Investigational New Drug

Minocin® (minocycline) for Injection

A Phase 1, Randomized, Double-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Study of the Safety, Tolerability, Pharmacokinetics of Minocin® (minocycline) for Injection in Healthy Adult Subjects

Protocol No.: MDCO-MIN-16-02
U.S. NDA No.: 50-444
EudraCT No.: 2016-002246-24
PROTOCOL VERSION: 5.0, Amendment 3.0

Development Phase:	Phase 1
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IMI/Combacte NET		
Medical Lead:	Name:	Oliver Cornely, MD
Project Lead:	Name:	Dorothee Arenz

Issue Date: Date: **09 Nov 2017**

This study will be conducted in compliance with Good Clinical Practice (GCP) and protection of the subject as required by the regulations and directives in operation at this time.

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Sponsor and Principal Investigator—Signatories

Study MDCO-MIN-16-02, A Phase 1, Randomized, Double-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Study of the Safety, Tolerability, Pharmacokinetics of Minocin® (minocycline) for Injection in Healthy Adult Subjects

Version: 5.0, Amendment 3.0	Version Date: 09 November 2017
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Signature: Brooke Lowe on Behalf of Jeff Loutit — Date: 13NOV2017

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Signature: Voors-Pette — Date: 13 Nov 2017

IMI / COMBACTE NET Academic Work Package Lead	Prof. Dr. Oliver A. Cornely Department I for Internal Medicine University Hospital Cologne Kerpener Str. 62 50937 Koeln, Germany

Signature — Date: 13 Nov 2017

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1. PROTOCOL SYNOPSIS

Name of Sponsor/Company: The Medicines Company
Name of Finished Drug: Minocin® (minocycline) for Injection, referred to as Minocin IV in this protocol
Name of Active Ingredient: Minocycline
Title of Study: A Phase 1, Randomized, Double-Blind, Placebo-Controlled, Single and Multiple Ascending Dose Study of the Safety, Tolerability, Pharmacokinetics of Minocin® (minocycline) for Injection in Healthy Adult Subjects
Phase of Development: Phase 1
Study Center: Single study site is QPS Netherlands BV, Groningen, The Netherlands
Number of Subjects: Approximately 60 healthy subjects (approximately 30 men and 30 women). Each randomized subject can be part of one cohort only, i.e. cannot be randomized twice.
Principal Investigator: Christine Voors-Pette, MD
Study Period: The estimated study period will be approximately 36 weeks from screening of the first subject to completion of the last subject.
Objectives: • The primary objective is to assess the safety and tolerability of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects. • The secondary objective is to assess the pharmacokinetics of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects.
Rationale: Minocin® (minocycline) for Injection, and referred to as Minocin IV in this protocol, is an intravenous (IV) formulation of minocycline, a derivative of tetracycline. Minocin IV is similar to other drugs developed in the era prior to the maturation of the field of pharmacodynamics, in that human doses were not systematically determined or optimized. Minocycline was first approved in the United States (US) as both oral and IV formulations in the 1970s. Only oral dosage formulations of minocycline have been approved for use in countries in the EU. An updated development program for Minocin IV would enable the availability of a new agent in the European Union (EU) to address highly resistant strains of *Acinetobacter baumannii*. The program will build on recently acquired information in preclinical studies and allow for optimization of the dosage regimen prior to use in controlled studies. The overall development strategy in the EU is summarized below: • Establish safety, tolerability, and PK of escalating doses of Minocin IV in normal subjects • Link PK data in human subjects with PK-PD efficacy targets in animal models of infection to select dosage regimen • Conduct a pathogen-directed clinical study of Minocin IV in infections due to *Acinetobacter baumannii*, assisted by the use of a rapid diagnostic test Two recent studies using high doses of minocycline (single doses up to 800 mg and multiple doses of 400 mg twice a day) have been conducted in patients with stroke (Fagan 2010) and spinal cord injury

(Casha 2011). These studies were conducted to investigate the potential clinical uses of higher doses of minocycline taking advantage of its properties unrelated to its antimicrobial effects (Garrido-Mesa, 2013), and resulted in pharmacokinetic and safety data of intravenous minocycline at high doses. The results of these studies indicate that doses of minocycline higher than those currently approved for IV administration in the US, and oral in the US and EU, appear to be safe and well tolerated in humans. Based on these data, the sponsor believes it is rational to investigate higher doses of minocycline to optimize PK-PD properties for treatment of serious gram-negative pathogens, particularly non-fermenting organisms such as *Acinetobacter baumannii* complex.

This study is the first in the EU development program described above and will start with doses already approved in the US, namely 100 mg and 200 mg, in order to build a population PK model. Doses will be increased up to 600 mg if safety is observed at the lower doses. Data from this study will determine the dose of Minocin IV to be used in the Phase 3 study in the treatment of subjects with pneumonia secondary to *Acinetobacter baumannii*.

Methodology:

This is a double-blind, randomized, placebo-controlled, single- and multiple ascending dose study of up to 5 doses (cohorts) of Minocin IV. An effort should be made to include balanced numbers of male and female subjects. At least 2 male and 2 female subjects must be assigned to receive Minocin, 1 male and 1 female to receive placebo in each cohort.

Each cohort will consist of 10 subjects (8 active drug and 2 placebo).

Planned Cohorts:

Cohort 1: 100 mg*

Cohort 2: 200 mg*

Cohort 3: 300 mg

Cohort 4: 400 mg

In the ongoing study, dose escalation was stopped after the multiple dose phase of 400 mg cohort due to 6/10 subjects in this cohort discontinuing due to AEs of dizziness and nausea. Consequently, cohorts 5 and 6 are adapted to 500 mg or 600 mg respectively as a single dose on day 1 and as a loading dose on day 4 followed 12 hours later by multiple doses of 300 mg every 12 hours.

Cohort 5: 500/300 mg

Cohort 6: 600/300 mg

* Cohorts 1 and 2 may be run concurrently as they are the approved doses of minocycline IV in the US. Cohorts 1/2, 3, 4, 5 and 6 will be run sequentially.

Within each cohort, subjects will receive a single dose on Day 1, followed by 7 days of multiple doses (Days 4-10), followed by a single dose on Day 11. Subjects will be discharged from the clinic on Day 14 upon having an EOS evaluation and a follow-up phone call will occur on Day 17 (See Schedule and Events Table for details).

Subjects are required to remain admitted in the research unit from the time of randomization (Day 1) until the discharge on Day 14. Subjects that discontinue the study early are required to complete Early Termination procedures.

Safety will be assessed throughout the study and serial blood and urine samples will be collected for the safety and PK assessment of Minocin IV.

Adverse events and serious adverse events will be collected and recorded. Safety will be evaluated by the assessment of clinical safety laboratory results, vital sign measurements, ECG and physical

examination findings. A Day 17 follow up phone call will be performed to evaluate safety by the assessment of adverse events and serious adverse events.

Main Criteria for Selection:

Inclusion Criteria

Subjects must meet all of the following criteria in order to be eligible for the study:

1. A signed informed consent form, the ability to understand the study conduct and tasks that are required for study participation, and a willingness to cooperate with all tasks, tests, and examinations as required by the protocol, whether in the research unit or after discharge, for the duration of the study;
2. Male or female between 18 and 50 years of age inclusive;
3. Subject has a body mass index (BMI) ≥18 kg/m2 and ≤ 30 kg/m^2;
4. Subject is non-smoker or smokes up to 5 cigarettes per day (or equivalent);
5. Subject is in good health based on medical history and physical examination findings and has no clinically meaningful safety laboratory abnormalities (Haematology, blood chemistry, and urinalysis) or 12-lead ECG results, as assessed by the Principal Investigator (PI);
6. Vital signs (BP, pulse, respiratory rate and temperature) measured at screening/baseline must be within the following ranges: SBP ≥90 to ≤150 mm Hg, DBP ≥45 to ≤90 mm Hg; Heart Rate ≥ 45 to ≤90 bpm (taken after resting in a supine position for at least 5 minutes);
7. Expectation that intravenous access will be sufficient to allow for ease of study drug infusion, and for all protocol required blood sampling to take place;
8. Subject commits to remaining admitted in the research unit for the course of the study;
9. Female subject is surgically sterile, postmenopausal: period of amenorrhea for at least 2 years, or if of childbearing potential, agrees to abstinence or to use at least 2 acceptable methods of birth control (e.g. prescription oral contraceptives, contraceptive injections, contraceptive patch, intrauterine device, barrier methods, etc.) or male partner sterilization alone, between the first dose (Day 1) and for 90 days after the completion of the study.

Exclusion Criteria:

Subjects who meet any of the following exclusion criteria will not be enrolled in the study:

1. Has any condition, including findings in the medical history or in pre-study assessments that constitutes a risk or a contraindication for the participation in the study or completing the study;
2. Positive breath test for alcohol and/or positive urine test for drugs of abuse at Screening and Day -1 Visits;
3. Has a history or presence of alcohol/drug abuse within 2 years. Alcohol abuse is defined as regularly consuming >3 units/day (21 units per week for men), >2 units/day (14 units/week) for women. A unit is defined as a can of 4% beer (330 mL), approximately 190 mL of 6-7% beer (malt liquor), a glass of 40% spirits (30 mL), a glass of wine (100 mL);
4. Subject shows positive hepatitis B surface antigen (HBsAg), hepatitis C virus antibodies (anti-HCV), or human immunodeficiency virus (HIV) I/II antibodies and antigen tests;
5. Subject has active or ongoing candida infection;
6. Blood or plasma donation within past 2 months;
7. Females who are pregnant or nursing or who have a positive pregnancy test result at the

Screening Visit or Day -1 prior to dosing;

8. Males who are unwilling to practice abstinence or use an acceptable method of birth control during the entire study period and for 90 days after the completion of the study (i.e. condom with spermicide, where locally available);
9. Presence of known raised intracranial pressure;
10. Use of retinoids (e.g., Isotretinoin);
11. History of significant hypersensitivity or allergic reaction to any of the tetracycline class of antibiotics or the components of those antibiotics;
12. Receipt of any investigational medication or investigational device during the last 30 days prior to randomization;
13. Treatment with any prescription, vitamins or OTC drugs, within 2 weeks or five half-lives, whichever is longer, or herbal nutritional supplements within 2 weeks of screening, with the exception of acetaminophen/paracetamol for minor headache. Subjects will not be allowed to receive medications for the duration of the study (except the abovementioned acetaminophen/paracetamol). Birth control or other hormone replacement is also permitted as long as it has been taken at a stable dose for at least three months before the Screening Visit and remains stable for the duration of the study;
14. A QTcF >480 msec;
15. Calculated creatinine clearance less than 50 mL/min (Cockcroft-Gault method) at screening or check-in (Day -1);
16. Unable or unwilling, in the judgment of the Investigator, to comply with the protocol;
17. An employee of the Investigator, the study center, the sponsor or The Medicines Company with direct involvement in the proposed study or other studies under the direction of that Investigator or study center, or a family member of the employee or the Investigator;
18. Prior enrollment in any minocycline study including prior cohorts in this trial.

Test Drug Mode of Administration:

Minocin IV or placebo doses should be prepared by an unblinded pharmacist (or qualified designee).

Subjects randomized to Minocin IV will receive 100-600 mg depending on cohort. Cohorts 1-4 received 100-400 mg, respectively. Cohorts 5 and 6 are adapted to 500 mg or 600 mg respectively as a single dose on Day 1 and as a loading dose on Day 4 followed 12 hours later by multiple doses of 300 mg every 12 hours. Doses will be administered over 60 minutes in 0.9% Sodium Chloride Injection (referred to as Normal Saline for the remainder of the document). Detailed dosing preparation is described in the Pharmacy Manual.

The placebo will be Normal Saline and will be administered at a rate and volume matching the rate and volume of Minocin IV for each respective Cohort.

All doses of Minocin IV or placebo will be administered intravenously into a peripheral vein.

Duration of Treatment: The Screening period for all cohorts is ≤28 days prior to admission on Day -1, with single dose administration on Days 1 and 11; multiple-dose administration on Days 4-10; Serial PK collection on Days 1-4 and 11-14. Subjects are discharged on Day 14 after the final study assessments are completed. A Follow-up phone call will occur on Day 17.

Reference Therapy, Dose and Mode of Administration: Normal Saline (placebo) as administered at a rate and volume matching the rate and volume of Minocin IV for each respective Cohort.

Criteria for Evaluation:

Endpoints:

- Assess the safety and tolerability of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects (see below section **Safety**).
- Assess the pharmacokinetics of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects (see below section **Pharmacology**).

Efficacy: Efficacy will not be assessed in this study.

Safety Evaluations:
Safety will be evaluated by the assessment of AEs/SAEs, clinical safety laboratory results, ECG, vital sign measurements, and physical examination findings.

Dose Escalations and Stopping Rules:

Intra-Cohort Data Review:
For Cohorts 3-6, single dose safety data will be reviewed by the Medical Monitor and Principal Investigator on Day 3 prior to the start of the multi- dosing on Day 4. During the review, blinded safety data from the single dose infusions will be assessed and one of the following will be decided before the start of the Multiple Dose Phase on Day 4:

1. To continue with the Multi-dose phase as planned,
2. To continue with the Multi-dose phase and add additional safety evaluations,
3. To not continue with the Multi-dose phase.

Between- Cohort Data Monitoring Committee Review:
Safety data will be reviewed by the Data Monitoring Committee (DMC) following each cohort and prior to the start of the next cohort.

After completion of cohorts 1 and 2, which may run concurrently, and prior to dosing of each cohort 3, 4,5 and 6, the DMC will review all pertinent blinded safety data (e.g., physical examinations, vital signs, electrocardiograms (ECGs), clinical laboratory tests, and AEs) from the preceding cohort to determine one of the following:

1. To continue with the next cohort as planned,
2. To continue with the next cohort and add additional safety evaluations,
3. To continue with the next cohort by adjusting the planned dose of Minocin IV, or
4. To not continue with the next cohort.

Stopping Rules

The study will be terminated if ≥ 2 subjects in a cohort meet any of the following criteria and the subject was determined to have received active drug after unblinding:

- Has a drug related SAE
- Experiences a drug related AE grade ≥ 3 toxicity

Plasma and Urine PK Assessments:

Plasma and urine samples will be collected during the Single and Multiple Dose Phases to determine concentrations of Minocin IV and metabolites in plasma and urine (see Schedule of Events Flowchart for specific collection times).

Pharmacology:

The plasma concentration-time data for Minocin IV will be analyzed by non-compartmental methods.

Treatment arms (doses, placebo) are compared regarding plasma AUC_{0-t}, AUC_{0-inf}, C_{max}, and T_{max}. Individual as well as mean time-concentration profiles will be graphed. Statistical analysis of dose proportionality of exposure parameters will be performed. Urine PK parameters such as amount excreted and % dose excreted will be calculated from urinary excretion data. Detailed methods used and the results obtained will be included in a separate PK report.
Statistical Methods: Ten subjects per cohort are planned, hence n = 60 subjects in total. This sample size was selected based on previous experience attempting to minimize the number of subjects on study drug while obtaining sufficient PK data from different subjects to produce a good estimate of exposure. Descriptive statistics will be performed for patient characteristics, laboratory evaluations, vital signs and ECG results. Additional shift tables will be provided for laboratory evaluations and ECG results. TEAEs will be listed and number (percentage) of subjects reporting TEAEs and number (percentage) of TEAEs will be tabulated.

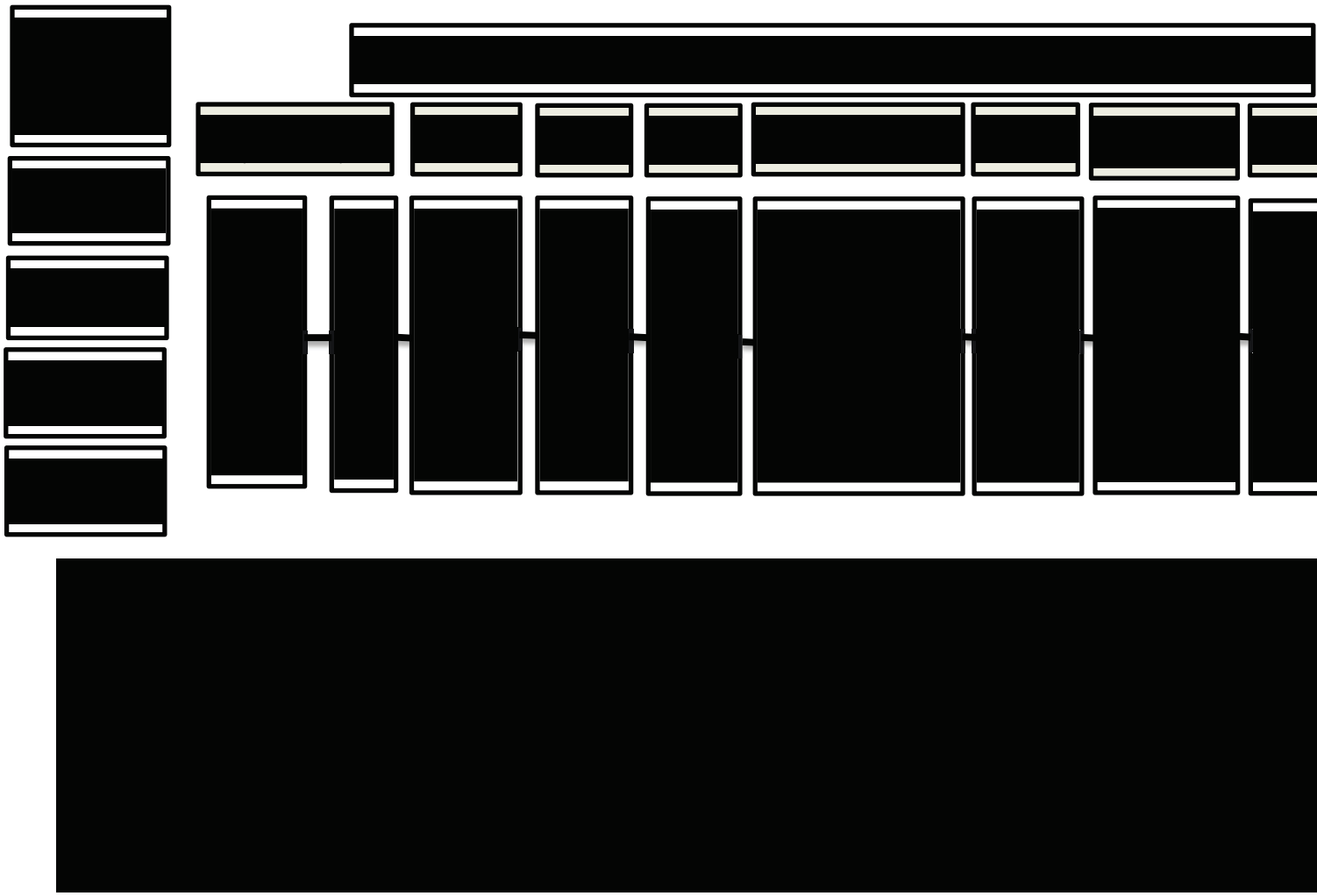
2. SCHEDULE OF EVENTS/ASSESSMENTS COHORT 1-6

Study Procedures	Screening (≤28 days to -1)	Day -1[1]	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6	Day 7	Day 8	Day 9	Day 10	Day 11	
Informed consent	X													
Medical History & Demographics	X	X												
Height, weight and BMI[3]	X	X										X		
Physical Examination	X	X			X						X			
Vital Signs:RR/BP/HR/Temperature[4]	X	X	X	X	X	X	X	X	X	X	X	X	X	
ECG (12-lead)[5]	X	X	X			X	X	X	X	X	X	X	X	
Laboratory Assessments[6]	X	X		X		X		X		X		X		
Creatinine clearance[7]			X											
Test for HIV, Hep B sAg, HCV Ab	X													
Urine Drug Screen / Alcohol breath test[8]	X	X												
Pregnancy Test[9]	X	X	X											
PK Blood Collection[10]			X	X	X	X[2]							X	
PK Urine Collection[11]			X	X	X	X							X	
Inclusion / Exclusion Criteria	X	X												
Dose Administration (1h Infusion)[12]			X			X	X	X	X	X	X	X	X	
Concomitant Medications	<----------X----------													
Adverse Events / SAEs			< ----------X----------											
Follow-Up Phone Call										------->				

[1] Day -1 procedures should be completed within approximately 24 hours before dosing on Day 1.
[2] The 72 h post-dose PK collection should occur before the start of the 2nd infusion on Day 4.
[3] BMI will only be calculated at screening. Weight will be collected on Day -1, 10 and 14.
[4] Vital signs (blood pressure, pulse, temperature, respirations) will be measured before and at 1, 3, and 6 h after the start of the morning dose and before and at 1 hour a
On Day 1 vitals will be collected at 1, 3, and 6 h after the start of dosing. Vital signs may be collected once in the morning on days where no dosing occurs.
[5] On Day -1, ECGs should be performed in triplicate. On Days 1 a triplicate ECGs will be performed 1h/end of infusion (+/- 10 minutes). Day4 triplicate ECGs will be
h/end of infusion (+/-10 minutes) after the start of the morning dosing. On Days 5-11 and 14, single ECGs are performed. Triplicate recording may be required depend
11.1.7.
[6] Includes serum chemistry, hematology, and urinalysis.On Days 4, 6, 8, and 10, collect blood and urine for clinical laboratory tests approx. 4 h after the start of the mo
[7] Urine from the PK collection on Day 1 (hours: 0-4, 4-8, 8-12, 12-24) will also be used to determine 24h creatinine clearance.
[8] Required for all subjects.
[9] Serum pregnancy should be performed during Screening, Day -1 and Day 14 EOS. Urine pregnancy test should be performed before dosing on Day 1.

[10] Serial plasma PK samples at the following time points (in hours) on **Day 1**:Before dosing and at 1 (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48, and 72 h (pre-dose or
Post-**Day 4 dose**: 1 (end-of-infusion), 2, 4, 8, and 12 h after the start of dosing; On **Day 11**: Before the Final Dose and at 1 (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48
[11] Urine for PK analysis will be collected at **Day 1** before dosing and during intervals: 0-4, 4-8, 8-12, 12-24, 24-48, and 48-72 h (pre-dose on Day 4) after dosing. **Day**
intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing.
[12] Subjects will receive a single dose on Day 1. Dosing on Days 4-10 will occur every 12 h. Only the one morning dose on Day 11 will be administered.

3. SCHEMATIC DIAGRAM OF TRIAL DESIGN



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4. LIST OF ABBREVIATIONS

A. baumannii	*Acinetobacter baumannii*
AE	Adverse event
AESI	Adverse event of special interest
ALT	Alanine aminotransferase
AM	*Ante meridiem,* morning
aPTT	Activated partial thromboplastin time
ARO	Academic Research Organization
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration-time curve
BMI	Body mass index
BP	Blood pressure
bpm	Beats per minute
BUN	Blood urea nitrogen
ºC	Degrees Celsius
CDAD	*Clostridium difficile*-associated diarrhea
CDC	Centers for Disease Control and Prevention
C. difficile	*Clostridium difficile*
CFR	Code of Federal Regulations
C_{max}	Maximum observed drug concentration
CS	Clinically significant
CTCAE	Common Terminology Criteria for Adverse Events
CTCC	Clinical Trials Center Cologne
d	Day(s)
DBP	Diastolic blood pressure
DMC	Data monitoring committee
EC	Ethics Committee
ECG	Electrocardiogram
eCRF	Electronic Case Report Form

eDC	Electronic Data Capture
EOS	End of study
ET	Early termination
EU	European Union
FDA	Food and Drug Administration
g	Gram(s)
GCP	Good Clinical Practice
GPV	Global pharmacovigilance
h	Hour
Hg	Mercury
ICF	Informed Consent Form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals
IEC	Independent Ethics Committee
IH	Intracranial hypertension
IMI	Innovative Medicines Initiative
IMP	Investigational medicinal product
IMSIE	Institute of Medical Statistics, Informatics and Epidemiology
ITT	Intent-to-treat
IU	International Units
IV	Intravenous
kg	Kilogram(s)
L	Liter
m^2	Meters squared
MAA	Marketing Authorization Application
max	Maximum
MDCO	The Medicines Company
MedDRA	Medical Dictionary for Regulatory Activities
mg	Milligram(s)

MI	Myocardial infarction
mITT	Modified intent to treat
mL	Milliliter(s)
min	Minute(s)
n	Number
NCS	Not clinically significant
NDA	New drug application
OTC	Over-the-counter
PCS	Potentially clinically significant
PD	Pharmacodynamics
pH	Power of hydrogen
PI	Principal Investigator
PK	Pharmacokinetic or pharmacokinetics
PP	Per-protocol (population)
PR	PR interval (time from onset of P wave to start of QRS complex)
PT	Prothrombin time
Q	Quartile
q12h	Every twelve hours
QTc	Corrected QT interval
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBP	Systolic blood pressure
SD	Standard deviation
SmPC	Summary of Product Characteristics
T_{max}	Time of the maximum drug concentration
UA	Urinalysis
UKK	University Hospital of Cologne
ULN	Upper limit of normal
US, USA	United States of America

USP	United States Pharmacopeia
WHO	World Health Organization

5. INTRODUCTION

This study will evaluate the safety, tolerability and PK of Minocin IV (Minocycline) for Injection.

Minocycline was first approved in the United States (US) as both oral and IV formulations in the 1970s. Only oral dosage formulations of minocycline have been approved for use in countries in the EU. A new formulation of Minocin IV has been approved in the US that enables administration of minocycline in a smaller volume of fluid. The Medicines Company plans to conduct a development program in the EU for Minocin IV for the treatment of infections due to *A. baumannii*, and this new formulation supports the planned study of higher doses of minocycline.

This study will be conducted in compliance with Good Clinical Practices (GCP) including the Declaration of Helsinki and all applicable regulatory requirements.

Additional details regarding Minocin IV can be found in the Investigator's Brochure.

5.1. Background

Minocycline is a tetracycline derivative. The approved indication for Minocin IV in the United States includes the treatment of infections due to susceptible strains of several important Gram-positive and Gram-negative pathogens, including *Acinetobacter* species.

Acinetobacter infections are associated with high morbidity and mortality [Munoz-Price and Weinstein, 2008]. Antibacterial agents frequently used to treat *Acinetobacter* infections include aminoglycosides, cephalosporins, and carbapenems; however, resistance to these first-line agents is increasing, leaving only unproven agents available for treatment [Shlaes et al, 2013; Munoz-Price and Weinstein, 2008]. In view of this increasing resistance, both EU and US governments have identified multi-drug resistant *Acinetobacter* infections as a public health risk [EU MEMO/08/788, 2008; US Generating Antibiotic Incentives Now (GAIN) Act, 2012; and Centers for Disease Control (CDC) Antibiotic Resistance Threats Report, 2013].

It is noteworthy that while minocycline has been approved since the 1970s, recent studies indicate that minocycline remains highly active in vitro against *Acinetobacter* species. Importantly, minocycline has been found to be active against strains that are resistant to doxycycline or tigecycline [Munoz-Price and Weinstein, 2008].

The original IV formulation of minocycline, Minocin (minocycline) for Injection, was approved in the US on 26 October 1972. Only oral dosage formulations of minocycline and Minocin have been approved for use in countries in the EU since the 1970s [Minocycline tablets SmPC, 2011; Minocin MR Capsules SmPC, 2014]. The new formulation of Minocin IV was approved in the US on 17 April 2015 (NDA 50,444). The new formulation is comprised of minocycline hydrochloride with magnesium sulfate to improve the solubility and stability of minocycline solutions at a more physiological pH, which enables administration of minocycline in a smaller volume of fluid.

The Medicines Company plans to conduct three Phase I safety and pharmacology studies (including this study) and one Phase III study to support the overall development strategy for Minocin IV in the EU.

5.2. Minocin IV (minocycline) for Injection

Minocin® (minocycline) for Injection, and referred to as Minocin IV in this protocol, is an intravenous (IV) formulation of minocycline, a derivative of tetracycline.

Minocin IV is supplied as a sterile lyophilized cake in a single-use 10 mL glass vial with a rubber stopper and an aluminum over-seal. Each vial contains 108 mg of minocycline hydrochloride equivalent to 100 mg minocycline, 269 mg of magnesium sulfate heptahydrate equivalent to 27 mg of magnesium (an inactive ingredient), and sodium hydroxide (to adjust pH).

5.2.1. Nonclinical Studies

The nonclinical development program for Minocin IV consists of studies of antibacterial potency in vitro against clinical isolates and in vivo in mouse and rat infection models; in vitro and in vivo pharmacokinetic and metabolism studies; and a battery of nonclinical toxicity, safety, and local tolerance studies, including 14 day IV repeat-dose studies in rats and dogs, safety pharmacology studies in rats and dogs, and local tolerance studies in mice and rabbits.

Additional details regarding the nonclinical development program for Minocin IV can be found in the Investigator's Brochure.

5.2.2. Clinical Studies

No clinical studies have been conducted to date by the sponsor with the new formulation of Minocin IV. However, clinical pharmacokinetic and safety information are available from the US Prescribing Information for Minocin IV [Minocin for Injection PI, 2015], and while no randomized controlled trials of IV minocycline in patients with *A. baumannii* infection exist, clinical efficacy

data are available from published case report series that indicated successful treatment.

Following a single dose of Minocin IV 200mg administered to 10 healthy male subjects, serum concentrations of minocycline ranged from 2.52 µg/mL to 6.63 µg/mL (average of 4.18 µg/mL) at the end of infusion and 0.82 µg/mL to 2.64 µg/mL (average of 1.38 mcg/mL) after 12 hours [Minocin IV Investigator's Brochure Ed. 2 Addendum, 2017].

The serum t½ of minocycline following administration of Minocin IV 100 mg every 12 hours or 200 mg once daily was not significantly different and ranged from 15 hours to 23 hours [Minocin IV Investigator's Brochure Ed. 2 Addendum, 2017.

IV minocycline has been in use for over four decades and has a well-established safety profile. Recent clinical safety data have been generated from studies of potential non-antimicrobial effects of minocycline [Casha et al, 2012; Fagan et al, 2010].

Additional details regarding the clinical studies conducted for Minocin IV can be found in the Investigator's Brochure.

5.2.3. Known and Potential Risks and Benefits

In general, the risk of significant adverse events related to the Minocin IV is minimal and consistent with taking intravenous doses of a tetracycline antibiotic. The following adverse reactions have been observed in patients receiving tetracyclines and are included in the Minocin® (minocycline) Investigator's Brochure.

Drug rash with eosinophilia and systemic symptoms, including fatal cases, have been reported with minocycline use. If this syndrome is recognized, the drug should be discontinued immediately.

Photosensitivity manifested by an exaggerated sunburn reaction has been observed in some individuals taking tetracyclines. This has been reported with minocycline.

Central nervous system side effects including light-headedness, dizziness, or vertigo have been reported with minocycline. These symptoms may disappear during therapy and usually disappear rapidly when the drug is discontinued.

Clostridium difficile (*C. difficile*) associated diarrhea (CDAD) has been reported with use of nearly all antibacterial agents, including Minocin, and may range in severity from mild diarrhea to fatal colitis. Treatment with antibacterial agents alters the normal flora of the colon leading to overgrowth of *C. difficile*.

Intracranial hypertension (IH; *pseudotumor cerebri*) has been associated with the use of tetracyclines, including Minocin. Clinical manifestations of IH include headache, blurred vision, diplopia, and vision loss; papilledema can be found on fundoscopy. Women of childbearing age who are overweight or have a history of IH are at greater risk for developing tetracycline-associated IH. Concomitant use of isotretinoin and Minocin should be avoided because isotretinoin is also known to cause pseudotumor cerebri. Although IH typically resolves after discontinuation of treatment, the possibility for permanent visual loss exists.

The anti-anabolic action of the tetracyclines may cause an increase in BUN. While this is not a problem in those with normal renal function, in patients with significantly impaired function, higher serum levels of tetracycline may lead to azotemia, hyperphosphatemia, and acidosis.

Minocin IV, like other tetracycline-class antibacterials, can cause fetal harm when administered to a pregnant woman. The use of drugs of the tetracycline class during tooth development (last half of pregnancy, infancy, and childhood to the age of 8 years) may cause permanent discoloration of the teeth (yellow-gray-brown). Enamel hypoplasia has also been reported.

All tetracyclines form a stable calcium complex in any bone-forming tissue. A decrease in the fibula growth rate has been observed in premature human infants given oral tetracycline in doses of 25 mg/kg every 6 hours.

Results of animal studies indicate that tetracyclines cross the placenta, are found in fetal tissues, and can have toxic effects on the developing fetus (often related to retardation of skeletal development). Evidence of embryotoxicity has been noted in animals treated early in pregnancy.

As with other antibacterial preparations, use of Minocin may result in overgrowth of nonsusceptible organisms, including fungi.

Because Minocin IV contains magnesium, close monitoring is recommended; it should be used with caution in patients with heart block or myocardial damage.

Minocin IV is contraindicated in individuals who have shown hypersensitivity to any of the tetracyclines or to any components of the product formulation.

The safety monitoring practices employed by this protocol (i.e. physical examination, vital signs, 12-lead ECG, hematology, serum chemistry, urinalysis, and AE collection) are adequate to protect the subjects' safety and should detect adverse events.

The approximate volume of blood planned for collection from each subject over the course of the study presents no undue risk to the subjects.

There will be no direct health benefit for trial participants from receipt of study drug. An indirect health benefit to the healthy subjects enrolled in this trial is the free medical tests received at screening and during the study.

A complete description of relevant risks of Minocin IV, including reported adverse reactions that have been observed in patients receiving tetracyclines, can be found in the Investigator's Brochure Ed. 2 Addendum, 2017.

5.3. Study Rationale

The purpose of this study is to collect safety, tolerability and PK data on ascending dose regimens of Minocin IV. The safety, tolerability and PK data will support the compound as a potential clinical candidate in Europe and allow recommendations of dose levels to be used in future registration studies.

Minocin IV is similar to other drugs developed in the era prior to the maturation of the field of pharmacodynamics, in that human doses were not systematically determined or optimized. An updated development program for Minocin IV would enable the availability of a new agent in the European Union (EU) to address highly resistant strains of *Acinetobacter baumannii*. The program will build on recently acquired information in preclinical studies and allow for optimization of the dosage regimen prior to use in controlled studies.

The overall development strategy in the EU is summarized below:

- Establish safety, tolerability, and PK of escalating doses of Minocin IV in normal subjects
- Link PK data in human subjects with PK-PD efficacy targets in animal models of infection to select dosage regimen
- Conduct a pathogen-directed clinical study of Minocin IV in infections due to *Acinetobacter baumannii*, assisted by the use of a rapid diagnostic test

PK-PD and Dosing Strategy. Studies of minocycline in 2 animal models of infection have been completed and identified the 24h free (non-protein bound) minocycline plasma AUC: MIC ratio as the PK-PD index associated with antimicrobial effects. These findings are consistent with studies with other tetracyclines/glycylcyclines. Studies in lung models of infection in rats show that an AUC:MIC ratio of 11 is required for bacterial stasis and a ratio of 18 is required for a 1 log drop in bacterial counts; these data and the current approved dosage regimens are consistent with the CLSI and US FDA breakpoint of 4 ug/ml.

There has been interest in the potential clinical uses of higher doses of minocycline for its properties unrelated to its antimicrobial effects (Garrido-Mesa, 2013), and resulted in studies of the pharmacokinetics and safety of intravenous minocycline at higher doses than those currently approved for treatment of infections. In a Phase 1 study in subjects with acute traumatic spinal cord injury, 52 subjects received IV minocycline in a dose escalation protocol (Casha, 2011). Treatment was given for 7 days (14 doses). The first five patients received 200 mg twice daily for 7 days. Steady state minocycline concentrations were 4.2 µg/mL within 48 hours. Subsequent patients were treated with a loading dose of 800 mg, with subsequent doses tapered by 100 mg every 12 hours until a dose of 400 mg was reached; the dose of 400 mg every 12 hours was maintained through Day 7. A mean steady-state serum minocycline concentration of 12.7 µg/mL was reported on this regimen. Two subjects died in the study, one in the placebo group (multiorgan failure and acute respiratory distress syndrome at Day 20) and one in the minocycline group (narcotic drug overdose at 6 months). The incidence of adverse events was not significantly different between the minocycline and placebo groups. One subject on minocycline developed elevated liver enzymes that normalized after discontinuation of drug, but was asymptomatic.

Sixty patients with acute ischemic stroke received total daily intravenous doses of up to 700 mg for 72 hours and serum PK data obtained (Fagan, 2010). No dose-limiting toxicities were observed. The plasma concentrations and exposure to minocycline increased in proportion to dose and were well-tolerated with doses up to 10 mg/kg/day (highest tested), or approximately 3.5x the current maximum labelled IV dose in the US; the mean 24h AUC was in the high dose group 348 mg-h/L. Adverse events were predominantly mild in severity, self-limiting, and were unrelated to minocycline dose. The most commonly reported adverse events outside of the cardiovascular system were headache (10%), nausea/vomiting/diarrhoea (9%), and local infusion reactions (8%).

The results of these studies indicate that doses of minocycline higher than those currently approved for the IV in the US (and oral in the US and EU) appear to be safe and well tolerated in humans. Based on these data, the sponsor believes it is rational to investigate higher doses of minocycline to optimize PK-PD properties for treatment of serious gram-negative pathogens, particularly non-fermenting organisms such as *Acinetobacter baumannii* complex.

This study is the first in the EU development program described above and will start with doses already approved in the US, namely 100 mg and 200 mg, in order to build a population PK model. Doses will be increased up to 600 mg if safety is observed at the lower doses.

5.4. Study Population

Healthy male and female volunteer subjects between 18 and 50 years of age, inclusive, will participate in the study. Six cohorts of approximately 10 subjects are planned. Each cohort should include at least three men and three women, with at least 1 man and 1 woman each receiving placebo.

6. TRIAL OBJECTIVES AND PURPOSE

6.1. Primary Objective

The primary objective is to assess the safety and tolerability of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects.

6.2. Secondary Objective

The secondary objective is to assess the pharmacokinetics of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects.

7. TRIAL DESIGN

7.1. Type/Design of Trial

This is a double-blind, randomized, placebo-controlled, single- and multiple ascending dose study of up to 5 doses/cohorts of Minocin IV.

Each cohort will consist of 10 subjects (8 active drug and 2 placebo).

Planned Cohorts:

- Cohort 1: 100 mg*
- Cohort 2: 200 mg*
- Cohort 3: 300 mg
- Cohort 4: 400 mg
- Cohort 5: 500/300 mg
- Cohort 6: 600/300 mg

* Cohorts 1 and 2 may be run concurrently as they are the approved doses of minocycline IV in the US. Cohorts 3, 4, 5 and 6 will be run sequentially.

Within each cohort, subjects will receive a single dose on Day 1, followed by 7 days of multiple-doses (Days 4-10), followed by a single dose on Day 11. Subjects will be discharged from the clinic on Day 14 and follow-up phone call will occur on Day 17 (See Schedule of Events for details).

Subjects are required to remain admitted in the research unit from the time of randomization until discharge on Day 14.

Subjects that discontinue the study early are required to complete Early Termination procedures.

Safety will be assessed throughout the study and serial blood and urine samples will be collected for the safety and PK assessment of Minocin IV.

7.2. Study Endpoints

The endpoints of this trial are:

- Assess the safety and tolerability of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects.
- Assess the pharmacokinetics of single and multiple intravenous doses of Minocin IV when administered to healthy adult subjects.

7.3. Measures to Minimize/Avoid Bias

7.3.1. Blinded Study Where Pharmacist is Unblinded

The study will be conducted using a double-blind design, with a placebo comparator. Specifics on how the blind for the study drug is maintained are provided in Section 9.4. Allocation of treatment is not disclosed to the blinded study team and subject. Study drug will be prepared by an unblinded pharmacist (or qualified designee) and will be provided to the blinded team in such a way that the appearance, volume, and infusion rate is the same for both study groups. Unblinded pharmacists (and qualified designees) will be required by signature to keep the study personnel blinded and will be prohibited from performing any study duties other than prepare study drug and associated tasks (e.g., drug accountability, etc.).

8. SUBJECT POPULATION

8.1. Number of Subjects

Sixty subjects will be studied at one study center. Each randomized subject can be part of one cohort only, i.e. cannot be randomized twice. Subjects who drop-out/are withdrawn before taking any study drug may be replaced at the discretion and agreement of the site and Sponsor.

8.2. Inclusion Criteria

Subjects must meet all of the following criteria in order to be eligible for the study:

1. A signed informed consent form, the ability to understand the study conduct and tasks that are required for study participation, and a willingness to cooperate with all tasks, tests, and examinations as required by the protocol, whether in the research unit or after discharge, for the duration of the study;
2. Male or female between 18 and 50 years of age inclusive;
3. Subject has a body mass index (BMI) ≥18 kg/m2 and ≤ 30kg/m^2;
4. Subject is non-smoker or smokes up to 5 cigarettes per day (or equivalent);
5. Subject is in good health based on medical history and physical examination findings and has no clinically meaningful safety laboratory abnormalities (Hematology, blood chemistry, and urinalysis) or 12-lead ECG results, as assessed by the Principal Investigator (PI);
6. Vital signs (BP, pulse and temperature) measured at screening/baseline must be within the following ranges: SBP ≥90 to ≤150 mm Hg, DBP ≥45 to ≤90 mm Hg; Heart Rate ≥ 45 to ≤90 bpm (taken after resting in a supine position for at least 5 minutes);
7. Expectation that intravenous access will be sufficient to allow for ease of study drug infusion, and for all protocol required blood sampling to take place;
8. Subject commits to remaining admitted in the research unit for the course of the study;
9. Female subject is surgically sterile, postmenopausal: period of amenorrhea for at least 2 years, or if of childbearing potential, agrees to abstinence or to use at least 2 acceptable methods of birth control (e.g. prescription oral contraceptives, contraceptive injections, contraceptive patch, intrauterine device, barrier methods, etc.) or male partner sterilization alone, between the first dose (Day 1) and for 90 days after the completion of the study.

8.3. Exclusion Criteria

Subjects who meet any of the following exclusion criteria at baseline will not be enrolled in the study:

1. Has any condition, including findings in the medical history or in pre-study assessments that constitutes a risk or a contraindication for the participation in the study or completing the study;
2. Positive breath test for alcohol and/or positive urine test for drugs of abuse at Screening and Day -1 Visits;
3. Has a history or presence of alcohol/drug abuse within 2 years. Alcohol abuse is defined as regularly consuming >3 units/day (21 units per week for men), >2 units/day (14 units/week) for women. A unit is defined as a can of 4% beer (330 mL), approximately 190 mL of 6-7% beer (malt liquor), a glass of 40% spirits (30 mL), a glass of wine (100 mL);
4. Subject shows positive hepatitis B surface antigen (HBsAg), hepatitis C virus antibodies (anti-HCV), or human immunodeficiency virus (HIV) I/II antibodies and antigen tests;
5. Subject has active or ongoing candida infection;
6. Blood or plasma donation within past 2 months;
7. Females who are pregnant or nursing or who have a positive pregnancy test result at the Screening or Day -1 Visits;
8. Males who are unwilling to practice abstinence or use an acceptable method of birth control during the entire study period and for 90 days after the completion of the study (i.e. condom with spermicide, where locally available);
9. Presence of known raised intracranial pressure;
10. Use of retinoids (e.g., Isotretinoin);
11. History of significant hypersensitivity or allergic reaction to any of the tetracycline class of antibiotics or the components of those antibiotics;
12. Receipt of any investigational medication or investigational device during the last 30 days prior to randomization;
13. Treatment with any prescription, vitamins or OTC drugs, within 2 weeks or five half-lives, whichever is longer, or herbal nutritional supplements within 2 weeks of screening, with the exception of acetaminophen/paracetamol for minor headache. Subjects will not be allowed to receive medications for the duration of the study (except the abovementioned acetaminophen/paracetamol). Birth control or other hormone replacement is also permitted as long as it has been taken at a stable dose for at least three months before the Screening Visit and remains stable for the duration of the study;
14. A QTcF of >480 msec;
15. Calculated creatinine clearance less than 50 mL/min (Cockcroft-Gault method) at screening or check-in (Day -1);

16. Unable or unwilling, in the judgment of the investigator, to comply with the protocol;
17. An employee of the investigator, the study center, the sponsor or The Medicines Company with direct involvement in the proposed study or other studies under the direction of that investigator or study center, or a family member of the employee or the investigator;
18. Prior enrollment in any minocycline study including prior cohorts in this trial.

8.4. Withdrawal Criteria

All subjects have the right to withdraw at any point during treatment without prejudice. The investigator can discontinue any subject at any time if medically necessary. It will be documented whether or not each subject completed the clinical study. If for any subject, study treatment or observations were discontinued, the reason will be recorded and the Sponsor should be notified promptly. Reasons that a subject may discontinue participation in a clinical study are considered to constitute one of the following:

1. Any SAE, clinically significant AE, severe laboratory abnormality, intercurrent illness, or other medical condition that indicates to the PI that continued participation is not in the best interest of the subject, that exposes the subject to substantial risk and/or does not allow the subject to adhere to the requirements of the protocol.
2. Subject's decision to withdraw.
3. Requirement of prohibited concomitant medication.
4. Physician’s decision.
5. Subject’s failure to comply with protocol requirements or study related procedures.
6. Lost to follow-up.
7. Termination of the study by the Sponsor, or designee, ARO or regulatory authorities.

It is imperative to obtain complete follow-up data for all subjects whether or not they receive their assigned treatment or have discontinued study drug. All data collected up until the time of subject withdrawal is to be entered into the eCRF. In addition, every attempt should be made to collect follow-up information except for those subjects who specifically withdraw consent for release of such information. All procedures and laboratory specimens or tests requested for evaluation following administration of the study drug should be carried out when possible whether or not a subject continues to receive treatment according to the protocol.

The clinical report will include reason(s) for subject withdrawals as well as details relevant to the subject withdrawal. If a subject is withdrawn from the trial prior to study completion, the subject will undergo all procedures scheduled for study completion (end of study evaluations) as appropriate (see Section 10.2.14). Any subject withdrawn due to an AE (whether serious or non-serious) or clinically significant abnormal laboratory test values will be evaluated by the PI or a monitoring physician and will be treated and/or followed up until the symptoms or values return to normal or acceptable levels, as judged by the PI.

Subjects who drop-out/are withdrawn may be replaced at the discretion and agreement of the site and Sponsor. Randomization numbers for replacements will be generated by Biostatistics.

9. TREATMENT OF SUBJECTS

9.1. Study Medications

Each subject will receive a single dose of Minocin IV or placebo, followed by multiple doses of Minocin IV or placebo, given q12h (+/- 1 h) over 60 minutes in 0.9% Sodium Chloride Injection (referred to as Normal Saline for the remainder of the document) for 7 days and then a single dose on day 11.

9.1.1. Minocin IV

Minocin IV will be supplied as a sterile lyophilized powder in single-use 10 mL glass vials. Each vial contains 108 mg of minocycline hydrochloride equivalent to 100 mg of minocycline.

Each dose will be reconstituted and prepared in Sodium Chloride Injection USP bag per the Table below and administered as a constant rate IV infusion over 1 hour via a single dedicated peripheral venous line. Details of Minocin IV preparation and administration are included in the study Pharmacy Manual.

9.1.2. Placebo

Sodium Chloride Injection USP (placebo) is administered at a rate and volume matching the rate and volume of Minocin IV for each respective Cohort. Dosing is to the same schedule as subjects randomized to Minocin IV. Details of placebo preparation and administration are included in the study Pharmacy Manual.

9.1.3. Packaging and Labeling

Minocin IV will be provided by the Sponsor. Infusion bags of normal saline will be provided by the study site pharmacy.

Medication labels will comply with regulatory requirements.

9.1.4. Storage

Minocin IV will be stored in a secure area at a controlled room temperature of 20° to 25°C. Once diluted into an IV bag, Minocin IV may be stored at either room temperature for up to 4 hours or refrigerated at 2 to 8C for up to 24 hours. Access should be strictly limited to the study pharmacists or designees.

Normal Saline should be stored per the manufacturer's instructions. Normal saline used as placebo should be treated the same as Minocin IV once it leaves the pharmacy to ensure blinding is maintained (e.g., refrigerate, etc.).

Further details of Minocin IV and normal saline storage can be found in the Pharmacy Manual.

9.1.5. Accountability

The investigator or designee must maintain an inventory record of Minocin IV received and all doses administered to assure the regulatory authorities and the Sponsor that the study drug will not be dispensed to any person who is not a subject under the terms and conditions set forth in this protocol. Study drug accountability forms and specific instructions can be found in the Pharmacy Manual.

The Minocin IV supplied for use in this study is to be prescribed only by the Principal Investigator or designated sub-investigators and may not be used for any purpose other than that outlined in this protocol.

During the study all used Minocin IV vials will be kept until the monitor has reviewed the accountability records.

All unused Minocin IV will be destroyed on site once it has been inventoried and the monitor has reviewed the accountability records. In the event that Minocin IV needs to be returned for any other reason, the site will receive a written request listing the lot number(s) to be returned and the reason for the return request.

9.1.6. Product Complaints

Sites are required to report any product complaints to MDCO immediately but no later than 24 hours from the time of awareness, by phone or e-mail as follows:

United States of America: +1-888-977-6326
Contact information for all other geographic areas:
www.themedicinescompany.com/contact/global-medical-info
Email: Medical.information@themedco.com

Product Complaint: Is defined as any written, electronic, or oral communication that alleges deficiencies related to the identity, durability, reliability, quality, safety, effectiveness or performance of a product, after it is released for distribution (EU DIR 2001/83/EC). (Derived from Ref US 21 CFR 211.198).

There are two types of Product Complaints: **Technical Quality Complaints**: A report of dissatisfaction with product with regard to its efficacy, strength, integrity, purity, or quality; thus a potential failure to meet product specifications. **Preference Complaints**: A report of dissatisfaction with service, delivery, packaging or other preference.

Technical Quality Complaint: A report of dissatisfaction with the product with regard to its efficacy, strength, integrity, purity, or quality; thus a potential failure to meet product specifications. Examples include:

- An indication that there is an unexpected physical change in the drug product such as discoloration, change in shape of the drug product, presence of particulates or any other physical change that might indicate contamination, a manufacturing defect or any other event that might indicate a compromise in product quality.
- An indication that the content does not meet its labeled volume, count, etc.
- An indication that there is an unexpected physical change in any part of the container (this includes the bottle, any part of the seal, the cap or the label).
- An indication that the product is mislabeled.
- An indication that there is an unexpected physical change of the product or container once the product is diluted or reconstituted (the container includes the vial, bag, IV line, syringe or any other item that is in contact with the product).
- An indication that the product is falsified, tampered with or adulterated.

9.2. Concomitant Medications

All medications taken during the 14 days prior to dosing will be recorded and reviewed by the PI or their designee.

9.2.1. Prohibited Concomitant Medications

During the study, 2 grams per day or less of acetaminophen/paracetamol may be administered orally at the discretion of the PI or their designee for intercurrent illness or AEs. If needed on dosing day, acetaminophen/paracetamol should not be taken until at least 2 hours after dosing.

Vitamin supplements will be prohibited for 14 days prior to Day 1 until discharge.

All medication (other than study drug, acetaminophen/paracetamol or birth control and hormone replacement therapy) is prohibited during the study. If

drug therapy other than that specified by the protocol is taken, a joint decision will be made by the PI and Sponsor to continue or discontinue the subject.

9.3. Restrictions

Beverages containing alcohol should not be consumed 48 hours or less prior to Day -1 until the end of the study.

Subjects that smoke more than 5 cigarettes per day will be excluded from the study.

Subjects participating in strenuous exercise or sports as assessed by the PI will be excluded from the study.

Extreme exposure to the sun or sunbathing has to be avoided, as well as the use of tanning devices (e.g. sunbed, solarium) from screening until 7 days after discharge.

9.4. Blinding

9.4.1. Blinding of study medications

The study drug will be prepared by an unblinded pharmacist (or qualified unblinded designee) in a secure area where blinded staff are not present. Once prepared, the IV bags containing the study drug will be labeled without revealing the treatment. Additional steps may be required to ensure that the blinded staff do not become unblinded by the color of the IV solution (e.g., colored IV tubing, IV bag covers, etc.) which are described in detail in the study Pharmacy Manual.

9.4.2. Method and Maintenance of Blinding

This is a double-blind study, other than the unblinded statistician (or designee) who prepares the randomization list and the unblinded study pharmacist (and designee(s). All others will be blinded, including, but not limited to: PI, site staff, subjects, Sponsor and third party vendors.

9.5. Unblinding

One set of sealed, printed code break slips containing the randomization code for each subject will be supplied.

Breaking of the blind is expressly forbidden except in the event of a medical emergency where the identity of the drug must be known to properly treat the subject or to assess the stopping rules.

In the event of a medical emergency, it is requested that the investigator make every effort to contact the study Medical Monitor or designee prior to breaking the blind. If breaking the blind is required because of a medical emergency, the

treatment identity would be revealed for that subject only. In the event that the emergency is one in which it appears that the other subjects may be at imminent risk, the blind may be broken for all subjects dosed at that dose level.

In all cases where the blind is broken, the investigator should record the date, reason for blind breaking and sign his/her name.

At the end of the study, the envelopes will be reviewed by the sponsor at a designated unblinded monitoring visit.

The study will be unblinded at the end of the study post clinical database lock or as needed after the completion of a cohort for review of AEs.

10. SCHEDULE AND SEQUENCE OF PROCEDURES

The Study Schematic Tables are outlined in Sections 2 and 3, which summarize the study assessments by time point.

10.1. Informed consent

Prior to any study-related activities, an Informed Consent Form (ICF) and subject information sheet, if applicable, approved by a regional ethical or institutional review board must be signed and personally dated by the subject. The format and content of the ICF and subject information sheet must be agreed upon by the Principal Investigator(s), appropriate regional ethical or institutional review board, and MDCO, or designee. The subject's original signed and dated ICF (together with any subsequent amended versions approved by the regional ethical or institutional review board) must be retained by the investigator in the subject's file. A copy of the original signed and dated ICF and subject information sheet must be given to the subject.

10.2. Subject Confinement

Subjects will be admitted to the study on Day 1 and will remain in the research unit through completion of all scheduled procedures on Day 14. Subjects who leave the research unit prior to Day 14 will be considered Early Terminations unless permission is granted by the PI and sponsor due to extraordinary circumstances.

10.2.1. Screening Period (Days -28 to -1)

Once consent is obtained, screening may begin within 28 days prior to first dosing. The subject informed consent form will be obtained prior to any study procedures being performed. Subjects will have to meet all eligibility criteria before being enrolled in the study.

The following assessments/procedures will be performed at the screening visit:

- Informed consent administered
- Inclusion/exclusion criteria assessment
- Medical history and Demographics will be recorded
- Review of concomitant medications
- Height, weight, and BMI calculated
- Physical examination

- Vital signs: systolic and diastolic blood pressure, pulse, respiratory rate, and temperature
- 12-lead ECG
- Hematology, serum chemistry, and urinalysis
- Urine drug/alcohol screen
- Serum pregnancy test (women of childbearing potential only)

10.2.2. Check-in Procedures (Day -1)

Day -1 procedures should be completed within approximately 24 hours before dosing on Day 1 to obtain baseline values. Baseline data may be captured pre-dose on Day 1 at the site's discretion.

Subject eligibility will be reviewed for each subject to ensure that subjects remain eligible for the study since the Screening Visit.

The following assessments/procedures will occur on Day -1:

- Review of inclusion/exclusion criteria
- Review of concomitant medications
- Review medical history and demographics
- Weight and BMI (calculated using height from Screening)
- Physical examination
- Vital signs: systolic and diastolic blood pressure, pulse, temperature and respiratory rate
- 12-lead ECG (performed in triplicate)
- Hematology, serum chemistry, and urinalysis collection
- Urine drug screen
- Alcohol screen
- Serum pregnancy test (women of childbearing potential only)

10.2.3. Randomization (Day 1)

Randomization should only occur once subject eligibility is confirmed and after all screening and Day -1 medical/physical assessments and laboratory tests have been completed (with lack of exclusion criteria or contraindications to treatment identified).

10.2.4. First Single-dose Treatment (Day 1)

Within each cohort, subjects will receive a single dose of either study drug or placebo (prepared by an unblinded pharmacist or qualified designee) on Day

1.

Day 1 **Pre-Dose** Assessments:

- Ensure all baseline procedures noted in Section 10.2.2 (Day -1) have been completed within 24 hours of dosing
- Urine pregnancy test (women of childbearing potential only)
- Blood collection for pharmacokinetic assessment within 24 h of dosing
- Urine collection for pharmacokinetic assessment within 24 h of dosing
- Dose administration

Day 1 **Post-Dose** assessments:

- Vital signs at: 1, 3, and 6 h after the start of dosing
- Blood collection for pharmacokinetic assessment at: **1 h** (end-of-infusion), **2, 4, 8, 12,18,** 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 1-4)
- Urine collection for pharmacokinetic assessment during intervals: **0-4, 4-8, 8-12, 12-24***, 24-48 and 48-72 hours after dosing (ongoing Days 1-4)
- Urine collected will also be used to determine 24 h creatinine clearance
- 12-lead ECG at 1 hour (+/-10 minutes) after dosing/end of infusion (performed in triplicate)
- Review of adverse events
- Review of concomitant medications

10.2.5. Day 2 Procedures

The following procedures will occur on Day 2:

- Vital signs, collected once in the morning: systolic and diastolic blood pressure, pulse, temperature and respirations
- Hematology, serum chemistry, and urinalysis
- Blood collection for pharmacokinetic assessment at: 1 h (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 1-4)
- Urine collection for pharmacokinetic assessment during intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing (ongoing Days 1-4)
- Review of adverse events

- Review of concomitant medications

10.2.6. Day 3 Procedures

The following procedures will occur on Day 3:

- Vital signs, collected once in the morning: systolic and diastolic blood pressure, pulse, temperature and respirations
- Blood collection for pharmacokinetic assessment at: 1 h (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 1-4)
- Urine collection for pharmacokinetic assessment during intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing (ongoing Days 1-4)
- Physical examination
- Review of adverse events
- Review of concomitant medications

10.2.7. Day 4 Procedures

The following procedures will occur on Day 4:

- Vital signs before and at 1, 3, and 6 h after the start of the morning dose and before and at 1 hour after the start of the evening dose: systolic and diastolic blood pressure, pulse, temperature and respirations12-lead ECG: before and 1 h (+/- 10 minutes)/end of infusion after the start the first dose (performed in triplicate)
- Dose administration q12 hours (+/- 1 h) (**Note:** the first dose on Day 4 will occur once the 72 h post- (Day 1) dose PK collection has been completed)
- Hematology, serum chemistry, and urinalysis: approximately 4 hours after the start of the morning dose
- Blood collection for pharmacokinetics assessment. **Note:** The 72-hour sample for Day 1 dosing is also the pre-dose sample for the Day 4 morning dose. Samples are also taken at 1 h (end-of-infusion), 2, 4, 8, and 12 hours after the start of the Day 4 morning dosing (the 12-hour post-dose PK collection should occur before the start of the second infusion on Day 4).
- Final urine collection for pharmacokinetics assessment of the Day 1 dosing (this is the end of the Day 1 dosing collection period, 48-72 h. There is no requirement for Urine PK to be collected for the multiple-dose period).
- Review of adverse events

- Review of concomitant medications

10.2.8. Days 5, 7, and 9 Procedures

The following procedures will occur on Days 5, 7, and 9:

- Vital signs before and at 1, 3, and 6 h after the start of the morning dose and before and at 1 hour after the start of the evening dose: systolic and diastolic blood pressure, pulse, temperature and respirations
- 12-lead ECG: before and 1 h/end of infusion (+/-10 minutes) after the start of the morning (AM) dosing
- Dose administration (q12h - +/- 1 h)
- Physical examination (Day 9 only)
- Review of adverse events
- Review of concomitant medications

10.2.9. Days 6 and 8 Procedures

The following procedures will occur on Days 6 and 8:

- Vital signs before and at 1, 3, and 6 h after the start of the morning dose and before and at 1 hour after the start of the evening dose: systolic and diastolic blood pressure, pulse, temperature and respirations
- 12-lead ECG: before and 1 h/end of infusion (+/-10 minutes) after the start of the morning (AM) dosing
- Hematology, serum chemistry, and urinalysis: approx. 4 hours after the start of the morning (AM) dose
- Dose administration (q12h - +/- 1 h)
- Review of adverse events
- Review of concomitant medications

10.2.10. Day 10 Procedures

The following procedures will occur on Day 10:

- Weight and BMI (calculated using height from Screening)
- Vital signs before and at 1, 3, and 6 h after the start of the morning dose and before and at 1 hour after the start of the evening dose: systolic and diastolic blood pressure, pulse, temperature and respirations
- 12-lead ECG: before and 1 h/end of infusion (+/-10 minutes) after the start

of the morning (AM) dosing

- Hematology, serum chemistry, and urinalysis: approx. 4 hours after the start of the morning dose
- Dose administration (q12h - +/- 1 h)
- Review of adverse events
- Review of concomitant medications

10.2.11. Second/Final Single-dose Treatment (Day 11)

The following procedures will occur on Day 11:

- Vital signs before and at 1, 3, and 6 h after the start of dose: systolic and diastolic blood pressure, pulse, temperature and respirations
- 12-lead ECG: before and 1 h/end of infusion (+/-10 minutes) after the start of dosing
- Blood collection for pharmacokinetics assessment: taken **before** the Final Dose and at **1** h (end-of-infusion), **2, 4, 8, 12, 18**, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 11-14)
- Urine collection for pharmacokinetic assessment **pre-dose** and during intervals: **0-4, 4-8, 8-12, 12-24**, 24-48 and 48-72 hours after dosing (ongoing Days 11-14)
- Dose administration
- Review of adverse events
- Review of concomitant medications

10.2.12. Day 12 Procedures

The following procedures will occur on Day 12:

- Vital signs: systolic and diastolic blood pressure, pulse, temperature and respirations
- Blood collection for pharmacokinetics assessment: taken before the Final Dose and at 1 h (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 11-14)
- Urine collection for pharmacokinetic assessment during intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing (ongoing Days 11-14)
- Review of adverse events
- Review of concomitant medications

10.2.13. Day 13 Procedures

The following procedures will occur on Day 13:

- Vital signs: systolic and diastolic blood pressure, pulse, temperature and respirations
- Hematology, serum chemistry, and urinalysis
- Blood collection for pharmacokinetics assessment: taken before the Final Dose and at 1 h (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 11-14)
- Urine collection for pharmacokinetic assessment during intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing (ongoing Days 11-14)
- Review of adverse events
- Review of concomitant medications

10.2.14. Discharge (Day 14)

Subjects will be discharged from the clinic on Day 14. Prior to discharge, the following procedures will occur:

- Weight and BMI (calculated using height from Screening)
- Physical examination
- Vital signs: systolic and diastolic blood pressure, pulse, temperature and respirations
- 12-lead ECG
- Hematology, serum chemistry, and urinalysis
- Serum pregnancy test (women of childbearing potential only)
- Blood collection for pharmacokinetics assessment: taken before the Final Dose and at 1 h (end-of-infusion), 2, 4, 8, 12, 18, 24, 36, 48 and 72 hours after the start of dosing (ongoing Days 11-14)
- Urine collection for pharmacokinetic assessment during intervals: 0-4, 4-8, 8-12, 12-24, 24-48 and 48-72 hours after dosing (ongoing Days 11-14)
- Review of adverse events
- Review of concomitant medications

10.2.15. Follow-up Phone Call (Day 17)

A Day 17 follow-up phone call will be performed to evaluate safety by the assessment of adverse events and serious adverse events. A subject's participation in the study is complete when:

- All procedures at the last study visit (Day 14) have been completed
- All procedures for the follow-up phone call (Day 17) have been completed
- All AEs/SAEs have been followed to resolution

10.2.16. Early Termination

An Early Termination Visit is required for any subject that discontinues the study prior to Day 14. All Day 14 visit procedures should occur during the Early Termination Visit, as well as the collection of any PK samples (if applicable). A follow-up phone call should occur approximately 3 days from the Early Termination Visit.

11. PROTOCOL ASSESSMENTS

11.1. Assessment of Safety

11.1.1. Data Review Meetings

Intra-Cohort Data Review:

For Cohorts 3-6, single dose safety data will be reviewed by the Medical Monitor and Principal Investigator on Day 3 prior to the start of the multi-dosing on Day 4. During the review, blinded safety data from the single dose infusions will be assessed and one of the following will be decided before the start of the Multiple Dose Phase on Day 4:

- To continue with the Multi-dose phase as planned,
- To continue with the Multi-dose phase and add additional safety evaluations,
- To not continue with the Multi-dose phase.

Between- Cohort Data Monitoring Committee Review:

Safety data will be reviewed by the Data Monitoring Committee (DMC) following each cohort and prior to the start of the next cohort.

After completion of cohorts 1 and 2, which may run concurrently, and prior to dosing of each cohort 3, 4, 5 and 6, the DMC Sponsor will review all pertinent blinded safety data (e.g., physical examinations, vital signs, electrocardiograms (ECGs), clinical laboratory tests, and AEs) from the preceding cohort to determine one of the following:

- To continue with the next cohort as planned,
- To continue with the next cohort and add additional safety evaluations,
- To continue with the next cohort by adjusting the planned dose of Minocin IV, or
- To not continue with the next cohort.

11.1.2. Stopping Rules

The study will be terminated if ≥ 2 subjects in a cohort meet any of the following criteria and the subject was determined to have received active drug after unblinding (see 9.5 Unblinding):

- Has a drug related SAE
- Experiences a drug related AE grade ≥ 3 toxicity

A written statement fully documenting the reasons for study termination will be provided to the HREC/IEC.

11.1.3. Adverse Events

Subjects will be carefully monitored for adverse events by the investigator during the designated study period (see Section 12 for details).

11.1.4. Body Mass Index (BMI)

BMI will be calculated using the following equation:

BMI= Weight (kg)/Height (m^2)

11.1.5. Vital Signs

Vital signs assessments will include systolic and diastolic blood pressure, pulse, body temperature, and respirations.

Subjects should be resting in a supine position for at least 5 minutes prior to and during vital signs measurements.

A +/- 10 minute time window is permitted.

11.1.6. Physical Exam

Physical examinations include, at a minimum, assessment of the following systems: skin, head, ears, eyes, nose and throat, respiratory system, cardiovascular system, gastrointestinal system, neurological condition, blood and lymphatic systems, and the musculoskeletal system. A licensed physician will conduct the examinations.

Physical examination may be performed at various unscheduled time points if deemed necessary by the investigator.

11.1.7. ECG

12-lead ECGs will be assessed.

Triplicate 12-lead ECGs each separated by at least 1 minute will be taken at Day -1 and post- dose at Day 1. Triplicate 12-lead will also be taken at Day 4 pre-dose and post-dose (for the morning dose). ECGs will be taken following resting in the supine position for 5 minutes. The average value for the triplicate will be utilized for assessing QTcF exclusion criteria.

All subsequent post-dose ECGs will be single readings. However, if one of the following occurs, then the ECG will be repeated in triplicate (separated by at least 1 minute):

- QTcF increase from baseline triplicate average > 30msec

- QRS duration > 130ms
- PR interval > 240ms

If the "recheck" triplicate's average is still above these parameters, then the investigator shall be notified for decision on further action.

The same model of ECG machine should be used for check-in and post-dose readings for all subjects.

A +/- 10 minute time window is permitted, -1 hour for pre-dose assessment.

ECGs will be interpreted and signed and dated by the PI, or qualified designee. The ECGs will be classified as normal, having a clinically insignificant abnormality (NCS), or having a clinically significant abnormality (CS). In addition, ECG parameters of ventricular rate, PQ or PR interval, QRS duration, and QT interval (corrected and uncorrected) will be noted on the eCRF.

All CS findings will be recorded as AEs.

11.1.8. Clinical Laboratory Tests

Clinical laboratory tests will be conducted at the site's local lab. Lab results and lab reports will be kept as source documents. Subjects will be required to fast for at least 4 hours before the clinical laboratory tests. Abnormal lab ranges must be documented as clinically significant or non-clinically significant by a study investigator and filed with the source documents.

At a minimum the following lab tests should be performed at the designated time points:

11.1.8.1. Hematology

Hemoglobin, Hematocrit, White blood cell count (with automated differential), Red blood cell count, Platelet count, Prothrombin time (PT), Activated partial thromboplastin time (aPTT).

11.1.8.2. Chemistry

Blood urea nitrogen, Serum creatinine, Total bilirubin, Direct bilirubin, Alkaline phosphatase, Aspartate aminotransferase, Alanine aminotransferase, Albumin, Total protein, Glucose, Calcium, Chloride, Sodium, Magnesium, Potassium, Uric acid, Lactate dehydrogenase, Bicarbonate, Phosphorus.

11.1.8.3. Urinalysis

Midstream urine sample analyzed by dipstick (Note: microscopic examination of sediment will only occur if a strong positive result is observed).

11.1.8.4. Human Chorionic Gonadotropin (Pregnancy Test)

Pregnancy tests are required for women of childbearing potential only. Serum pregnancy should be performed during Screening, Day -1 and 14. Urine pregnancy test should be performed before dosing on Day 1.

11.1.8.5. Urine Testing for Drugs of Abuse

Amphetamines, barbiturates, benzodiazepines, cannabinoids, cocaine, methadone and opiates.

11.1.8.6. Breath Alcohol Testing

Breath alcohol test will be performed on all subjects.

11.1.8.7. Creatinine Clearance

Urine will be obtained to measure 24-hour creatinine clearance on Day 1 only. Serum creatinine concentrations will also be measured from the clinical laboratory tests.

11.1.9. Meals

Standard meals will be provided uniformly to all subjects at approximately 1 hour prior to dosing, and at approximately 4 and 9 hours after dosing on Day 1 and Day 11. On all other study days, breakfast, lunch, dinner, and evening snacks will be served at appropriate times.

Water will be allowed as desired at all times. To encourage urine production, subjects will be asked to drink ~240 mL of water at 2 hours after start of dosing on Days 1 and 11.

11.2. Assessment of Efficacy

Efficacy will not be assessed in this study.

11.3. Assessment of Pharmacokinetics

The PK sample collection times are all from the start of the study drug infusion.

11.3.1. Sampling and Processing

For the PK blood samples, the following deviation windows from the actual sampling times are permitted:

Nominal Time	Reporting Standard
0-4 h	+/- 2 minutes
>4 h	+/- 5 minutes
>12 h	+/- 10 minutes

After collection of the PK samples, blood will be centrifuged and plasma collected, and frozen. For specific collection and storage procedures, please refer to the Study PK Manual.

11.3.2. Urine Sampling and Processing

Urine PK samples will be collected at designated time points. To begin the 0-4 h post-dose collection, subjects will void completely within 15 min prior to dosing. Subjects will be encouraged to void at the end of each collection interval. The time of each void should be documented.

A +/- 20 minute time window is permitted following scheduled collection time.

Subjects should be encouraged to drink water during the urine PK collection periods.

Urine will be refrigerated during the collection intervals. At the end of each interval, total urine volume will be measured and recorded. For specific collection and storage procedures, please refer to the Study PK Manual.

12. ADVERSE EVENTS

12.1. Definitions

12.1.1. Adverse Event

Adverse Event (AE): Any untoward medical occurrence in a patient or clinical trial subject administered a medicinal product and that does not necessarily have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (e.g. an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Planned hospital admissions and/or surgical operations for an illness or disease that existed before the study drug was given or the subject was randomized in a clinical study are not to be considered AEs.

Adverse events or abnormal test findings will be followed until the event (or its sequelae) or the abnormal test finding resolves or stabilizes at a level acceptable to the Sponsor/Investigator.

12.1.1.1. AE Severity

The severity of an AE will be assessed by the investigator. The investigator should ensure that any subject experiencing an AE receives appropriate medical support until the event resolves.

The investigator and/or sub investigator will classify the severity of AEs according to the current version of Common Terminology Criteria for Adverse Events (CTCAE) as follows:

Grade 1 = mild

Grade 2 = moderate

Grade 3 = severe

Grade 4 = life-threatening

Grade 5 = death related to AE

If a condition cannot be identified in the CTCAE, the following definitions will be used:

Mild: asymptomatic or mild symptoms OR clinical or diagnostic observations only OR intervention not indicated.

Moderate: minimal, local or noninvasive intervention indicated OR limiting age appropriate instrumental activities of daily living (e.g., preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.).

Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated OR disabling OR limiting self-care activities of daily living (e.g., bathing, dressing and undressing, feeding self, using the toilet, taking medications, not being bedridden).

Life-threatening: urgent intervention indicated.

12.1.1.2. Study Drug Causality

The relationship of an AE to study drug will be assessed with consideration to the following criteria:

- Temporal relationship to the initiation of study medication
- Response of the event to withdrawal of study medication
- AE profile of concomitant therapies
- Clinical circumstances during which the AE occurred
- Subject's clinical condition and medical history

Categorization of causality will be designated by the investigator as stated below:

Reasonable possibility - There are facts (evidence) or arguments to suggest a causal relationship between the event and the IMP.

No Reasonable possibility - There are few to no facts (evidence) or arguments to suggest a causal relationship between the event and the IMP.

12.1.2. Serious Adverse Event

A serious adverse event (SAE) is any untoward medical occurrence that at any dose:

- Results in death,
- Is life-threatening, i.e. the subject was, in the opinion of the investigator, at immediate risk of death from the event as it

occurred (it does not include an event that, had it occurred in a more severe form, might have caused death),

- Results in a significant, persistent or permanent change, impairment, damage or disruption in the subject's body function/structure, physical activities and/or quality of life,
- Requires in-subject hospitalization or prolongs hospitalization,
- Is a congenital anomaly/birth defect, or
- Is another medically significant event where medical and scientific judgement should be exercised in deciding whether other situations should be considered serious reactions, such as important medical events that might not be immediately life threatening or result in death or hospitalization but might jeopardize the patient or might require intervention to prevent one of the other outcomes listed above. 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 hospitalization or development of dependency or abuse. Any suspected transmission via a medicinal product of an infectious agent is also considered a serious adverse reaction.

A distinction should be drawn between serious and severe AEs. Severity is an estimate or measure of the intensity of an AE, while the criteria for serious AEs are indications of adverse subject outcomes for regulatory reporting purposes. A severe AE need not necessarily be considered serious and a serious AE need not be considered severe. For example, nausea that persists for several hours may be considered severe nausea, but not an SAE. On the other hand, a myocardial infarction (MI) that may be considered minor could also be an SAE if it prolonged hospitalization.

12.1.3. Medication Errors

Medication error refers to any unintended error in the dosing and/or administration of the study drug as per instructions in the protocol. Medication errors generally fall into 4 categories as follows:

- wrong study drug
- wrong dose (including dosing regimen, strength, form, concentration, amount);
- wrong route of administration;
- wrong patient (i.e. not administered to the intended patient)

Medication Errors include occurrences of overdose and under dose of the study drug, and abuse and misuse.

Overdose: Unintentional administration of a quantity of the study drug given per administration or per day which is above the maximum recommended dose according to the reference safety information or protocol for the study drug. This also takes into account cumulative effects due to overdose.

Under dose: Unintentional administration of a quantity of the study drug given per administration or per day which is under the minimum recommended dose according to the reference safety information or protocol.

Abuse of a medicinal product: Persistent or sporadic, intentional excessive use of medicinal products which is accompanied by harmful physical or psychological effects.

Misuse: Intentional and inappropriate use of a study drug not in accordance with the prescribed or authorized dose, route of administration, or not within the legal status of its supply.

12.1.4. Adverse Event of Special Interest (AESIs)

An adverse event of special interest (serious or non-serious) is one of scientific and medical concern specific to the Sponsor's study drug or program, which warrants ongoing monitoring and rapid communication by the investigator to the sponsor. Such an event might warrant further investigation in order to characterize and understand it.

There are no AESIs identified for the study drug Minocin IV in this protocol.

12.1.5. Special Situations

Special Situations is a comprehensive term that encompasses safety information related to products for which global regulations require collection, evaluation, and/or reporting to regulatory authorities.
Additional special situations not previously defined include the following:

- Suspected transmission via a medicinal product of an infectious agent
- Drug interactions
- Occupational exposure

12.2. Procedure for Non-Serious Adverse Event Recording

All non-serious AEs that occur between first administration of study medication and the completion of the Day 17 follow up must be assessed and recorded on the source documents and eCRF, regardless of causal relationship to the study drug.

12.3. Procedure for Serious Adverse Event Reporting

Note: This procedure may need also to be followed for reporting Special Situations.

All SAEs that occur between first administration of study medication and the completion of the Day 17 follow up must be reported to the Sponsor's Global Pharmacovigilance Department (GPV) within 24 hours of awareness of the event using the provided study specific SAE Report Form. In addition to completing the SAE Report Form, each SAE must be entered on the appropriate page of the eCRF.

When death occurs with an SAE, the cause of death must be reported as an SAE. "Fatal" will be reported as the outcome for these events.

The investigator must assess the causality for each SAE.

The Sponsor will contact the investigator, if necessary, to clarify any of the event information. The investigator should provide any follow-up information for the event to the Sponsor on an updated SAE report form as soon as it becomes available.

If the investigator is notified of a SAE that occurs post-study period, that he or she wishes to report to the Sponsor (e.g., an event suspected to be causally related to study drug), the event should be reported through the process described above.

Where appropriate, if required by local regulations or procedures, the investigator should report these events to the Ethics Committee (EC) and/or national regulatory authority in addition to the Sponsor.

12.4. Procedure for Medication Error Reporting for Study Drugs

Medication errors with or without an associated AE need to be recorded as medication errors in the eCRF as described in Section 12.1.3.

Medication errors with an associated SAE need to be recorded as medication errors in the eCRF and reported to the Sponsor's Global Pharmacovigilance Department as described in Section 12.3.

A mis-dosing protocol deviation (refer to Section 16.2) would need to be reported as a medication error if it was an "unintended error" as defined in Section 12.1.3.

12.5. Procedure for Reporting Adverse Events of Special Interest (AESI's)

There are no AESIs identified for the study drug, Minocin IV, in this protocol.

12.6. Procedure for Reporting Pregnancies/Lactation Exposure

Occurrences of pregnancy/lactation exposure in a study subject or study subject's partner should be reported within 24 hours using the Pregnancy/Lactation Exposure Reporting form. In cases where a pregnancy/lactation exposure occurs with a Serious Adverse Event, the Serious Adverse Event reporting form should be used to report the SAE and the Pregnancy Reporting form should be used to report the pregnancy. When a pregnancy occurs without any concurrent SAE, the Pregnancy Reporting form may be submitted alone. The pregnancy must be followed through to outcome of pregnancy. Any pregnancy discovered from study drug administration until 30 days after last dose needs to be reported.

12.7. Procedure for Reporting Special Situations

If there is an occurrence of a Special Situation event, defined in 12.1.5, report this occurrence to the Sponsor as per Section 12.3, Procedure for Serious Adverse Event Reporting. Note: The Special Situations event does not need to be serious to be reported on the SAE Report form.

12.8. Procedure for SUSARs

Suspected unexpected serious adverse reactions (SUSAR or EIGI), new Safety Development or the requirement of Urgent Safety Measures will be reported to the Central Committee on Research Involving Human Subjects (CCMO) in accordance with agency timelines.

13. DATA COLLECTION

An electronic data capture (EDC) system will be used for this trial. The IT infrastructure and data management staff will be supplied by CTC Cologne. All users will be trained on the technical features of the EDC by performing an eLearning program prior to gaining access to the EDC. A data entry guideline will cover training of the content of the eCRF. A UserID/Password will be granted for eLearning. After successful completion of the training access to the study eCRF is granted. This UserID and Password is not to be shared amongst the study staff. All users must have a unique account to enter or review data. The eCRF should be filled out by the site continuously during study within 3 business days, basic safety data AEs, safety lab) within 1 business day. All subject data until day 14 within 1 business day from day 14, until day 17 within 3 business days. Discrepancies and implausible values are queried by data management electronically. The trial site should answer these queries without unreasonable delay.

It is not expected that the eCRF will serve as source for any data collected in this trial. If there is a reason for a site to do so, it must be approved by Sponsor and documented in the site files.

Prior to the database being locked, the investigator or designee will review, approve and sign/date each completed eCRF. This signature serves as attestation of the Investigator's responsibility for ensuring that all data entered into the eCRF are complete, accurate and authentic. After the end of the trial, a copy of the data will be provided to the site. This copy will contain the final data, an audit trail of activity on the data, and any queries and answers that were posted for data clarification.

14. STATISTICAL PLAN

Detailed methodology for summary and statistical analyses of the data collected in this trial will be documented in a Statistical Analysis Plan (SAP). The SAP will be prepared by Institute of Medical Statistics, Informatics and Epidemiology (IMSIE) and agreed upon with the Sponsor. This document may modify the plans outlined in the protocol; however, any major modifications of the primary endpoints definition and/or its analysis will also be reflected in a protocol amendment. Additional statistical analyses other than those described in this section may be performed if deemed appropriate and included in the plan.

14.1. Sample size

Ten subjects per cohort are planned, hence n = 60 subjects in total. This sample size was selected based on previous experience attempting to minimize the number of subjects on study drug while obtaining sufficient PK data from different subjects to produce a good estimate of exposure.

14.2. Randomization

Subjects will be allocated to Minocin IV or placebo arm based on permuted blocks of varying length after written informed consent. The randomization will be stratified by sex. A randomization list for each cohort is provided to the pharmacy by the statistician. At the time of inclusion (day 1 of each cohort) the pharmacist assigns treatment according to the randomization list to the subjects indicated by the investigator

Within each cohort, 8 subjects will receive Minocin IV (at least 2 male and 2 female) and 2 placebo (1 male and 1 female). Based on the study center's capabilities, subjects in Cohorts 1 and 2 may be run concurrently where subjects are randomly assigned to receive either 100 mg, 200 mg Minocin IV or placebo.

In case any randomized subject needs to be replaced (see section 8.4), the replacement subject gets the treatment (kit) as already assigned.

14.3. General Statistical Considerations and Definitions

14.3.1. General Statistical Methods

Descriptive statistical analyses will be performed. Number of subjects (n), arithmetic mean, standard deviation (SD), median, first quartile (Q1), third quartile (Q3), minimum (min) and maximum (max) will be given for quantitative variables. Absolute and relative

frequencies are given for qualitative variables. If variables are measured over time, each time point will be summarized. Summaries of the number and frequency and/or raw data listings of AEs will be presented. 95% confidence intervals will be given where reasonable.

No interim analysis is planned.

14.3.2. Analysis Population

The following populations will be used for data analyses and/or presentation.

14.3.2.1. Intent-to-Treat (ITT) Population

All subjects randomized into the trial. Treatment classification will be based on the randomized treatment.

14.3.2.2. Modified Intent-to-Treat (mITT) Population

All ITT subjects who receive at least one dose of study drug.

14.3.2.3. Pharmacokinetics (PK) Population

All subjects who have any valid samples measured for study drug levels. This will be used for PK analysis (on an as-treated basis).

14.3.2.4. Per-Protocol (PP) Population

All mITT subjects who received their assigned study drug without major protocol violations. The PP population will be finalized during a treatment-blind data review before database lock.

14.3.2.5. Safety Population

All subjects who received at least one dose of study drug. Treatment classification will be based on the actual treatment received. This will be the primary population for the safety analyses.

14.3.3. Missing Data Handling

Unless otherwise specified, missing data will not be imputed and will be excluded from the associated analysis.

14.4. Statistical Analyses

14.4.1. Demographic and Background Characteristics

Subject demographics and baseline characteristics will be summarized descriptively by treatment group using ITT, mITT, PK, PP, and safety populations.

14.4.2. Study Drug and Concomitant Medications

Listings and frequency counts of each prior (pre-baseline) medication and concomitant (baseline or later) medication will be provided by treatment. Medications will be coded using the most updated version of the WHO Drug Dictionary available (e.g. version WHODDE DEC. 1, 2015 or later).

14.4.2.1. Adverse Events

The MedDRA dictionary will be used for coding adverse events (AEs). The current version will be used when coding is started.

All AEs are listed by subject with severity, relationship to study drug, cohort (dose), treatment.

The number (percentage) of subjects reporting AEs and the total number (percentage) of AEs will be tabulated by each preferred term, system-organ class, severity, relationship to study drug, cohort (dose), treatment. AE analysis is done for Safety Population.

14.4.3. Laboratory Tests

Laboratory values will be summarized descriptively by treatment group and cohort, including changes and percent changes from baseline at each time point. Analyses will also be performed for each lab parameter by treatment group for incidence rates of potentially clinical significant (PCS) values for subjects without PCS value at baseline.

Numerical values of laboratory parameters from different (if any) local laboratories with different units and normal ranges (if any) will be converted to the conventional units and normalized to a standard set of reference/normal ranges. The normalization process will be performed and separated by each of the laboratory parameters. A shift analysis by normal range will be done which counts the number of patients with a low, normal or high value at baseline and a low, normal or high value post baseline.

14.4.4. Vital Signs

Change and percent change from baseline in vital signs will be summarized descriptively at each scheduled time point by treatment group.

14.4.5. ECGs

PR, QRS, QT, and QTc intervals will be summarized descriptively. A normal-abnormal shift table will be presented for ECGs. ECG results will be classified using frequency counts for normal, abnormality that is not clinically significant (NCS), and clinically significant abnormality (CS) by dose cohort and time point of collection.

14.4.6. Pharmacokinetic Parameters

The plasma concentration-time data for Minocin IV will be analyzed by non-compartmental methods. Treatment arms (doses, placebo) are compared regarding plasma AUC0-t, AUC0-inf, Cmax, and Tmax. Individual as well as mean time-concentration profiles will be graphed. Statistical analysis of dose proportionality of exposure parameters will be performed.

Urine PK parameters such as amount excreted and % dose excreted will be calculated from urinary excretion data.

Detailed methods used and the results obtained will be included in a separate PK report.

15. RECORDS RETENTION

Current EU Directives / Regulations and ICH guidelines collectively require that essential clinical trial documents (including case report forms) other than patient's medical files must be retained for the following time period:

- for at least 15 years after completion or discontinuation of the trial,
- or for at least two years after the granting of the last marketing authorization in the European Community and when there are no pending or contemplated marketing applications in the European Community,
- or for at least two years after formal discontinuation of clinical development of the study drug.

Subject's medical files should be retained in accordance with applicable legislation and in accordance with the maximum period of time permitted by the hospital, institution or private practice. The documents can be retained for a longer period, however, if required by the applicable regulatory requirements or by agreement with the sponsor.

To comply with these requirements, the Investigator will not dispose of any records relevant to this study without either (1) written permission from the Sponsor or (2) providing an opportunity for the Sponsor to collect such records. The Investigator shall take responsibility for maintaining adequate and accurate hard copy source documents of all observations and data generated during this study, including the hard copy or discs received from the sponsor of the final data. Such documentation is subject to inspection by the Sponsor or its agents, the Competent Authority, FDA and/or other regulatory agencies.

16. QUALITY CONTROL AND QUALITY ASSURANCE

The Sponsor has ethical, legal and scientific obligations to carefully follow this study in accordance with established research principles and applicable regulations. The Investigator, as part of his/her responsibilities, is expected to cooperate with the Sponsor (and designees) in ensuring that the study adheres to the protocol and ICH GCP requirements.

As part of a concerted effort to fulfill these obligations, the Sponsor's (or Sponsor designees') monitor will visit the center(s) during the study in accordance with the Monitoring Plan set forth for this trial. The Investigator will permit the Sponsor (and designees) to monitor the study as frequently as is deemed necessary and provide access to medical records/source documents to ensure that data are being recorded adequately, that data are verifiable and that protocol adherence is satisfactory.

16.1. Auditing

The Sponsor (and designees) may conduct audits at the study center(s). Audits will include, but not be limited to, study drug supply, presence of required documents, the informed consent process, and comparison of eCRFs with source documents. The Investigator agrees to permit audits conducted at a reasonable time in a reasonable manner.

Regulatory authorities worldwide may also inspect the Investigator during or after the study. The Investigator should contact the Sponsor (and designees) immediately if this occurs, and must permit regulatory authority inspections.

16.2. Protocol Deviations

This study will be conducted as described in this protocol, except for an emergency situation in which the protection, safety, and well-being of the subject requires immediate intervention, based on the judgment of the Investigator (or a responsible, appropriately trained professional designated by the Investigator). In the event of a significant deviation from the protocol due to an emergency, accident, or mistake, the Investigator or designee must contact the Sponsor (and designees), or their agent, at the earliest possible time by telephone. This will allow an early joint decision regarding the subject's continuation in the study. The Investigator and the Sponsor will document this decision. The EC will be informed of all protocol changes by the Investigator in accordance with the EC established procedure. No deviations from the protocol of any type will be made without complying with all the EC established procedures.

The following Protocol Deviations will require additional information in the eCRF explaining why the deviation occurred and what will be done to prevent it from re-occurring:

- Inclusion criteria violation
- Exclusion criteria violation
- Dosing errors
- Unintentional unblinding

17. ETHICS AND RESPONSIBILITY

This study will be conducted in compliance with the protocol, the Sponsor's standard operating procedures and/or guidelines, the United States Food and Drug Administration (FDA) regulations, the International Conference on Harmonization (ICH) GCP guidelines, the Declaration of Helsinki and other local regulations, as applicable.

17.1. Informed Consent

Written informed consent will be obtained from all subjects before any study-related procedures (including any pre-treatment procedures) are performed. The Investigator(s) has both ethical and legal responsibility to ensure that each subject (and their guardian or legally authorized representative) being considered for inclusion in this study is given a full explanation of the protocol. This shall be documented on a written informed consent form, which shall be approved by the same EC responsible for approval of this protocol. Each informed consent form shall include the elements required by ICH, Part E6, Section 4.8 and any applicable local regulations. The Investigator agrees to obtain approval from the Sponsor of any written informed consent form used in the study, preferably prior to submission to the EC.

Once the appropriate essential information has been provided to the subject and fully explained by the Investigators (or a qualified designee) and it is felt that the subject understands the implications of participating, the subject and the Investigator (or designee) shall sign the EC-approved written informed consent form. The subject shall be given a copy of the signed informed consent form, and the original shall be filed appropriately, according to the institution. A second copy may be filed in the subject's medical record, if allowed by the institution.

17.2. Institutional Review Board/Ethics Committee

This protocol, the written informed consent form and any materials presented to subjects shall be submitted to the EC identified with this responsibility. Notification in writing of approval must come from the EC chairman or secretary, to the Investigator, either as a letter or as a copy of the appropriate section of the EC meeting minutes where this protocol and associated informed consent form were discussed. The Investigator will not participate in the decision. If the Investigator is an EC member, the written approval must indicate such non-participation in the voting session. The Investigator will submit status reports to the EC as required by the

governing body. The EC must be notified by the Investigator in writing of the interruption and/or completion of the study; the Investigator must promptly report to the EC all changes in research (protocol amendments) and will not make such changes without EC approval, except where necessary to eliminate apparent immediate hazards to human subjects. In cases where it is necessary to eliminate immediate hazards to subjects, the EC must then be notified of the change as per local requirements. The Investigator is required to maintain an accurate and complete record of all written correspondence to and received from the EC and must agree to share all such documents and reports with the Sponsor.

18. CONFIDENTIALITY

All information generated in this study must be considered highly confidential and must not be disclosed to any persons not directly concerned with the study without written prior permission from the Sponsor. However, authorized regulatory officials and Sponsor personnel will be allowed full access to the records. All medications provided and subject bodily fluids and/or other materials collected shall be used solely in accordance with this protocol, unless otherwise agreed to in writing by the Sponsor.

Only unique subject numbers in eCRFs will identify subjects. Their full names may, however, be made known to a product regulatory agency or other authorized official if necessary.

With respect to the clinical trial data that is received from countries in the European Economic Area and Switzerland, MDCO has certified adherence to the US-EU and the US-Swiss Safe Harbor Principles.

19. INVESTIGATOR AGREEMENT

I have read and understand the protocol (including the Investigator's Brochure) and agree that it contains all the ethical, legal and scientific information necessary to conduct this study. I will personally conduct the study as described.

I will provide copies of the protocol to all physicians, nurses and other professional personnel responsible to me who will participate in the study. I will discuss the protocol with them to assure myself that they are sufficiently informed regarding Minocin IV, safety parameters and the conduct of the study in general. I am aware that this protocol must be approved by the Ethics Committee (EC) responsible for such matters in the Clinical Study Facility where Minocin IV will be tested prior to commencement of this study. I agree to adhere strictly to the attached protocol. I understand that this EC approved protocol will be submitted to relevant regulatory authorities by the Sponsor, as appropriate. I agree that clinical data entered on case report forms by me and my staff will be utilized by the Sponsor in various ways such as for submission to governmental regulatory authorities and/or in combination with clinical data gathered from other research sites, whenever applicable. I agree to allow Sponsor monitors and auditors full access to all medical records/source documents at the research facility for subjects screened or randomized in the study.

I agree to provide all subjects with informed consent forms, as required by government and ICH regulations. I further agree to report to the Sponsor any adverse experiences in accordance with the terms of this protocol, ICH guideline, Part E6, Section 4.11 and applicable local regulations.

Principal Investigator (Signature)	Date
Principal Investigator (Printed Name)	Protocol Version: Original Version
Institution Name	

20. REFERENCES

Antibiotic Resistance Threats in the United States. Centers for Disease Control and Prevention (CDC). 2013.

Casha, S. et al. Results of a phase II placebo-controlled randomized trial of minocycline in acute spinal cord injury. Brain 135, 1224-1236 (2012).

Fagan, S. C. et al. Minocycline to improve neurologic outcome in stroke (MINOS): a dose-finding study. Stroke 41, 2283-2287 (2010).

Garrido-Mesa, N, A Zarzuelo and J Gálvez. Minocycline: far beyond an antibiotic. British Journal of Pharmacology 2013; 169: 337-352.

Generating Antibiotic Incentives Now (GAIN) Act, United States, 2012.

Memo/08/788, European Commission Press Release Database, 15 Dec 2008.

MINOCIN® (minocycline) for Injection 100 mg/vial [US Prescribing Information]. The Medicines Company; Parsippany, NJ. April 2015. Available at: http://www.accessdata.fda.gov/drugsatfda_docs/label/2015/050444s049lbl.pdf. Accessed 1 February 2016.

Minocin IV Investigator's Brochure, Edition 2 Addendum, October 2017.

MINOCIN® MR 100 mg Modified Release Capsules [Summary of Product Characteristics]. Meda Pharmaceuticals; Bishop's Stortford, UK. March 2014. Available at: http://www.medicines.org.uk/emc/medicine/21139. Accessed 1 February 2016.

Minocycline Tablets 100 mg [Summary of Product Characteristics]. Actavis UK Ltd; Devon UK. August 2011. Available at: http://www.medicines.org.uk/emc/medicine/24177. Accessed 1 February 2016.

Munoz-Price LS, Weinstein RA. Acinetobacter Infection. New Engl J Med. 2008;358:12.

Shlaes DM, Sahm D, Opiela C et al. The FDA reboot of antibiotic development. Antimicrobial Agents and Chemotherapy. 2013;57:4605-07.