Clinical Trial Protocol

	Document Number: **c40616508-02**
EudraCT No.	2021-006676-17
BI Trial No.	1346-0056
BI Investigational Medicinal Products	BI 425809 (iclepertin), Tracleer® (bosentan)
Title	The effect of multiple oral doses of bosentan on the steady state kinetics of BI 425809 after oral administration to healthy male subjects (an open-label, two-period fixed sequence trial)
Lay Title	A study in healthy men to test whether bosentan influences the amount of BI 425809 in the blood
Clinical Phase	I
Clinical Trial Leader	[redacted] Phone: [redacted] Fax: [redacted]
[redacted] **Investigator**	[redacted] Phone: [redacted] Fax: [redacted]
Current Version, Date	Version 2.0, 01 Feb 2023
Original Protocol Date	13 Dec 2022

CLINICAL TRIAL PROTOCOL SYNOPSIS

Company name	Boehringer Ingelheim
Original protocol date	13 December 2022
Revision date	Not applicable
BI trial number	1346-0056
Title of trial	The effect of multiple oral doses of bosentan on the steady state kinetics of BI 425809 after oral administration to healthy male subjects (an open-label, two-period fixed sequence trial)
[redacted] **Investigator**	[redacted]
Trial site	[redacted]
Clinical phase	I
Trial rationale	BI 425809 is a substrate of CYP3A4. Rifampicin, a strong inducer of 3A4, has reduced the bioavailability of BI 425809 by 90%. This trial should investigate the effect of bosentan, a moderate inducer of CYP3A4, on the kinetics of BI 425809.
Trial objective	The main objective of this trial is to investigate the relative bioavailability of BI 425809 given alone (Reference) compared to a combined administration with the moderate CYP3A4 inducer bosentan (Test) following repeated oral administration.
Trial endpoints	Primary endpoints: $AUC_{\tau,ss}$ and $C_{max,ss}$ of BI 425809 Secondary endpoints: $C_{min,ss}$ of BI 425809
Trial design	open-label, two-period fixed sequence design
Number of subjects **total entered** **on each treatment**	 14 14
Diagnosis	Not applicable
Main inclusion criteria	Healthy male subjects, age of 30 to 55 years (inclusive), body mass index (BMI) of 18.5 to 29.9 kg/m^2 (inclusive)
Trial product 1 **dose** **mode of administration**	BI 425809, 10 mg film-coated tablets 1 x 1 tablet (= daily dose of 10 mg BI 425809) Oral with 240 mL of water

Trial product 2	Tracleer®, 125 mg Filmtabletten (1 film tablet contains 125 mg bosentan)
dose	2 x 1 tablet Tracleer® (daily dose of 250 mg bosentan)
mode of admin.	Oral with 240 mL of water
Duration of treatment	Treatment Reference: 1 x 1 tablet BI 425809 daily for 10 days Treatment Test: 2 x 1 tablet Tracleer® plus 1 x 1 tablet BI 425809 for 14 days Treatment Test will follow immediately after Treatment Reference (total treatment duration of 24 days).
Statistical methods	Relative bioavailability will be estimated by the ratios of the geometric means (test/reference) for the primary 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 period and treatment. CIs will be calculated based on the residual error from the ANOVA. Descriptive statistics will be calculated for all endpoints.

FLOW CHART

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[9]	PK $_{blood}$ BI 425809	PK $_{blood}$ bosentan	12-lead ECG	Vital signs (BP, PR)	Questioning for AEs and concomitant therapy [6]
SCR	1	-21 to -1			Screening (SCR)[1]	A			x	x	
Reference period (Period 1)	2	1	0:00	08:00	Allocation of subject numbers, BI 425809 administration (ambulatory)	B[2]	x[2]		x[2]	x[2]	x[2]
		2	24:00	08:00	BI 425809 administration (ambulatory)						x
		3	48:00	08:00	BI 425809 administration (ambulatory)						x
		4	72:00	08:00	BI 425809 administration (ambulatory)						x
		5	96:00	08:00	BI 425809 administration (ambulatory)						x
		6	120:00	08:00	BI 425809 administration (ambulatory)						x
		7	144:00	08:00	BI 425809 administration (ambulatory)						x
		8	168:00	08:00	BI 425809 administration (ambulatory)						x
		9	192:00	08:00	BI 425809 administration (ambulatory)		x[7]				x
			204:00	20:00	Admission to trial site[8]	x[5]					x
		10	216:00	08:00	BI 425809 administration	B[2]	x[7]			x[2]	x
			216:30	08:30			x				
			217:00	09:00			x				
			217:30	09:30			x				
			218:00	10:00	240 mL fluid intake		x				x
			218:30	10:30			x				
			219:00	11:00	Suicidality assessment		x				
			219:30	11:30			x				
			220:00	12:00	240 mL fluid intake		x				
			220:30	12:30	Lunch[3]		x				x
			222:00	14:00			x				
			224:00	16:00	Snack (voluntary)[3]		x				
			226:00	18:00			x				
			227:00	19:00	Dinner						
			228:00	20:00			x				x
		11	240:00	08:00			x				x
Test period (period 2)	3	1	0:00	09:00	BI 425809 + bosentan administration, thereafter breakfast			x[2]	x[2]	x[2]	x[2]
			3:30	12:30	lunch						
			7:00	16:00	Snack (voluntary)[3]						
			10:00	19:00	Dinner						
			11:00	20:00	bosentan administration						x
		2	22:00	07:00	BI 425809 + bosentan administration, breakfast (voluntary)[3] and discharge					x[2]	x[2]
			34:00	19:00	bosentan administration						x
		3	46:00	07:00	BI 425809 + bosentan administration						x
			58:00	19:00	bosentan administration						x
		4	70:00	07:00	BI 425809 + bosentan administration	B[2]					x
			82:00	19:00	bosentan administration						x
		5	94:00	07:00	BI 425809 + bosentan administration					x[2]	x[2]
			106:00	19:00	bosentan administration						x

FLOW CHART, continued

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[9]	PK blood BI 425809	PK blood bosentan	12-lead ECG	Vital signs (BP, PR)	Questioning for AEs and concomitant therapy [6]
Test period (period 2)	3	6	118:00	07:00	BI 425809 + bosentan administration						x
			130:00	19:00	bosentan administration						x
		7	142:00	07:00	BI 425809 + bosentan administration	B[2]	x[2]	x[2]	x[2]	x[2]	x[2]
			154:00	19:00	bosentan administration						x
		8	166:00	07:00	BI 425809 + bosentan administration						x
			178:00	19:00	bosentan administration						x
		9	190:00	07:00	BI 425809 + bosentan administration						x
			202:00	19:00	bosentan administration						x
		10	214:00	07:00	BI 425809 + bosentan administration						x
			226:00	19:00	bosentan administration						x
		11	238:00	07:00	BI 425809 + bosentan administration	B[2]					x
			250:00	19:00	bosentan administration						x
		12	262:00	07:00	BI 425809 + bosentan administration						x
			274:00	19:00	bosentan administration						x
		13	287:00	08:00	BI 425809 + bosentan administration						x
			299:00	20:00	Admission to trial site[8]	x[5]					x
			300:00	21:00	bosentan administration						x
		14	311:00	08:00	BI 425809 administration	B[2]	x[7]	x[7]	x[2]	x[2]	x[2]
			311:30	08:30			x				
			312:00	09:00	bosentan administration (after PK)		x				x
			312:30	09:30			x				
			313:00	10:00	240 mL fluid intake		x				
			313:30	10:30			x				
			314:00	11:00	Suicidality Assessment		x				
			314:30	11:30			x				
			315:00	12:00	240 mL fluid intake		x				
			315:30	12:30	Lunch[3]		x				x
			317:00	14:00			x				
			319:00	16:00	Snack (voluntary)[3]		x				
			321:00	18:00			x				
			322:00	19:00	Dinner						
			323:00	20:00	bosentan administration (after PK)		x	x			x
		15	335:00	08:00	Breakfast[3] (voluntary) and discharge		x	x		x	x
FU	4	25-38			End of study (EoS) examination[4]	C			x	x	x

1. Subject must be informed and written informed consent obtained prior to starting any screening procedures. Screening procedures include physical examination, check of vital signs, ECG, safety laboratory (including drug screening), demographics (including determination of body height and weight, smoking status and alcohol history), relevant medical history, suicidality assessment, concomitant therapy and review of inclusion/exclusion criteria.
2. The time is approximate; the procedure is to be performed and completed within the 2 h prior to drug administration.
3. If several actions are indicated at the same time, the intake of meals will be the last action.
4. At the end of study (synonym for end of trial), the EoS examination includes physical examination, vital signs, ECG, safety laboratory, suicidality assessment, 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. blood sampling to be done within 10 min prior to dosing

8. The time is an approximate. The procedure is to be completed not later than 10 hours prior to BI drug administration.
9. Letters A, B and C define different sets of safety laboratory examinations (for details refer to Section 5.2.3).

Tolerance time for drug dosing on Days 1-8 (period 1) and 1-12 (period 2) is ± 1 hour.

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ABBREVIATIONS AND DEFINITIONS

ADME	Absorption, distribution, metabolism, and excretion
AE	Adverse event
AESI	Adverse events of special interest
ALCOA	attributable, legible, contemporaneous, original, and accurate
ANOVA	Analysis of variance
AUC	Area under the concentration-time curve of the analyte in plasma
AUC0-∞	Area under the concentration-time curve of the analyte in plasma over the time interval from 0 extrapolated to infinity
AUC0-tz	Area under the concentration-time curve of the analyte in plasma over the time interval from 0 to the last quantifiable data point
$AUC_{\tau,ss}$	Area under the concentration-time curve of the analyte in plasma at steady state over a uniform dosing interval τ
BA	Bioavailability
BI	Boehringer Ingelheim
BMI	Body mass index (weight divided by height squared)
BP	Blood pressure
CI	Confidence interval
CIAS	Cognitive impairment associated with schizophrenia
CL	Total clearance of the analyte in plasma after intravascular administration
Cmax,ss	Maximum measured concentration of the analyte in plasma in steady state
Cmin,ss	Minimum measured concentration of the analyte in plasma (at steady state)
CNS	Central Nervous System
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
DILI	Drug induced liver injury
ECG	Electrocardiogram
eCRF	Electronic case report form
eDC	Electronic data capture
EDTA	Ethylenediaminetetraacetic acid
EudraCT	European Clinical Trials Database
GCP	Good Clinical Practice
gCV	Geometric coefficient of variation
GlyT1	glycine transporter 1

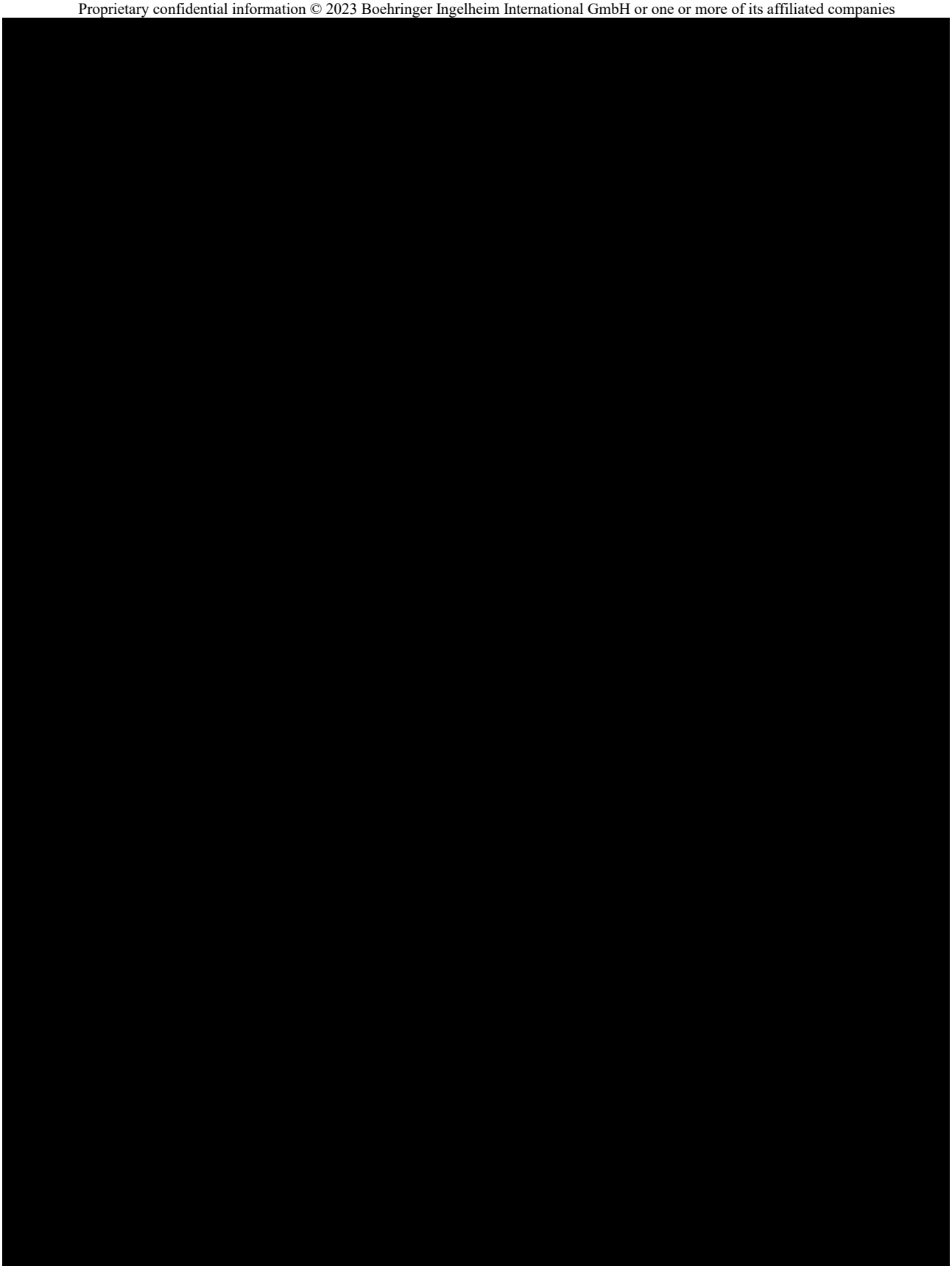
[redacted]

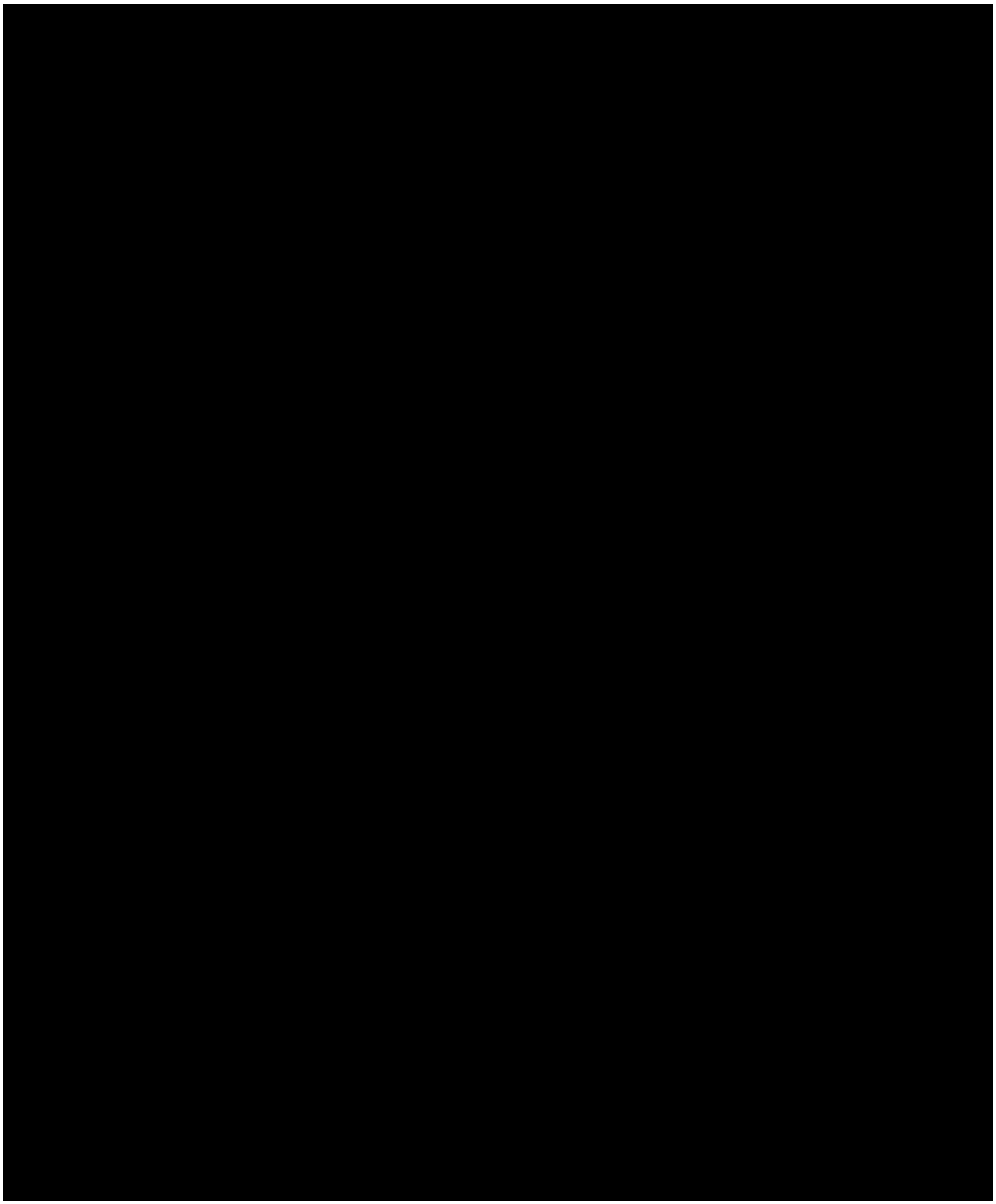
IB	Investigator's brochure
IEC	Independent Ethics Committee
IPD	Important protocol deviation
IRB	Institutional Review Board
ISF	Investigator site file
[redacted]	[redacted]
MDA	Methylenedioxyamphetamine
MDMA	Methylenedioxymethamphetamine
MedDRA	Medical Dictionary for Regulatory Activities
NMDA	N-methyl-D-aspartate
PD	Pharmacodynamic(s)
PK	Pharmacokinetic(s)
PKS	Pharmacokinetic set
PR	Pulse rate
qd	once daily
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) or Bazett (QTcB)
R	Reference treatment
REP	Residual effect period
SAE	Serious adverse event
SCR	Screening
SmPC	Summary of Product Characteristics
SOP	Standard operating procedure
T	Test product or treatment
TBA	Trial bioanalyst
tmax	Time from (last) dosing to the maximum measured concentration of the analyte in plasma
TSAP	Trial statistical analysis plan
ULN	Upper limit of normal
WOCBP	women of childbearing potential

1. INTRODUCTION

BI 425809 is a glycine transporter 1 (GlyT1) inhibitor under development for treatment of cognitive impairment associated with schizophrenia (CIAS).

1.2 DRUG PROFILE





1.2.2 Bosentan

1.2.2.1 Drug profile

Bosentan is an endothelin receptor antagonist used for the treatment of pulmonary arterial hypertension (PAH) and for prevention of digital ulcers in patients with systemic sclerosis. The therapeutic dose is 125 mg given twice daily.

The absolute bioavailability of bosentan in healthy subjects is about 50%. After oral dosing maximum plasma concentrations will be reached after 3-5 hours. After hepatic metabolism via CYP3A4 and CYP2C9 the drug is eliminated via bile. The terminal elimination half-life is 5.4 hours. Bosentan is known to be an inducer of CYP3A4 and CYP2C9. After multiple doses plasma concentrations reduce by 35-50% which is probably caused by autoinduction of its metabolism. Steady state conditions are reached after 3-5 days [R22-4121].

Chronic treatment with bosentan may cause an increase of liver transaminases and decreased haemoglobin values. After treatment of about 2500 patients with daily doses of 100 mg – 2000 mg (mean treatment duration: 45 weeks) the most frequent adverse events comprised headache, oedema (fluid retention), changed liver values and anaemia/decreased haemoglobin value [R15-5307].

In pre-clinical trials bosentan has been demonstrated to be teratogen. Therefore, no women will be included into this trial. Furthermore, bosentan may influence male fertility. Concentration of spermatocytes was reduced by at least 50% in 6 of 24 patients after treatment with bosentan for 6 months [R22-4121].

For a more detailed description of the bosentan profile, please refer to the current SmPC [R22-4121]

1.2.2.2 Clinical safety in healthy subjects

In the MRD study doses of 100 mg, 200 mg, 500 mg and 1000 mg were given once daily for 8 days (Part A). Headache and head discomfort was the most frequent adverse event which was reported by 4 of 6 subjects in the 100 mg, 200 mg and 500 mg dose group (DG) and by 3 of 6 subjects in the 1000 mg DG. Headache typically occurred on the first dosing day and reoccurred on different trail days for some subjects. It was always assessed to be possibly or probably related to trial medication. Headache was of mild intensity with the exception of 1 moderate case in the 1000 mg dose group. Further AEs comprise gastrointestinal and respiratory disorders (no statement on relationship). Compared with placebo bosentan caused a reduction of standing blood pressure (systolic + diastolic) of 5-10 mmHg. A specific pattern of lab value changes has not been observed in the MRD study [R22-4129].

Bosentan has been administered to healthy subjects in several drug-drug-interaction (DDI) studies. In the first reported interaction trials with the P-gp substrate digoxin (Weber 1999, R15-5309) and the 2C9 substrate warfarin [R22-4130] high bosentan doses of 500 mg bid have been used. After combined administration of digoxin and bosentan over 7 days, mild to moderate headache was the most frequently reported adverse event. There was no pattern of change in any clinical laboratory parameters or vital signs [R15-5309]. In the warfarin interaction study 500 mg bosentan twice daily were given for 10 days. No statement on tolerability is given in this paper [R22-4130].

In DDI trials performed later, bosentan had been dosed 125 mg twice daily, which is the standard therapeutic maintenance dose. In general, it was well tolerated in these studies:

Tadalafil: Wrishko et al investigated the interaction with tadalafil. In this 3-period cross-over trial the 14 subjects received 40 mg tadalafil once daily for 10 days, 125 mg bosentan twice daily for 10 days and a combination of tadalafil and bosentan for 10 days. The treatment emergent adverse events in the bosentan alone period comprised headache (reported by 3 subjects), postural dizziness (3), fatigue (2), myalgia (2), flushing (1), nasal congestion (1), ocular hyperaemia (1) and pain in extremity (1) [R22-4132].

Glyburide: Investigating the interaction with glyburide, bosentan 125 mg bid was given for 10 days (treatment A: + glyburide 2.5 mg bid on day 6-10) or 5 days (treatment B: glyburide 2.5 mg bid for 10 days + bosentan co-medication on day 6-10). Reported adverse events comprise 2 cases of headache (mild, moderate), fatigue (mild), abdominal discomfort (mild) and 2 cases of elevated liver enzymes (AST: up to 2 x ULN, ALT: up to 9 x ULN). Both cases occurred on day 10 of the first treatment period, which was treatment B in the case of the 9fold ALT increase. Both cases resolved during the wash-out period. During the second treatment period, these subjects did not show increased liver enzymes [R11-0555]. The increased liver transaminases reported by van Giersbergen et al occurred at the end of combined administration of glyburide and bosentan. Both compounds are known to cause increased liver values. This may have contributed to the extraordinary finding in one subject.

Lopinavir/Ritonavir: The interaction between bosentan and lopinavir/ritonavir was investigated in a 3 period cross-over trial. The healthy male participants received 125 mg bosentan twice daily for 9.5 days, 400 mg lopinavir/100 mg ritonavir for 9.5 days and a combination of both treatments for 9.5 days. In the bosentan alone period adverse events were reported by 4 of 10 participants (mainly mild headache). Changes in lab parameters did not show treatment related patterns, specifically no increase in liver transaminases were observed during the study. While blood pressure was decreased in all treatment groups, there was no case of clinically relevant hypotension [R14-1160]

Simvastatin: To investigate the interaction of bosentan and simvastatin, 125 mg bosentan was given twice daily for 5.5 days. Mild to moderate headache was the most frequently reported adverse event. No pattern of abnormal laboratory values or vital signs was detected during the study that might suggest a treatment effect [P03-08686].

Treprostinil: Gotzkowsky et al investigated the interaction with treprostinil. In this 3-period cross-over trial the subjects received 1 mg treprostinil twice daily, 125 mg bosentan twice daily and a combination of both drugs for 4.5 days each. The treatment emergent adverse events in the bosentan alone period (N=23) comprised headache (reported by 2 subjects), dizziness (2), pharyngolaryngeal pain (2), constipation (1) and dysmenorrhea (1). There were no clinically relevant changes in lab parameters or vital signs. [R22-4131].

[redacted]

[redacted]

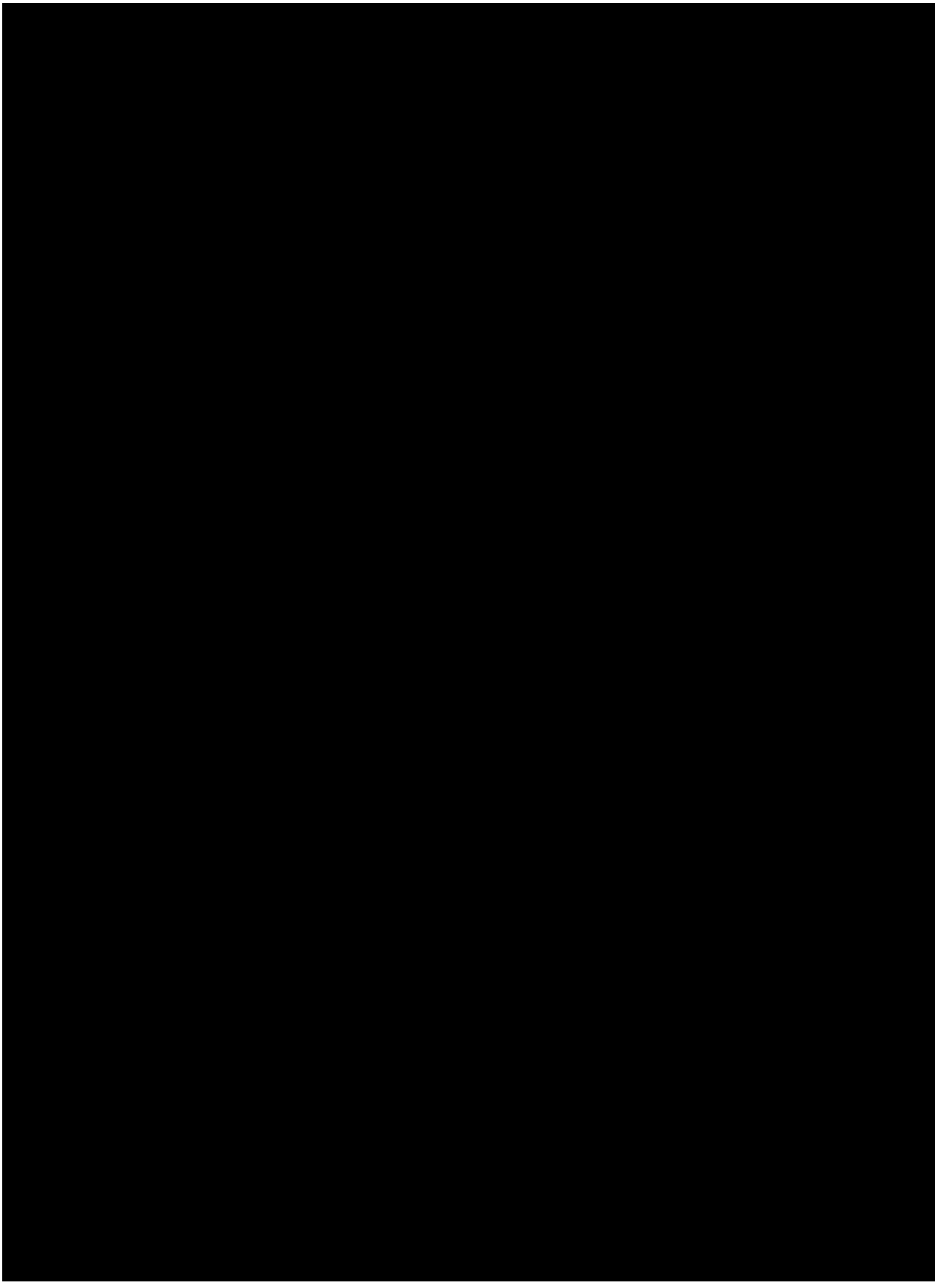
1.4 BENEFIT - RISK ASSESSMENT

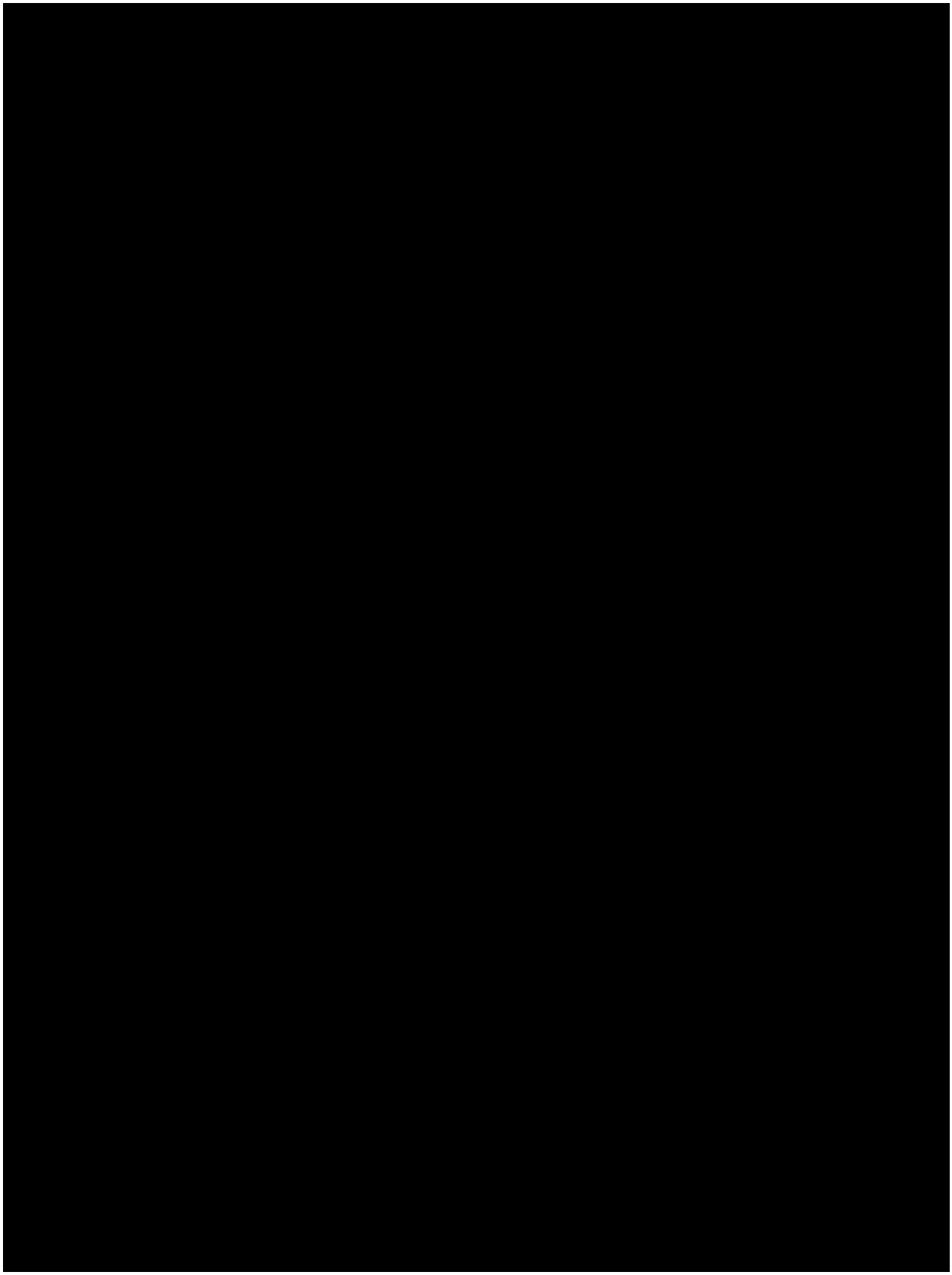
[redacted]

1.4.2 Risks

Subjects are exposed to risks of trial procedures and risks related to the exposure to the trial medication. An overview of trial-related risks is given in Table 1.4.2: 1.

[redacted]





The total volume of blood withdrawn per subject during the entire trial will not exceed 250 ml which is half of the volume of a normal blood donation. No health-related risk to healthy subjects is expected from withdrawal of this volume of blood.

2. TRIAL OBJECTIVES AND ENDPOINTS

2.1 MAIN OBJECTIVES, PRIMARY AND SECONDARY ENDPOINTS

2.1.1 Main objectives

The main objective of this trial is to investigate the relative bioavailability of BI 425809 given alone (Reference) compared to a combined administration with the moderate CYP3A4 inducer bosentan (Test) following repeated oral administration.

2.1.2 Primary endpoints

The following pharmacokinetic parameters will be determined for BI 425809:

- $AUC_{\tau,ss}$ (area under the concentration-time curve of the analyte in plasma at steady state over a uniform dosing interval τ)
- $C_{max,ss}$ (maximum measured concentration of the analyte in plasma at steady state over a uniform dosing interval τ)

2.1.3 Secondary endpoint

The following pharmacokinetic parameter will be determined for BI 425809:

- $C_{min,ss}$ (minimum concentration of the analyte in plasma at steady state within a uniform dosing interval τ)

[redacted]

[redacted]

2.2.2.2 Safety and tolerability

Safety and tolerability of BI 425809 will be assessed based on:

- Adverse events (including clinically relevant findings from the physical examination)
- [redacted]
- Safety laboratory tests
- 12-lead ECG
- Vital signs (blood pressure, pulse rate)

3. DESCRIPTION OF DESIGN AND TRIAL POPULATION

3.1 OVERALL TRIAL DESIGN

The trial will be performed as a non-randomised, open-label, 2 period fixed sequence trial in healthy male subjects in order to compare the test treatment (T) to the reference treatment (R). The treatments will be

Period 1 (R): 1 x 1 tablet BI 425809 10 mg for 10 days

Period 2 (T): 2 x 1 tablet Tracleer® 125 mg plus 1 x 1 tablet BI 425809 10 mg for 14 days

For details on drug administration, refer to Section 4.1.

There will be no washout period between the treatments. Period 2 will immediately follow Period 1 to maintain steady state conditions of BI 425809 (i.e. Day 1 of Period 2 will follow directly Day 10 of Period 1).

An overview of all relevant trial activities is provided in the Flow Chart. 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 GROUP

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

In mechanistic DDI trials the victim compound usually is administered single dose. In this case the offender drug has to be given also in the elimination phase of the victim drug (if the effect on drug metabolising enzymes is studied). In 1346-0056 BI 425809 is the victim compound and the moderate 3A4 inducer bosentan is the offender drug.

The open-label treatment is not expected to bias results, since the trial endpoints are derived from measurement of plasma concentrations of the analyte, that are not influenced by the subject`s knowledge of the treatment.

3.3 SELECTION OF TRIAL POPULATION

It is planned that 14 healthy male subjects will enter the trial. They will be recruited from the volunteers' pool of the trial site.

Female subjects will not be included into the trial due to the teratogenic potential of bosentan.

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 trial will be performed in healthy subjects.

Please refer to Section 8.3.1 (Source Documents) for the documentation requirements pertaining to the in- and exclusion criteria.

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), 12-lead ECG, and clinical laboratory tests
2. Age of 30 to 55 years (inclusive)
3. BMI of 18.5 to 29.9 kg/m^2 (inclusive)
4. Signed and dated written informed consent in accordance with ICH-GCP and local legislation prior to admission to the trial

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 45 to 90 bpm
3. Any laboratory value outside the reference range that the investigator considers to be of clinical relevance
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 or stroke), and other relevant neurological or psychiatric disorders

8. History of relevant orthostatic hypotension, fainting spells, or blackouts
9. Relevant chronic or acute infections
10. Any documented active or suspected malignancy or history of malignancy within 5 years prior to screening, except appropriately treated basal cell carcinoma of the skin
11. History of relevant allergy or hypersensitivity (including allergy to the trial medication or its excipients)
12. Use of drugs and any kind of vaccination within 30 days of planned administration of trial medication that might reasonably influence the results of the trial (including drugs that cause QT/QTc interval prolongation)
13. Intake of an investigational drug in another clinical trial within 60 days of planned administration of investigational drug in the current trial, or concurrent participation in another clinical trial in which investigational drug is administered
14. Smoker (more than 10 cigarettes or 3 cigars or 3 pipes per day)
15. Inability to refrain from smoking on specified trial days
16. Alcohol abuse (consumption of more than 24 g per day for males)
17. Drug abuse or positive drug screening
18. Blood donation of more than 100 mL within 30 days of planned administration of trial medication or intended blood donation during the trial
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 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
24. During COVID-19 pandemic: laboratory test indicative of an ongoing SARS-CoV-2 infection
25. Male subjects with WOCBP partner who are unwilling to use male contraception (condom or sexual abstinence) from time point of administration of trial medication until 30 days thereafter

[redacted]

27. Liver enzymes (ALT, AST, GGT) or serum creatinine above upper limit of normal range at screening examination, confirmed by a repeat test

For restrictions of the trial, refer to Section 4.2.2.

3.3.4 Withdrawal of subjects from treatment or assessments

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

If a subject is removed from 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 from 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, trial data will be included in the CRF and will be reported in the CTR.

Following removal or withdrawal after first study drug administration, a complete end-of-trial examination should be performed. If the discontinuation or withdrawal occurs before the end of the REP (see Section 1.2.3 , 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 Withdrawal from trial treatment

An individual subject will be withdrawn from trial treatment if:

1. The subject wants to withdraw from trial treatment. The subject will be asked to explain the reasons but has the right to refuse to answer
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, the safety of the subject cannot be guaranteed as he / she 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 (surgery, adverse events, or diseases)
5. 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.

If new efficacy or safety information becomes available, Boehringer Ingelheim will review the benefit-risk-assessment and, if needed, pause or discontinue the trial treatment for all subjects or take any other appropriate action to guarantee the safety of the trial subjects.

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, please see 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 (if reasons 4 and/or 5 are met, the trial should be discontinued immediately):

1. Failure to meet expected enrolment goals overall or at a particular trial site
2. The sponsor decides to discontinue the further development of the investigational products
3. Deviation from GCP, or the CTP impairing the appropriate conduct of the trial
4. New toxicological findings, serious adverse events, or any safety information invalidating the earlier positive benefit-risk-assessment (see Section 3.3.4.1)
5. More than 50% of the subjects show drug-related and clinically relevant adverse events of moderate or severe intensity, or if at least one drug-related serious adverse event is reported

3.3.5 Replacement of subjects

In case more than 2 subjects do not complete the trial (including subjects non-evaluable for PK), subjects may be replaced if considered necessary to reach the objective of the trial. The Clinical Trial Leader together with the Trial Pharmacokineticist and the Trial Statistician are to decide, if and how many subjects will be replaced. A replacement subject will be assigned a unique trial subject number, and will receive both treatments.

4. TREATMENTS

4.1 INVESTIGATIONAL TREATMENTS

BI 425809 has been manufactured by BI Pharma GmbH & Co, Germany.

Tracleer® has a EU market authorisation and is obtained from a public pharmacy.

4.1.1 Identity of the Investigational Medicinal Products

The characteristics of the trial product 1 are given below:

Substance:	BI 425809 (iclepertin)
Pharmaceutical formulation:	Film-coated tablet
Source:	BI Pharma GmbH & Co. KG, Germany
Unit strength:	10 mg
Posology:	1 – 0 – 0
Route of administration:	Oral
Duration of use:	24 days qd (once daily in period 1 and 2)

The characteristics of the trial product 2 are given below:

Name:	Tracleer®
Substance:	bosentan
Pharmaceutical formulation:	Film-coated tablet
Source:	[redacted]
Unit strength:	125 mg
Posology:	1 – 0 – 1
Mode of administration:	Oral
Duration of use:	14 days bid (twice daily in period 2 only)

4.1.2 Selection of doses in the trial

The dose of 10 mg BI 425809 is the expected therapeutic dose that is currently used in clinical trials.

The dose of 125 mg bosentan given twice daily is the standard clinical dose [R22-4121].

4.1.3 Method of assigning subjects to treatment groups

There is only one treatment sequence investigated in this trial, and each subject will be allocated to the same treatment sequence (R-T). The subjects will be allocated to a trial subject number by drawing lots prior to first administration of trial medication in the morning of Day 1 of Visit 2.

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

All subjects may be treated in one cohort, i.e. all subjects may receive treatment on the same calendar day. In case this is not feasible (e.g., due to logistical or recruitment reasons), the group may be split into several cohorts as required. Treatment of all subjects on the same calendar day is acceptable (for discussion of trial-associated risks and safety measures, see Section 1.4).

4.1.4 Drug assignment and administration of doses for each subject

This is a one-way crossover trial. All subjects will receive the 2 treatments in a fixed sequence. The treatments to be evaluated are summarised in Table 4.1.4: 1 below.

[redacted]

On PK-profile days (period 1/Day 10 and period 2/Day 14) administration of trial medication will be performed after subjects have fasted overnight; fasting is to start no later than 10 h before the scheduled first dosing. On all other dosing days fasting status is not required.

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. During the first 4 h after morning 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).

For all drug administrations, 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.

Dosing of BI 425809 and bosentan is performed in an ambulatory fashion throughout the trial with the exception of PK-profile days and Day 1 of period 2 (on this day subjects will be kept under close medical surveillance to monitor the tolerability of first bosentan doses).

There is no specific wash-out period in this trial. BI 425809 will be continuously dosed from the start of period 1 until the end of period 2 to maintain the steady state conditions.

4.1.5 Blinding and procedures for unblinding

This non-randomised open-label Phase I trial will be handled in an open fashion throughout. The treatment assignment will be available to all involved parties.

4.1.6 Packaging, labelling, and re-supply

BI 425809 tablets will be provided by BI. They will be packaged and labelled in accordance with the principles of Good Manufacturing Practice (GMP). For details of packing and the description of the label, refer to the ISF.

Bosentan will be obtained by the clinical trial site from a public pharmacy. The drug will be dispensed out of the original, unmodified packages.

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.

No re-supply is planned.

4.1.7 Storage conditions

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 Clinical Research Associate (as provided in the list of contacts) is to be contacted immediately.

4.1.8 Drug accountability

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

- Approval of the clinical trial protocol by the IRB / ethics committee
- 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 form 'Trial Staff List' may dispense investigational drugs to trial subjects. Investigational drugs are not allowed to be used outside of this protocol.

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 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, 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 trial days) on the appropriate pages of the CRF.

4.2.2.2 Restrictions on diet and life style

While admitted to the trial site, the 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.

On PK-profile days from 1 h before morning drug intake until lunch, fluid intake is restricted to the water administered with the drug, and an additional 240 mL of water at 2 h and 4 h post-dose (mandatory for all subjects). From lunch until 12 h post-dose, total fluid intake is restricted to 2000 mL (thereafter subjects may drink ad libidum).

Green tea, 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 PK sampling of the trial.

Alcoholic beverages are not permitted starting 48 h before first trial drug administration until last PK sampling.

Methylxanthine-containing drinks or foods (such as coffee, tea, cola, energy drinks, or chocolate) are not allowed during in-house confinement at the trial site.

Smoking is not allowed during in-house confinement at 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 end of trial examination.

4.3 TREATMENT COMPLIANCE

Compliance will be assured by administration of all trial medication in the trial centre under supervision of the investigating physician or a designee. The measured plasma concentrations 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, please see Section 3.3.4.1).

5. ASSESSMENTS

5.1 ASSESSMENT OF EFFICACY

Not applicable.

5.2 ASSESSMENT OF SAFETY

5.2.1 Physical examination

At screening, the medical examination will include demographics, height and body weight, smoking and alcohol history (alcohol history 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), 12-lead ECG, laboratory tests, and a physical examination, and assessment of suicidal ideation and behavior using the C-SSRS ('baseline/screening scale'). At the end of trial examination, it will include review of vital signs, 12-lead ECG, laboratory tests, and a physical examination, and assessment of suicidal ideation and behaviour using C-SSRS ('since last visit scale').

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 (Dinamap Pro 100, [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 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 lab panel A and C; for lab panel B no specific fasting period is required). For retests, at the discretion of the investigator or designee, overnight fasting is not required.

The parameters to be assessed are listed in Tables 5.2.3: 1 and 5.2.3: 2. Reference ranges will be provided in the ISF.

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 in the urinalysis, respectively.

Table 5.2.3: 1 Routine laboratory tests

Functional lab group	BI test name [comment/abbreviation]	A[1]	B[1]	C[1]
Haematology	Haematocrit	X	X	X
	Haemoglobin	X	X	X
	Red Blood Cell Count/Erythrocytes	X	X	X
	Reticulocytes, absol.	X	X	X
	Reticulocytes/Erythrocyte	X	X	X
	White Blood Cells/Leucocytes	X	X	X
	Platelet Count/Thrombocytes (quant)	X	X	X
Automatic WBC differential, relative	Neutrophils/Leukocytes; Eosinophils/Leukocytes; Basophils/ Leukocytes; Monocytes/Leukocytes; Lymphocytes/Leukocytes	X	X	X
Automatic WBC differential, absolute	Neutrophil, absol.; Eosinophils, absol.; Basophils, absol.; Monocytes, absol.; Lymphocytes, absol.	X	X	X
Manual differential WBC (if automatic differential WBC is abnormal)	Neut. Poly (segs)/Leukocytes; Neut. Poly (segs), absol.; Neutrophils Bands/Leukocytes; Neutrophils Bands, absol.; Eosinophils/Leukocytes; Eosinophils, absol.; Basophils/ Leukocytes; Basophils, absol.; Monocytes/Leukocytes; Monocytes, absol.; Lymphocytes/Leukocytes; Lymphocytes, absol.			
Coagulation	Activated Partial Thromboplastin Time	X	--	X
	Prothrombin time (Quick)	X	--	X
	Prothrombin time – INR (International Normalization Ratio)	X	--	X
Enzymes	AST [Aspartate aminotransferase] /GOT, SGOT	X	X	X
	ALT [Alanine aminotransferase] /GPT, SGPT	X	X	X
	Alkaline Phosphatase	X	--	X
	Gamma-Glutamyl Transferase	X	X	X
Hormones	Thyroid Stimulating Hormone	X	--	--
Substrates	Glucose (Plasma)	X	--	X
	Creatinine	X	--	X
	Bilirubin, Total	X	--	X
	Bilirubin, Direct	X	--	X
	C-Reactive Protein (Quant)	X	--	X
Electrolytes	Sodium	X	--	X
	Potassium	X	--	X
	Calcium	X	--	X
Urinalysis[2] (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[2] (microscopic examination)	Only positive findings will be reported (for instance, the presence of sediment bacteria, casts in sediment, squamous epithelial cells, erythrocytes, leukocytes)			

1 A: Parameters to be determined at Visit 1 (screening examination)
B: Parameters to be determined during Visit 2 and 3 (for time points please refer Flow Chart)
C: Parameters to be determined at Visit 4 (end-of-study examination)
2 Microscopic examination if erythrocytes, leukocytes, nitrite or protein are abnormal in urine

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. Except for drug screening, it is planned to perform these tests during screening only. Drug screening will be performed at screening and at each admission to the trial site for in-house stay.

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/Ecstasy 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 [redacted]) will be performed at each admission to the trial site for in-house stay, and may be repeated at any time during the trial 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. These tests will be performed at the trial site using M-10/14-PDT Surestep Multiline test or comparable test systems.

In case of positive drug screen, confirmatory test may be done at [redacted]. Laboratory data will be transmitted electronically from the laboratory to the trial site. It is the responsibility of the Investigator to evaluate the laboratory reports. Clinically relevant abnormal findings as judged by the Investigator are to be reported as adverse events (please refer to Section 5.2.6).

In case the criteria for hepatic injury are fulfilled, a number of additional measures will be performed (please see Section 5.2.6.1.4).

5.2.4 Electrocardiogram

Twelve-lead ECGs (I, II, III, aVR, aVL, aVF, V1 - V6) will be recorded using a computerised electrocardiograph (CardioSoft EKG System, [redacted]) at the times provided in the Flow Chart.

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 sec duration after subjects have rested for at least 5 min in a supine position. ECG assessment will always precede all other trial procedures scheduled for the same time to avoid compromising ECG quality.

All ECGs will be stored electronically on the Muse CV Cardiology System ([redacted]). Electrode placement will be performed according to the method of Wilson, Goldberger and Einthoven modified by Mason and Likar (hips and shoulders instead of ankles and wrists).

All locally printed ECGs will be evaluated by the investigator or a designee. Abnormal findings will be reported as AEs (during the trial) or baseline conditions (if identified at the screening visit) if assessed to be clinically relevant by the investigator. Any ECG abnormalities will be carefully monitored and, if necessary, the subject will be removed from the trial and will receive the appropriate medical treatment.

ECGs may be repeated for quality reasons (for instance, due to alternating current artefacts, muscle movements, or electrode dislocation) and the repeated ECG will be used for analysis. Additional (unscheduled) ECGs may be collected by the investigator for safety reasons.

[redacted]

5.2.6 Assessment of adverse events

5.2.6.1 Definitions of adverse events

5.2.6.1.1 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 considered related or not.

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.

5.2.6.1.2 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 (except hospitalisation for routine check-up, diagnostic purposes only or for a planned surgery), or 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

5.2.6.1.3 AEs considered 'Always Serious'

In accordance with the European Medicines Agency initiative on Important Medical Events, Boehringer Ingelheim has set up a list of 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. A copy of the latest list of 'Always Serious AEs' will be provided upon request. These events should always be reported as SAEs as described in Section 5.2.6.2.

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

5.2.6.1.4 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, please see Section 5.2.6.2.2.

The following are considered as AESIs:

- Potential severe DILI

  A potential severe Drug Induced Liver Injury (DILI) that requires follow-up is defined by the following alterations of hepatic laboratory parameters:
  - o An elevation of AST (aspartate aminotransferase) and/or ALT (alanine aminotransferase) ≥3-fold ULN combined with an elevation of total bilirubin ≥2-fold ULN measured in the same blood sample, or in samples drawn within 30 days of each other, or
  - o 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.

5.2.6.1.5 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

5.2.6.1.6 Causal relationship of AEs

Medical judgment should be used to determine whether there is a reasonable possibility of a causal relationship between the AE and the given trial treatment, 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
- There is an 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

5.2.6.2 Adverse event collection and reporting

5.2.6.2.1 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. 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 (the End of Study (EoS) visit):
    - 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 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 trial treatment related SAEs and trial treatment related AESIs of which the investigator may become aware of by any means of communication, e.g. phone call. Those AEs should be reported on the BI SAE form (see Section 5.2.6.2.2), but not on the CRF.

5.2.6.2.2 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 to the sponsor's unique entry point within 24 hours of becoming aware of the event (the only exception to this rule are SAEs 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), the country specific reporting process will be provided in the ISF. The same timeline applies if follow-up information becomes available. On specific occasions, the Investigator could inform the sponsor upfront via telephone. This does not replace the requirement to complete and send 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. All (S)AEs, including those persisting after the individual subject's end of trial, must be followed up until they have resolved, have been sufficiently characterized (e.g. as 'chronic' or 'stable'), or no further information can be obtained.

5.3 DRUG CONCENTRATION MEASUREMENTS AND PHARMACOKINETICS

5.3.1 Assessment of pharmacokinetics

For the assessment of pharmacokinetics, blood samples will be collected at the time points indicated in the Flow Chart. The actual sampling times will be recorded and used for determination of pharmacokinetic parameters.

5.3.2 Methods of sample collection

5.3.2.1 Blood sampling for pharmacokinetic analysis of BI 425809

For quantification of BI 425809 concentrations in plasma, 2.7 mL of blood will be drawn from an antecubital or forearm vein into a K_2-EDTA (dipotassium ethylenediamine-tetraacetic 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 venipuncture with a metal needle.

The EDTA-anticoagulated blood samples will be centrifuged for about 10 min at approximately 2000 x g to 4000 x 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 0.5 mL of plasma. The process from blood collection until transfer of plasma aliquots into the freezer should be completed within 90 min with interim storage of blood samples and aliquots at room temperature. The time 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 aliquot. At the analytical laboratory, the plasma samples will be stored at approximately -20°C or below until analysis.

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

The back-up samples in period 2 (2^{nd} aliquots of BI 425809 samples in period 2) can be used to get a profile of bosentan after confirmation of the TBA about the successful analysis of BI 425809 in the 1^{st} aliquots.

5.3.2.2 Blood sampling for pharmacokinetic analysis of bosentan

For quantification of bosentan concentrations in plasma, 2.7 mL of blood will be drawn from an antecubital or forearm vein into a K_2-EDTA-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 venipuncture with a metal needle.

The EDTA-anticoagulated blood samples will be centrifuged for about 10 min at approximately 2000 x g to 4000 x 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 0.5 mL of plasma. The process from blood collection until transfer of plasma aliquots into the freezer should be completed within 90 min with interim storage of blood samples and aliquots at room

temperature. The time 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 lab after the trial bioanalyst (TBA) has acknowledged safe arrival of the first aliquot. At the analytical laboratory, the plasma samples will be stored at approximately -20°C or below until analysis.

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

After analysis, all plasma samples of this trial may be used for further methodological investigations (e.g. for stability testing or assessment of metabolites) or to address Health Authority questions regarding the results/methodology. However, only data related to the analyte and its metabolites 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.

5.4 ASSESSMENT OF BIOMARKERS

Not applicable.

5.5 BIOBANKING

Not applicable.

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 parameters in an appropriate way. The scheduled measurements will allow monitoring of changes in vital signs, standard laboratory values, ECG parameters and suicidal behavior 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. The pharmacokinetic parameters and measurements outlined in Section 5.3 are generally used assessments of drug exposure.

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 end of trial examination are provided in the Flow Chart.

The tolerance time for drug dosing on Days 1-8 (period 1) and 1-12 (period 2) is ± 1 hour.

If not stated otherwise in the Flow Chart, the acceptable deviation from the scheduled time for vital signs, ECG, and lab tests will be -60 min. The tolerance for suicidality assessment is ± 60 min.

If scheduled in the Flow Chart at the same time as a meal, blood sampling, ECG and vital signs have to be done first. 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.

For planned blood sampling times, 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.

6.2 DETAILS OF TRIAL PROCEDURES AT SELECTED VISITS

6.2.1 Screening period

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 trial.

For information regarding laboratory tests (including drug and virus screening), ECG, vital signs, suicidality assessment and physical examination, refer to Sections 5.2.1 to 5.2.5.

Genotyping will be performed in those volunteers whose genotypes have not been previously determined (for details, see Section 5.6).

6.2.2 Treatment periods

Each subject is expected to participate in 2 treatment periods (Period 1 = 10 dosing days, Period 2 = 14 dosing days) that follow directly after each other.

On Day 9 of period 1, trial participants will be admitted to the trial site and kept under close medical surveillance for the last day of period 1 and for the first day of period 2. The subjects will be allowed to leave the trial site in the morning of Day 2 / period 2 after formal assessment and confirmation of their fitness.

On Day 13 of period 2, trial participants will be admitted to the trial site and kept under close medical surveillance for at least 12 hours after last bosentan dosing. The subjects will be allowed to leave the trial site after formal assessment and confirmation of their fitness.

On all other trial days, subjects will be treated in an ambulatory fashion.

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

The safety measurements performed during the treatment period are specified in Section 5.2 of this protocol and in the Flow Chart. AEs and concomitant therapy will be assessed continuously from obtaining subject's written informed consent until the end of trial examination.

For details on times of all other trial procedures, refer to the Flow Chart.

6.2.3 Follow-up period and trial completion

For AE assessment, laboratory tests, recording of ECG and vital signs, suicidality assessment and physical examination during the follow-up period, see Section 5.2.

Subjects who discontinue treatment before the end of the planned treatment period should undergo the EoS Visit.

If needed in the opinion of the investigator, additional visits may be scheduled after the EoS Visit for continued safety monitoring.

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 EoS 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 NULL AND ALTERNATIVE HYPOTHESES

The relative bioavailability of BI 425809 given alone (Reference) compared to a combined administration with the moderate CYP3A4 inducer bosentan (Test) will be estimated by the ratios of the geometric means (test/reference), 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.

7.2 PLANNED ANALYSES

7.2.1 General considerations

7.2.1.1 Analysis sets

Statistical analyses will be based on the following analysis sets:

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

Descriptions of additional analysis sets may be provided in the TSAP.

Adherence to the protocol will be assessed by the trial team. Important protocol deviation (IPD) categories will be suggested in the IPD specification file. IPDs will be identified no later than in the Report Planning Meeting, and the IPD categories will be updated as needed.

7.2.1.2 Pharmacokinetics

The pharmacokinetic parameters listed in Section 2.1 [redacted] for drug BI 425809 will be calculated according to the relevant BI internal procedures.

Plasma concentration data and parameters of a subject will be included in the statistical pharmacokinetic (PK) analyses if they are not flagged for exclusion due to a protocol deviation relevant to the evaluation of PK (to be decided no later than in the Report Planning Meeting) or due to PK 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.

Important protocol deviations 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

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

- The subject experienced emesis that occurred at or before two times median t_{max} of the respective treatment (Median t_{max} is to be determined excluding the subjects experiencing emesis),
- If a subject experiences emesis at any time during ambulatory dosing of BI 425809 the potential impact on the attainment of steady state will be assessed at the report planning meeting
- Missing samples/concentration data at important phases of PK disposition curve

Plasma concentration data and parameters of a subject which are flagged for exclusion will be reported with its individual values but will not be included in the statistical analyses. Descriptive and inferential statistics of PK parameters will be based on the PKS.

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 provided in the bioanalytical report, (that is, to the same number of decimal places provided in the bioanalytical report).

7.2.2 Primary endpoint analyses

Primary analyses

The statistical model used for the analysis of the primary endpoints (refer to Section 2.1.2) will be an analysis of variance (ANOVA) model on the logarithmic scale. That is, the PK endpoints will be log-transformed (natural logarithm) prior to fitting the ANOVA model. This model will include effects accounting for the following sources of variation: subjects and treatment. The effect 'subjects' will be considered as random, whereas the other effects will be considered as fixed. The model is described by the following equation:

$y_{km} = \mu + s_m + \tau_k + e_{km}$, where

y_{km} = logarithm of response measured on subject m receiving treatment k,

μ = the overall mean,

s_m = the effect associated with the m^{th} subject, m = 1, 2, ..., n

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

e_{km} = the random error associated with the m^{th} subject who received treatment k.

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

Point estimates for the ratios of the geometric means (test/reference) 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]

7.2.3 Secondary endpoint analyses

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

[redacted]

[redacted]

7.2.5 Safety analyses

Safety will be analysed based on the assessments described in Section 2.2.2.2. All treated subjects (TS, refer to Section 7.2) will be included in the safety analysis. Safety analyses will be descriptive in nature and based on BI standards. No hypothesis testing is planned.

For all analyses, the treatment actually administered (= treatment at onset) to the subject will be used.

Treatments will be compared in a descriptive way. Tabulations of frequencies/proportions will be used to evaluate categorical (qualitative) data, and tabulations of descriptive statistics will be used to analyse continuous (quantitative) data.

Measurements (such as ECG, vital signs, or laboratory parameters) or AEs will be assigned to treatments (see Section 4.1) based on the actual treatment at the time of the measurement or on the recorded time of AE onset (concept of treatment emergent AEs).

Therefore, measurements performed or AEs recorded prior to first intake of trial medication will be assigned to the screening period, those between first trial medication intake and end of REP (see Section 1.2.3) will be assigned to the treatment period. Events occurring after the REP but prior to next intake or end of trial termination date will be assigned to 'follow-up'. In case of two or more treatments, the follow-up will be summarized according to the previous treatment. These assignments including the corresponding time intervals will be defined in detail in the TSAP. Note that AEs occurring after the last per protocol contact will not be captured in the trial database (see 5.2.6.1.1).

Additionally, further treatment intervals (analysing treatments) may be defined in the TSAP in order to provide summary statistics for time intervals, such as combined treatments, on-treatment totals, or periods without treatment effects (such as screening and follow-up 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 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.

For laboratory data, ECG and vital signs, baseline is defined as the last measurement before first intake of BI 425809 in the respective trial period.

Relevant ECG findings will be reported as AEs.

Results regarding the C-SSRS will only be listed.

7.2.6 Interim analyses

No interim analysis is planned.

7.3 HANDLING OF MISSING DATA

7.3.1 Safety

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

7.3.2 Pharmacokinetics

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

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

7.4 RANDOMISATION

The trial will not be randomised, thus this section is not applicable.

7.5 DETERMINATION OF SAMPLE SIZE

It is planned to enter a total of 14 subjects in the trial, because this sample size is considered sufficient to achieve the aims of this exploratory trial. 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.

The observed intra-individual coefficient of variation (gCV) for BI 425809 in the previous trial 1346-0039 was roughly 12.3% for $C_{max,ss}$ and 12.2% for $AUC_{\tau,ss}$. These results were used to approximate the precision for the endpoints in this trial.

For various assumptions around the gCV of 12%, Table 7.5: 1 provides an overview of the achievable precision for estimating the ratio of geometric means (test/reference). For illustrative purposes, the expected 90% confidence intervals are displayed for different values of the ratios T/R of geometric means.

Table 7.5: 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 2-period fixed sequence trial *(N=14)*

gCV [%]	Precision upper CL / relative BA estimate	Ratio [%]*	Lower CL [%]	Upper CL [%]
10	1.09	40	36.65	43.66
10	1.09	60	54.97	65.49
10	1.09	80	73.29	87.32
10	1.09	100	91.62	109.15
12	1.11	40	36.01	44.43
12	1.11	60	54.02	66.64
12	1.11	80	72.03	88.85
12	1.11	100	90.04	111.07
14	1.13	40	35.40	45.20
14	1.13	60	53.09	67.81
14	1.13	80	70.79	90.41
14	1.13	100	88.49	113.01

*Ratio of geometric means (test/reference) for a PK endpoint is defined by $\exp(\mu_T)/\exp(\mu_R)$.

The expected 90% confidence interval limits in the table were derived by

$$\text{CI limit}_{\text{upper,lower}} = \exp(\ln(\theta) \pm \omega),$$

with θ being the ratio (T/R) on original scale and ω the distance from the estimate θ to either confidence interval limit on the log-scale, which was obtained from the achievable precision on the original scale.

The calculation was performed as described by Julious [R11-5230] using R Version 4.2.1.

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

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 directive 2001/20/EC) and other relevant regulations. Investigators and site staff must adhere to these principles. Deviation from the protocol, the principles of ICH GCP or applicable regulations will be treated as 'protocol deviation'.

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. As a general rule, no trial results should be published prior to finalisation 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 respective 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 or alternative plan, in line with the guidance provided by ICH Q9 and ICH-GCP E6, for fully outsourced trials, 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.

[redacted]

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: gender, 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, 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 a 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.

[redacted]

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 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

Data protection and data security measures are implemented for the collection, storage and processing of subject data in accordance with the principles 7 and 12 of the WHO GCP handbook.

Individual subject data obtained as a result of this trial is considered confidential and disclosure to third parties is prohibited with the following exceptions:
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 have to be in accordance with the informed consent
- The BI-internal facilities storing biological samples from clinical trial participants as well as the external storage 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 (e.g. biomarker proposal, analysis plan and report) ensures compliant usage
- 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 the whole trial ('Last Subject Completed').

Early termination of the trial is defined as the premature termination of the trial due to 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 IEC / 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.

The sponsor will submit to the EU database a summary of the final trial results within one year from the end of a clinical trial as a whole, regardless of the country of the last subject (EU or non-EU).

8.7 ADMINISTRATIVE STRUCTURE OF THE TRIAL

The trial is sponsored by Boehringer Ingelheim (BI).

The trial will be conducted at the [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. The investigators will have access to the BI web portal Clinergize to access documents provided by the sponsor.

BI has appointed a Clinical Trial Leader (CT 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 Trial Mangers (CT Managers), Clinical Research Associates (CRAs), 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 425809 concentrations in plasma will be performed at [redacted]

Analyses of bosentan concentrations in plasma will be performed at [redacted]

On-site monitoring will be performed by BI or a contract research organisation appointed by BI.

Data management will be done by BI according to BI SOPs.

Statistical evaluation will be done according to BI standards.

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.

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10. APPENDICES

Not applicable.

11. DESCRIPTION OF GLOBAL AMENDMENTS

11.1 GLOBAL AMENDMENT 1

Date of amendment		01 Feb 2023
EudraCT number		2021-006676-17
BI Trial number		1346-0056
BI Investigational Medicinal Product(s)		BI 425809 (iclepertin), Tracleer® (bosentan)
Title of protocol		The effect of multiple oral doses of bosentan on the steady state kinetics of BI 425809 after oral administration to healthy male subjects (an open-label, two-period fixed sequence trial)
Substantial Global Amendment due to urgent safety reasons		☐
Substantial Global Amendment		☒
Non-substantial Global Amendment		☐
Section to be changed		1.) Title page, synopsis, section 1.2.1 and 1.3. 2.) Flow Chart 3.) Section 8.1
Description of change		[redacted]
Rationale for change		1.) Request of Ethics committee 2.) Request of Ethics committee 3.) Request of Competent Authority

APPROVAL / SIGNATURE PAGE

Document Number: c40616508 **Technical Version Number:2.0**

Document Name: clinical-trial-protocol-version-02

Title: The effect of multiple oral doses of bosentan on the steady state kinetics of BI 425809 after oral administration to healthy male subjects (an open-label, two-period fixed sequence trial)

Signatures (obtained electronically)

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Author-Clinical Trial Leader	[redacted]	02 Feb 2023 12:43 CET
Verification-Paper Signature Completion		02 Feb 2023 14:27 CET
Author-Trial Statistician		02 Feb 2023 14:32 CET
Approval-Clinical Program [redacted]		06 Feb 2023 14:20 CET

(Continued) Signatures (obtained electronically)

Meaning of Signature	Signed by	Date Signed