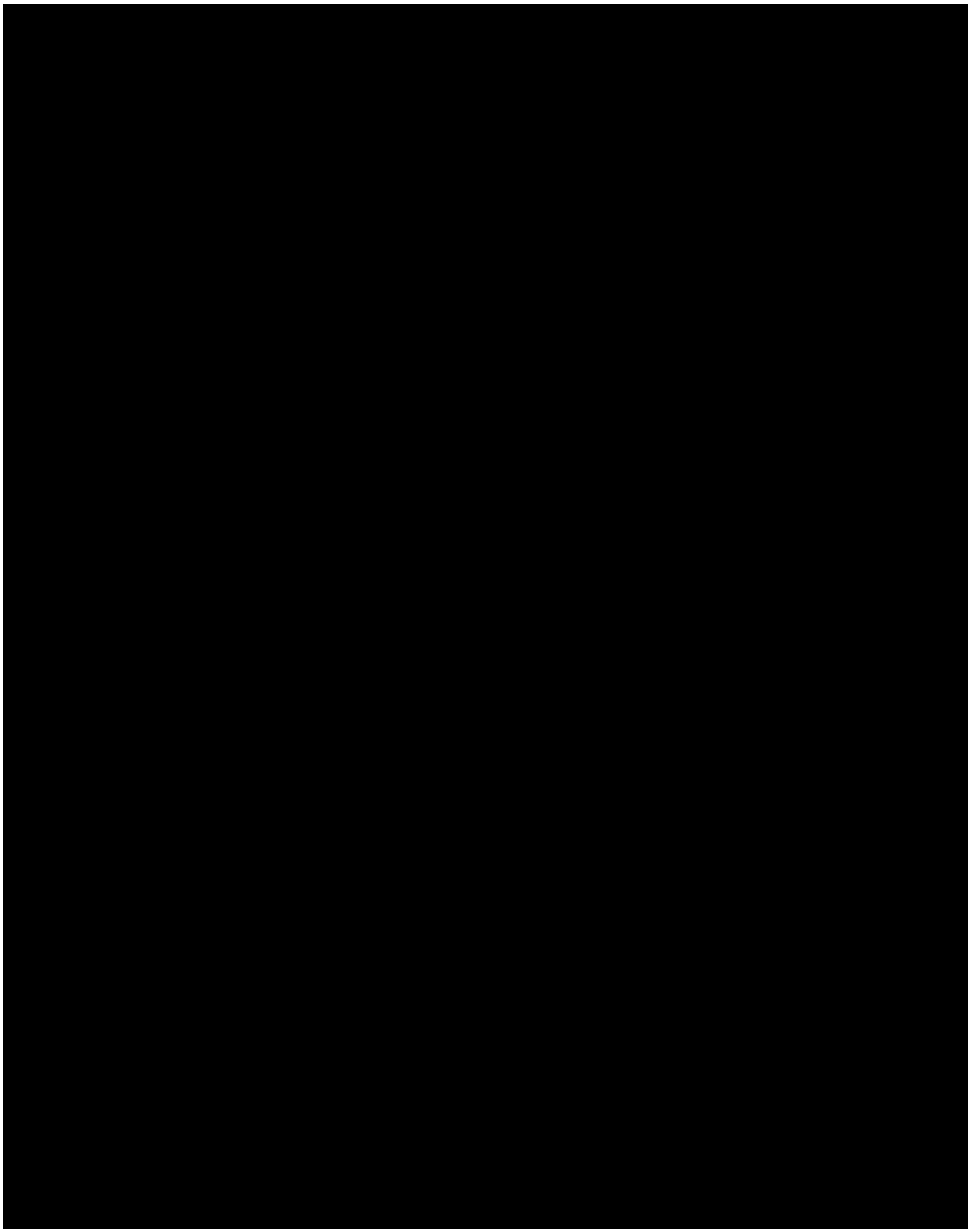
[redacted]











10.5 SARS-COV-2/ COVID-19 RELATED MEASURES

10.5.1 Introduction

Due to the SARS-CoV-2/ COVID-19 pandemic outbreak early 2020 additional measures were implemented to protect study participants and personal involved in clinical trials. This document summarizes these measures.

10.5.2 Screening

The following measures will be performed:

(1) Study participants will be informed about, and asked to agree to SARS-CoV-2/ COVID-19 specific requirements with their diagnostic, therapeutic, and legal implications

(2) **A PCR for SARS-CoV-2/ COVID-19, as offered by the trial site, will be performed at screening.**

(3) Evaluation of subjects before admission to the unit:
- Body temperature assessment
- Questionnaire and medical assessment
- SARS-CoV-2 test (mandatory)

10.5.3 Trial Conduct

A PCR for SARS-CoV-2/ COVID-19, will be performed on Day -3 of the SRD-Part and on Day -3 as well as before each period (should they leave the study premises) of the BA/ FE-Part (i.e. one day before admitted to the trial site)

10.5.3.1 Site-specific Measures

The following site specific measures will be adhered to during the conduct of the trial:

(1) Requirement that everyone (staff/ subjects/ visitors) wear masks.

(2) Medical staff with direct subject contact: use only employees passing 14 day quarantine or with at least one negative PCR.

(3) Daily evaluation for unit staff (at beginning of shift):
- Body temperature assessment.
- Self-assessment and disclosure of any symptoms.

(4) Ongoing evaluation of subjects during their stay in the unit.

(5) Separation of subject in the unit:
- Maximum 2 subjects per room.
- Food is brought to the subject's room.
- Subject related teams – i.e. can more readily relate individual staff to individual subjects.

- Until further notice, visitors for subjects are not allowed.

(6) Mandatory: SARS-CoV-2 PCR test in case of suspicion of infection.

(7) Protocols to be followed, in the event of SARS-CoV-2/ COVID-19 confirmation (e.g., isolation, sponsor/ authority notification, contact tracing, quarantined transport home, drop-out/ replacement).

(8) Additional measures such as use of telemedicine, remote monitoring (potentially), replacement of central labs with local labs are under discussion with the sponsor.

(9) General measures are in place including: entry checks for monitors, CRO visitors; minimization of Face-to-Face meetings; hygiene and social distancing measures.

10.5.3.2 Documentation of Adverse Events

SARS-CoV-2/ COVID-19 related adverse events will be documented as follows:

(1) Continue regular AE and SAE documentation, there is no change in the requirements of what to document and how.

(2) Continue also with expedited reporting of SAEs and AESI to PV in addition to documentation on the eCRF. The ways of transmitting the information stays as given on the contact sheet.

(3) If a patient experiences a SARS-CoV-2 infection, this will be entered as (S)AE (even if the subject did not experience symptoms).

(4) AE Start Date: The day when the subject experienced SARS-CoV-2/ COVID-19 symptoms or the day of the positive test should be entered as AE start date, whichever occurred first.

(5) AE End Date: The date of the last available negative test should be entered as AE end date. If the negative test date is not available, the date by when the subject has been received notice to be virus free should be used.

For any AE related to SARS-CoV-2/ COVID-19, please note that our standard processes should be followed, meaning:

(1) If a SARS-CoV-2 infection is associated with clinical symptoms (AE´s):

- Report as a non-serious AE, if the serious criteria are not met
- Report as a SAE, if serious criteria are met – e.g., hospitalization, serious for medical reasons, or AE term describing the clinical symptoms is on the “always serious list”
- The mere fact that someone is infected with SARS-CoV-2 should not lead to a judgement of seriousness
- Thus, no adaptation to standard procedures, CTPs or CRFs is required

(2) If a SARS-CoV-2 or any other infection is not associated with clinical symptoms, meaning there is just a positive SARS-CoV-2 test:

- The recommendation would be to consistently capture as a (non-serious) AE, as a positive Corona test means a patient has an infection
- This also would not require any adaptation to standard processes, CTP or CRF

Here are some examples from other events to illustrate the standard process:

(1) Event of Pneumonia

- Pneumonia is not on the always serious list, from that perspective it is not serious AE
- However, if the patient is hospitalized with acute hypoxaemic respiratory failure due to SARS-CoV-2/ COVID-19, then it certainly qualifies for a SAE

(2) Event of Viral Infection

- Viral infection treated as an out-patient or day-care is an AE (irrespective of SARS-CoV-2/ COVID-19 positive, negative or unknown)
- Viral infection treated with hospitalization is a SAE (irrespective of SARS-CoV-2/ COVID-19 positive, negative or unknown)

10.5.4 Monitoring

The [redacted] has confirmed that on-site monitoring will be possible, by applying safety measures as described under number (9) of Section 10.5.3.1 'Trial Conduct'.

The safety of trial participants overrules all other aspects of the trial, also and particularly during a COVID-19 pandemic. Study 1447-0001 is a single-dose trial in health volunteers at the [redacted], a dedicated phase 1 trial site, specialized in healthy volunteer studies. Therefore, the application of the Investigational Medicinal Product (IMP) can be stopped at any time without causing any harm to trial participants.

Should on-site monitoring not be possible, e.g., due to a SARS-CoV-2/ COVID-19 pandemic, the sponsor has put in place a number of working instructions (How-to-Guides), describing the conduct of remote monitoring visits. These documents also specify considerations how to weigh the interests of subjects (e.g., participant's safety, protection of privacy and the confidentiality of personal data) against the interests of the sponsor (e.g., regulatory compliance and data integrity). Source data verification will be performed as soon as possible but before data base lock.

In the following, trial specific aspects from BI's 'How-to Guides' regarding remote monitoring visits during a COVID-19 pandemic are summarized:

Site Initiation Visit (SIV)

BI's current processes allow for site initiation visits to be performed remotely, if the requirements have been met, i.e. if there is documented evidence that (1) Site Release Checklist is completed, (2) the first shipment of all relevant drug supplies is arranged to be available at the site and ready for use before the first subject is entered, (3) a valid record of

training completion is available for the PI for all items on the Trial Training Plan that are required to be performed in a face-to-face manner, (4) the monitoring frequency, as determined in the Monitoring Manual, can be complied with.

A site-risk assessment based on previous experience with the site and site-staff, as well as major concerns previously identified, will be performed. Under the COVID-19 situation, the resumption of site initiations and recruitment in the absence of on-site monitoring may be approved based on the outcome of this assessment. The site acceptance and ability to accommodate remote-site monitoring visits, and timely data entry during the time when on-site monitoring visits are not possible has to be confirmed.

If the SIV is done remotely, BI has still the obligation to ensure adequate monitoring once the first subject is enrolled or entered as required by the monitoring manual. While usually it will be acceptable to have this first monitoring visit also done remotely, the expectation is that it is soon followed by an on-site visit to ensure that the site is following the protocol and meeting the sponsor's expectations as to the subject's safety, and data integrity.

Site Monitoring Visit (SMV) during the Couse of a Trial

BI guidelines recommend to maintain the planned site monitoring frequency and, if site visits cannot be performed on-site, to perform them remotely, if possible.

It should be noted that *remote Source Data Verification is not included in the guidance for remote site monitoring visits* and would require special permission.

Site Close-out Visit (SCV)

It is recommended not to perform remote SCVs, but postpone them until the ban on visiting the site is lifted. Any deviations from the sponsor's timeline for closing the site can be documented in the SCV report.

If the IMP is still at the site, performing the SCV remotely should not be considered.

11. DESCRIPTION OF GLOBAL AMENDMENTS

11.1 GLOBAL AMENDMENT 1

Date of amendment		22 April 2020
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☐
Section to be changed		1.4.7 Measures of risk minimization (including safety precautions and stopping rules) 3.3.3 Exclusion criteria 3.3.4.3 Discontinuation of the trial by the sponsor 3.3.5 Replacement of subjects 4.1.1. Identity of the Investigational Medicinal Products 4.1.3 Method of assigning subjects to treatment groups 5.2.1 Physical examination 5.2.3 Safety laboratory parameters 5.2.7 Assessment of adverse events 6.2.1 Screening periods 8 Informed Consent, Trial Records, Data Protection, Publication, Policy and Administrative Structure 8.1 Trial Approval, Subject Information, Informed Consent 10.5 SARS-CoV-2 / COVID-19 Related Measures
Description of change		1.4.7: clarification and addition of ‘Measures of risk minimization’

		3.3.3: modification of exclusion criteria No. 3, 11 and 12, addition of exclusion criterion No. 29 3.3.4.3: alignment of discontinuation criteria 3.3.5: clarification of numbers of subjects being replaced 4.1.1: clarification dosing in BA/ FE-Part 4.1.3: clarification of allocation of subjects to dose groups 5.2.1: 'body temperature' added 5.2.3: reference to Appendix 10.5 added 5.2.7: reference to Appendix 10.5 added 6.2.1: reference to Appendix 10.5 added 8: reference to Appendix 10.5 added 8.1: 'Subjects legally accepted representative' taken out 10.5: 'Document that describes SARS-COV-2/ COVID-19 related measures' added
Rationale for change		Changes based on the request by BfArM and EC Berlin Minor editorial changes

11.2 GLOBAL AMENDMENT 2

Date of amendment		16 June 2020
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☐
Section to be changed		Section 1.3.3 Section 1.4.7 Section 3.1 Section 3.3.5 Appendix 10.5.4 Throughout the document
Description of change		Section 1.3.3, Section 1.4.7, Section 3.1: Inclusion of a substantial amendment - 'Before starting the BA/ FE-Part, a summary of safety and tolerability data (including AEs, SAEs, ARs and SARs) generated so fare during the course of the trial will be submitted via substantial amendment.' Section 1.4.7: Inclusion of additional stopping rule 'when the risk profile deteriorates and a necessary adjustment of the maximum insurance sum is not possible'. Section 3.3.5: Limitation of 'replacement subjects' - 'A maximum of 4 subjects can replace those who dropped-out during the course the trial for safety reasons, related or not related to the study medication.

		Appendix 10.5.4: Description of monitoring during a COVID-19 lock-down including remote monitoring measures. Throughout the document: Minor editorial and formatting changes.
Rationale for change		Factual changes are based on the request by the BfArM and EC Berlin. Minor editorial and formatting changes for consistency reasons.

11.3 GLOBAL AMENDMENT 3

Date of amendment		19 June 2020
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☐
Section to be changed		Flow Chart Abbreviations Section 1.4.7 Section 2.2.2.2 Section 5.2.6.3 Section 9.1 Section 10.4 Throughout the document
Description of change		Flow Chart, Abbreviations, Section 1.4.7, Section 2.2.2.2, Section 5.2.6.3, Section 9.1 and Section 10.4: Replacement of the DSS-akut by the CADSS questionnaire Throughout the document: Minor editorial and formatting changes
Rationale for change		A clinician administered questionnaire was selected because people in dissociated states may have trouble filling out questionnaires accurately. In addition, the clinician administered CADSS is intended to be used in other studies, therefore

		replacement of the DSS-akut by the CADSS questionnaire will allow for better comparison of findings across different studies within the project and compounds. Minor editorial and formatting changes for consistency reasons

11.4 GLOBAL AMENDMENT 4

Date of amendment		31 July 2020
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☐
Section to be changed		Flow Chart of SRD and BA/ FE part Abbreviations Section 4.1.4 Section 5.2.3 Section 5.2.4.1 Section 5.2.5 Section 5.3.2.1 Section 5.3.2.2 Section 5.3.2.4 Section 7.3.4 Section 8.7 Throughout the document
Description of change		Flow Chart of SRD and BA/ FE part: Change of safety laboratory from Day -1 to Day -3, and deletion of foot note 7; insertion of a 'fictive' planned time at screening of -504:00 h Flow Chart of SRD part: Additional time point (at -0:45 h) at start of continuous ECG monitoring within 3 hours prior drug administration; Flow Chart of BA/ FE part: Addition of foot note 14,

		to indicate base line ECGs at time point Day -1, -25:00 h; Flow Chart of BA/ FE part: Deletion of EEG/ qEEG time points on Day 1, -1:00 h and -0:45 h Flow Chart of BA/ FE part: Correction of time range of End of Trial examination from 4 to 14 to 16 to 26 days after first drug administration; adaptation of foot note 9; Abbreviations and Section 7.3.4: Deletion of QT interval corrected for heart rate, using now only the method of Fridericia (QTcF); Section 4.1.4: Prolongation of time during which subjects are not allowed to lie down from the first 2 to the first 4 h after drug administration; Section 5.2.3: Exclusionary laboratory tests can also be done at the trial site; no further test specification is made; Sections 5.2.4.1 and 5.2.5: Inclusion of ECG service provider [redacted] for ECG parameter capturing, and a description of data transfer; Sections 5.3.2.1 and 5.3.2.2: Increase of tube size for PK-sampling from 2.7 mL to 3.0 mL; Section 5.3.2.4: Increase of tube size for PK-sampling (stability-testing) from 1.2 mL to 2.0 mL, and adaptation of blood volume from 2.4 to 4 mL, accordingly; Section 5.3.2.4: Change of time point of blood drawing for PK-sampling (for stability-testing) from 4 h to 3 h after drug administration; Section 8.7: Insertion of ECG service provider [redacted]; Throughout the document: Minor editorial and formatting changes
Rationale for change		Inclusion of ECG service provider [redacted] for ECG parameter capturing, and a description of ECG-data transfer as well as ECG-evaluation; Change of safety laboratory time point from Day -1 to Day -3; insertion of a ‘fictive’ planned time at screening for data transfer (technical) reasons

		Deletion of EEG/ qEEG registration time points on Day 1 of the BA/ FE part; Increase of tube size for PK-sampling from 2.7 mL to 3.0 mL; Increase of tube size for PK-sampling (stability-testing) from 1.2 mL to 2.0 mL; Minor changes to make descriptions in the body of the protocol consistent with Flow Charts and vice versa; Minor editorial and formatting changes for consistency reasons;

11.5 GLOBAL AMENDMENT 5

Date of amendment		16 Oct 2020
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☐
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☒
Section to be changed		10.5.4
Description of change		Deletion of the wording, linking the impossibility of on-site monitoring to the discontinuation of the study
Rationale for change		The wording does not adequately reflect the present pandemic situation. Measures for remote monitoring are in place. The safety and wellbeing of subjects, subject's rights and data integrity is not compromised by this change.

11.6 GLOBAL AMENDMENT 6

Date of amendment		18 Feb 2021
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)
To be implemented only after approval of the IRB/ IEC/ Competent Authorities		☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval		☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only		☐
Section to be changed		Flow Chart – SRD part; Flow Chart – BA/ FE part; Section 5.2.6.4; Section 5.3.1; Throughout the document.
Description of change		Insertion of the 0:05 (08:15) PK sampling time point Shift of the 2:15 (10:15) PK sampling time point to 2:30 (10:30) Deletion of the 10:00 (18:00) PK sampling time point; Insertion of Footnote 15 (Sampling times and periods may be adapted based on information obtained during the trial …);

		Description of the adjudication process at [redacted], if two readers have deviating opinions on EEGs; inclusion of outcome categories ('normal', 'abnormal/ epileptiform', 'abnormal/ non-epileptiform'); BA/FE-part: Permission of flexibility concerning PK sampling time points and -periods based on information obtained during the course of the trial; Minor editorial and formatting changes for consistency reasons.
Rationale for change		Minor modification of the PK sampling scheme, based on the experience gained so fare. This change does not change the total volume of blood being sampled during the course of this trial; Description of an established reporting process at [redacted] Optimisation of the scientific value of the study, by allowing, also during the BA/ FE-part, to modify PK-sampling time points within the stipulated limits, without having to submit a substantial amendment; Minor editorial and formatting changes for consistency reasons.

11.7 GLOBAL AMENDMENT 7

Date of amendment		14-May-2021
EudraCT number		2019-004836-51
BI Trial number		1447-0001
BI Investigational Medicinal Product(s)		BI 1569912
Title of protocol		Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)

To be implemented only after approval of the IRB/ IEC/ Competent Authorities	☒
To be implemented immediately in order to eliminate hazard – IRB/ IEC/ Competent Authority to be notified of change with request for approval	☐
Can be implemented without IRB/ IEC/ Competent Authority approval as changes involve logistical or administrative aspects only	☐

Section to be changed		Flow Chart – Single Rising Dose Part Section 1.2.3.2 Section 1.2.6 Section 1.3.2.1 Section 1.4.7 Section 4.1.5.1 Section 4.2.2.2 Section 5.2.4.1 Section 6.1 Section 7.3 Section 7.3.1 Section 7.3.2 Section 7.3.4 Section 7.3.5 Section 9.1 Section 9.2 Throughout the document
Description of change		Flow Chart – Single Rising Dose Part: Inclusion of triplicate ECGs on Day1; removal of the light breakfast at 2 hours post dose on Day1.

		Section 1.2.3.2: Inclusion of toxicological date from the dog 'dose range finding' and the dog 'dose escalation' study (1.8 mg/ kg/ day). Section 1.2.6: Inclusion of human date of dose groups 1 to 6 (study 1447-0001). Section 1.3.2.1: Rational for higher exposure limits (stopping rule). Section 1.4.7: Dose escalation rules (stopping rules) adapted to new exposure limits. Section 4.1.5.1: Blinding of the central ECG lab staff. Section 4.2.2.2: Removal of the light breakfast at 2 hours post dose on Day1. Section 5.2.4.1: Inclusion of triplicate ECGs. Section 6.1: Sequence of procedures was accommodated to enable best ECG-quality. Section 7.3: Reference to SOP 001-MCS-36-472 was deleted. Section 7.3.1: Reference to SOP 001-MCS-36-472 was deleted. Section 7.3.2: Reference to SOP 001-MCS-36-472 was deleted. Section 7.3.4: Evaluation of ECG data specified. Section 7.3.5: Evaluation of ECG data specified. Section 9.1: Referenced added. Section 9.2: Referenced added. Throughout the document: Minor editorial and formatting changes.
Rationale for change		Inclusion of triplicate ECGs, to generate data that would justify a waver for a dedicated thorough QT-

		study with BI 1569912. [redacted] Minor editorial and formatting changes for consistency reasons.

APPROVAL / SIGNATURE PAGE

Document Number: c30179584 **Technical Version Number:8.0**

Document Name: clinical-trial-protocol-version-08

Title: Safety, tolerability, pharmacokinetics and pharmacodynamics of single rising oral doses of BI 1569912 in healthy male subjects (single-blind, partially randomized within dose groups, placebo-controlled, parallel-group design) with an additional relative bioavailability/ food effect part (open-label, randomized, three-way crossover design)

Signatures (obtained electronically)

Meaning of Signature	Signed by	Date Signed
Author-Clinical Trial Leader	[redacted]	14 May 2021 16:02 CEST
Approval-Clinical Program Leaders		16 May 2021 17:06 CEST
Author-Trial Statistician		17 May 2021 15:43 CEST
Verification-Paper Signature Completion		19 May 2021 14:58 CEST

(Continued) Signatures (obtained electronically)

Meaning of Signature	Signed by	Date Signed