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LYS228

Clinical Trial Protocol CLYS228X2202

A randomized, controlled, evaluator-blinded, multi-center study to evaluate LYS228 pharmacokinetics, clinical response, safety, and tolerability in patients with complicated intra-abdominal infection

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Site Operations Manual (SOM)

A Site Operations Manual (SOM) accompanies this protocol, providing the operational details for study conduct. Note: The SOM will not form part of the Clinical Study Report.

Notification of serious adverse events

Dear Investigator,

You must report a serious adverse event (SAE) (initial or follow-up) to Novartis as summarized below. Refer to Section 9.2 of the protocol for SAE criteria and additional requirements. See also page 2 of the SOM for further details on the method of reporting a SAE.

- Complete SAE report
- Submit SAE report to Novartis Chief Medical Office & Patient Safety (CMO&PS) **within 24 hours after awareness of the SAE**
- Notify the Novartis Medical Lead
- The fax number(s) and email address(es) are located in the SOM.

Table of contents

Corporate Confidential Information

Corporate Confidential Information

Corporate Confidential Information

List of tables

List of figures

List of abbreviations

AE	adverse event
ALP	alkaline phosphatase
ALT	alanine aminotransferase
APACHE II	Acute Physiology and Chronic Health Evaluation II
AST	aspartate aminotransferase
BMI	Body Mass Index
BUN	blood urea nitrogen
CDC	Centers for Disease Control and Prevention
CE	clinically evaluable
CFR	U.S. Code of Federal Regulations
CFU	colony forming units
cIAI	complicated intra-abdominal infection
CK	creatinine kinase
cm	centimeter(s)
CMO&PS	Chief Medical Office & Patient Safety
CrCl	creatinine clearance
CRF	Case Report/Record Form (paper or electronic)
CRO	Contract Research Organization
CTCAE	Common Terminology Criteria for Adverse Events
CV	coefficient of variation
CVVH	continuous veno-venous hemofiltration
dL	deciliter(s)
ECG	electrocardiogram
eCrCl	estimated creatinine clearance
EDC	Electronic Data Capture
EEG	electroencephalogram
EOAT	End of Antibiotic Treatment/Therapy
EOS	End of Study
EOSD	End of Study Drug
ESBLs	extended-spectrum β-lactamases
FDA	Food and Drug Administration

GABA	gamma-aminobutyric acid
GCP	Good Clinical Practice
GGT	Gamma-glutamyl transferase
h	hour
HBsAg	hepatitis B surface antigen
HBV	hepatitis B virus
hCG	human chorionic gonadotropin
HCV	hepatitis C virus
HIV	human immunodeficiency virus
i.v.	intravenous
ICF	Informed Consent Form
ICH	International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use
IEC	Independent Ethics Committee
INR	International Normalized Ratio
IRB	Institutional Review Board
IRT	Interactive Response Technology
KPC	*Klebsiella pneumoniae* carbapenemases
kg	kilogram
LDH	lactate dehydrogenase
LFT	liver function test
LLN	lower limit of normal
LLOQ	lower limit of quantification
MBL	metallo-β-lactamase
ME	microbiologically evaluable
MedDRA	Medical dictionary for regulatory activities
mg	milligram(s)
µg	microgram(s)
µL	microliters(s)
MIC	minimal inhibitory concentration
micro-	microbiologic
mITT	modified intention-to-treat

mL	milliliter(s)
MRSA	methicillin-resistant *Staphylococcus aureus*
MTD	maximum tolerated dose
NCDS	Novartis Clinical Data Standards
NDM-1	New Delhi metal-β-lactamase-1
ng	Nanogram(s)
OAT	organic anion transporter
p.o.	oral
PBP	penicillin binding protein
PD	pharmacodynamic(s)
PK	pharmacokinetic(s)
PM	Pharmacy Manual
PTA	probability of target attainment
qSOFA	quick sequential sepsis-related organ failure assessment
RBC	red blood cell(s)
RDC	Remote Data Capture
SAE	serious adverse event
SBL	serine-β-lactamase
sCR	serum creatinine
SD	standard deviation
SOM	Site Operations Manual
SUSAR	Suspected Unexpected Serious Adverse Reactions
TBL	total bilirubin
TD	Study Treatment Discontinuation
TOC	Test of Cure
ULN	upper limit of normal
ULQ	upper limit of quantification
WBC	white blood cell(s)
WHO	World Health Organization
WoC	Withdrawal of Consent

Pharmacokinetic definitions and symbols

%fT>MIC	Percentage of time that free drug concentration is above the MIC in plasma
AUCtau	The area under the plasma (or serum or blood) concentration-time curve from time zero to the end of the dosing interval tau [mass x time / volume]
AUCtau,ss	The area under the plasma (or serum or blood) concentration-time curve from time zero to the end of the dosing interval tau at steady state [mass x time / volume]
CL	The systemic (or total body) clearance from plasma (or serum or blood) following intravenous administration [volume / time]
Cmax	The observed maximum plasma (or serum or blood) concentration following drug administration [mass / volume]
Cmax,ss	The observed maximum plasma (or serum or blood) concentration following drug administration at steady state [mass / volume]
MIC_{90}	The minimal inhibitory concentration (MIC) to inhibit the growth of 90% of organisms tested
PTA	Probability of target attainment
T1/2	The terminal elimination half-life [time]
Tmax	The time to reach the maximum concentration after drug administration [time]
Vss	The volume of distribution at steady state following intravenous administration [volume]

Glossary of terms

Assessment	A procedure used to generate data required by the study
Cohort	A specific group of patients fulfilling certain criteria
Control drug	Any drug(s) (an active drug or an inactive drug, such as a placebo) which is used as a comparator to the investigational drug being tested in the trial
Dosage	Dose of the study treatment given to the patient in a time unit (e.g. 100 mg once a day, 75 mg twice a day)
Electronic Data Capture (EDC)	Electronic data capture (EDC) is the electronic acquisition of clinical study data using data collection systems, such as Web-based applications, interactive voice response systems and clinical laboratory interfaces. EDC includes the use of Electronic Case Report Forms (eCRFs) which are used to capture data transcribed from paper source forms used at the point of care.
Enrollment	Point/time of patient entry into the study at which informed consent must be obtained (i.e. prior to starting any of the procedures described in the protocol)
Epoch	Interval of time in the planned conduct of a study. An epoch is associated with a purpose (e.g. screening, randomization, treatment, follow-up) which applies across all arms of a study.
Investigational drug	The Study Drug whose properties are being tested in the study; this definition is consistent with US CFR 21 Section 312.3 and Directive 2001/20/EC and is synonymous with “investigational new drug,” "Investigational Medicinal Product," or “test substance”
Medication pack number	A unique identifier on the label of each drug package in studies that dispense study treatment using an IRT system
Patient	An individual with the condition of interest
Patient Number	A unique number assigned to each patient upon signing the informed consent. This number is the definitive, unique identifier for the patient and should be used to identify the patient throughout the study for all data collected, sample labels, etc.
Personal data	Patient information collected by the Investigator that is transferred to Novartis for the purpose of the clinical trial. This data includes patient identifier information, study information and biological samples.
Randomization number	A unique identifier assigned to each randomized patient, corresponding to a specific treatment assignment
Screen Failure	A patient who is screened but is not treated or randomized

Source Data/Document	Source data refers to the initial record, document, or primary location from where data comes. The data source can be a database, a dataset, a spreadsheet or even hard-coded data, such as paper or eSource
Study Drug	Intravenous LYS228 in combination with intravenous vancomycin and intravenous or oral metronidazole, or standard of care intravenous antibiotic therapy for the treatment of cIAI, administered between randomization and End of Study Drug Visit.
Study treatment	See definition for Study Drug
Study treatment discontinuation	When the patient permanently stops taking study treatment prior to the defined study treatment completion date
Treatment number	A unique identifier assigned in non-randomized studies to each dosed patient, corresponding to a specific treatment
Variable	A measured value or assessed response that is determined in specific assessments and used in data analysis to evaluate the drug being tested in the study
Withdrawal of consent (WoC)	Withdrawal of consent from the study is defined as when a subject does not want to participate in the study any longer, and does not allow any further collection of personal data.

Protocol summary

Protocol number	CLYS228X2202
Full Title	A randomized, controlled, evaluator-blinded, multi-center study to evaluate LYS228 pharmacokinetics, clinical response, safety, and tolerability in patients with complicated intra-abdominal infection
Brief title	LYS228 pharmacokinetics, clinical response, safety and tolerability in patients with complicated intra-abdominal infection (cIAI)
Sponsor and Clinical Trial Phase	Novartis Phase II
Intervention type	Drug
Study type	Interventional
Purpose and rationale	The purpose of this non-confirmatory study is to evaluate the probability of LYS228 pharmacokinetic-pharmacodynamic (PK/PD) target attainment and clinical response in patients with cIAI in order to refine human efficacious dose selection for future confirmatory studies
Primary Objectives	• To evaluate the plasma pharmacokinetics of LYS228 in patients with cIAI • To evaluate the clinical response to LYS228 in combination with vancomycin and metronidazole compared to standard of care antibiotics for treating patients with cIAI
Secondary Objectives	• To evaluate the safety and tolerability of LYS228 in combination with vancomycin and metronidazole in patients with cIAI • To evaluate the microbiological response to LYS228 in combination with vancomycin and metronidazole compared to standard of care antibiotics for treating patients with cIAI
Study design	This is a randomized, comparator-controlled, evaluator-blinded, non-confirmatory study to evaluate the pharmacokinetics, clinical response, safety and tolerability of intravenous LYS228 in patients with cIAI. The study will enroll approximately 60 patients with cIAI who have not yet been treated for their current infection. Patients will be randomized to receive LYS228 in combination with vancomycin and metronidazole or standard of care antibiotic therapy for cIAI for no less than 5 days and up to 14 days. The study treatment will be administered on an open-label basis. A blinded evaluator will perform safety and clinical response assessments. The study consists of a Screening and Baseline Period (D-1 to D1), a Treatment Period of variable duration (D1 to D5, and possibly up to D15) and a Follow-up period of at least 14 days and an End of Study (EOS) visit on Day 28. Patients who meet the eligibility criteria at screening will be randomized to one of the 2 treatment arms: (1) LYS228 in combination with vancomycin and metronidazole, or (2) standard of care antibiotic therapy for the treatment of cIAI. Blood cultures will be obtained before administration of Study Drug and preferably before administration of any antibiotic therapy. In addition, cultures from the intra-abdominal focus of infection will be obtained during surgery, or at the time of percutaneous drainage. The Screening/Baseline (D-1 to 1) and first dosing visits (D1) may occur on the same day. Study Drug will be administered for no less than 5 days in an in-patient setting. Please refer to the full protocol for definitions of End of Study Drug visit (EOSD) and End of Antibiotic Treatment visit (EOAT). The EOS visit will be completed on Day 28 after randomization.

Population	Approximately 60 patients will be randomized to LYS228 or a comparator (standard of care therapy) in 2:1 ratio and are expected to be included in the microbiologic modified intention to treat (micro-mITT) population.

Key Inclusion criteria	• Male and female patients 18 to 65 years of age inclusive and after an interim analysis of 8 patients, inclusion of patients up to 85 years of age with visual confirmation (presence of pus within the abdominal cavity) of an intra-abdominal infection associated with peritonitis including at least 1 of the following inclusionary diagnoses during surgical intervention (e.g. laparotomy, laparoscopic surgery, or percutaneous draining of an abscess):
	  • Cholecystitis (including gangrenous cholecystitis) with rupture, perforation, or progression of the infection beyond the gallbladder wall;
	  • Diverticular disease with perforation or abscess;
	  • Appendiceal perforation or periappendiceal abscess;
	  • Acute gastric or duodenal perforation, only if operated on >24 hours after perforation occurs;
	  • Traumatic perforation of the intestine, only if operated on >12 hours after perforation occurs;
	  • Secondary peritonitis due to perforated viscus, postoperative or spread from other focus of infection (but not spontaneous [primary] bacterial peritonitis or peritonitis associated with cirrhosis and chronic ascites);
	  • Intra-abdominal abscess (including of liver or spleen provided that there was extension beyond the organ with evidence of intraperitoneal involvement)
	• Provided all other inclusion criteria are met, patients can be enrolled before surgical intervention in certain conditions, as described in the full protocol.

Key Exclusion criteria	• Any of the excluded diagnoses: abdominal wall abscess, small bowel obstruction, traumatic bowel perforation undergoing surgery within 12 hours, perforation of gastroduodenal ulcer with surgery within 24 hours, an intra-abdominal process that is not likely caused by infection. Please refer to the full protocol for the full list. • Pre-operative treatment of any duration with non-Study Drug systemic antibiotic therapy for peritonitis or abscess is not allowed unless certain criteria are met. Please refer to the full protocol for the required criteria. • Concomitant bacterial infection at time of enrollment requiring non-Study Drug antibiotics and that may interfere with the evaluation of clinical response to the study antibiotic. • Known non-abdominal source of infection, including endocarditis, osteomyelitis, abscess, meningitis, or pneumonia diagnosed within 7 days prior to enrollment. • Patient has an Acute Physiology and Chronic Health Evaluation II (APACHE II) score > 30 or is considered, in the judgement of the investigator, unlikely to survive 4 weeks (e.g. rapidly progressive terminal illness, including septic shock). • Patients that meet sepsis criteria as defined by the quick sequential sepsis-related organ failure assessment (qSOFA) • Women of child-bearing potential unless they are using highly effective methods of contraception during dosing and for 7 days after stopping study treatment.

	• Significant lab abnormalities, as described in the full protocol.
Study treatment	Patients will be randomized to one of the following two treatment arms in a ratio of 2:1. • Arm 1: LYS228 in addition to vancomycin and metronidazole (n=40). LYS228 dose for each patient will be based on the renal function as described in the full protocol. • Arm 2: Standard of care intravenous therapy for the treatment of cIAI (n=20)
Pharmacokinetic (PK) assessments	Cmax, Tmax, AUCTau, T1/2, %fT>MIC, CL and Vss will be assessed from the plasma concentration-time data
Efficacy/PD assessments	• Clinical evaluation of cIAI infection • Microbiologic evaluation
Key safety assessments	• Adverse event monitoring • Physical examination and vital signs • Monitoring of laboratory markers in blood and urine • Electrocardiogram (ECG) • Assessment of fertility and pregnancy
Other assessments	• Blood cultures Corporate Confidential Information
Data analysis	Noncompartmental PK analysis will be performed on LYS228 plasma concentration-time profiles. Descriptive summary statistics of LYS228 concentrations and PK parameters will be provided by visit/sampling time-point. Clinical Response The proportion of patients who achieved clinical success or failure will be computed for each treatment arm and provided along with a 80% two-sided confidence interval based on the Clopper-Pearson method. The 80% confidence interval for the difference in clinical success between treatment arms will be computed according to the Wilson score method without continuity correction. Clinical success or failure will be evaluated at the EOSD, at the EOAT, and at 28 days post-randomization (EOS). Bayesian methods will be implemented based on the Beta-Binomial model to compare the clinical success rate in micro-mITT population at 28 days post-randomization. Informative prior on LYS228 and control will be used. An 80% posterior credible interval of the clinical success rate at Test of Cure (TOC) visit in the micro-mITT population will be provided for each treatment group as well as the treatment difference.
Key words	Intra-abdominal infection, LYS228, beta-lactam antibiotics, creatinine clearance, *Enterobactericeae*

1 Introduction

1.1 Background

Intra-abdominal infections (IAIs) are common in clinical practice and comprise a variety of pathological conditions, ranging from uncomplicated appendicitis to fecal peritonitis. Intra-abdominal infections are considered complicated (cIAI) when they are associated with perforation or necrosis of the gastrointestinal tract, and extension beyond the hollow viscus of origin into the peritoneal space. The release of enteric bacteria into the peritoneal and retroperitoneal space is associated with systemic signs and symptoms of infection, and either abscess formation or peritonitis (Solomkin et al 2010). Enteric flora includes Gram-negative *Enterobacteriaceae*, as well as Gram-positive and anaerobic bacteria.

Patients with infections caused by multi drug resistant *Enterobacteriaceae* are currently treated with carbapenems (Hooten et al 2010, Golan 2015). Carbapenems are characterized by a broad spectrum of antibacterial activity, however many bacterial strains have acquired the ability to express carbapenemases of the serine- β-lactamase (SBL) and/or metallo-β-lactamase (MBL) classes (i.e. *Klebsiella pneumonia* carbapenemase and New Delhi metallo-β-lactamase-1), rendering carbapenems ineffective. Infections caused by these strains often require the administration of poorly-tolerated antibiotics such as colistin (Pogue et al 2011, Navarro-San Francisco et al 2013).

The US CDC classifies carbapenem-resistant *Enterobacteriaceae* as an "urgent threat" and Enterobacteriaceae that express extended-spectrum β-lactamases (ESBLs) as a "serious threat" (CDC 2013). Furthermore, the World Health Organization has issued a global priority list of antibiotic-resistant bacteria for drug development of new antibiotics. *Enterobacteriaceae*, which are a common cause of hospital and community-acquired infections, have been assigned a critical priority status (WHO 2017).

LYS228 is a novel monobactam antibiotic which kills bacteria by inhibiting cell wall synthesis through covalent modification of the active site serine of penicillin binding protein 3 (PBP3). Monobactams represent a class of beta-lactam antibiotics which, unlike carbapenems, are intrinsically-stable to MBLs. PBP3 is a clinically validated antibacterial target, as it is targeted by aztreonam, a monobactam widely that is widely used in clinical settings (Tunkel and Scheld 1990). Monobactams are efficacious for treating infections with susceptible strains, are generally well tolerated, are primarily eliminated through urine, and have a predictable pharmacokinetic profile.

LYS228 is stable to most SBLs and all known MBLs and is therefore expected to be effective against bacterial infections caused by clinical strains of *Enterobacteriaceae* expressing β-lactamases which are susceptible to LYS228. MBL-expressing *Enterobacteriaceae* can inactivate all classes of β-lactam drugs, except monocyclic β-lactams like the monobactams. MBLs are frequently co-expressed with SBLs. The prevalence of strains expressing the New Delhi metallo-β-lactamase-1 (NDM-1) is as high as 12% in clinical isolates from patients with invasive infections in India (Biedenbach et al 2015, Rahman et al 2014); infections caused by strains expressing NDM-1 and other MBLs have been detected globally, including in Europe, the United States, China, and Japan (Kazmierczak et al 2016).

Further, the prevalence of MBL-expressing *Enterobacteriaceae* is increasing in environmental isolates and is expected to increase rapidly in the clinic as they are likely to be selected following the recent introduction of novel β-lactam antibiotics such as ceftazidime/avibactam that cover strains expressing SBLs but not MBLs.

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In summary, LYS228 is a monobactam antibiotic potent against *Enterobacteriaceae* and is stable in the presence of many serine- and all known metallo-β-lactamases. It is expected that LYS228 as monotherapy will be effective in treating patients with infections caused by LYS228-susceptible *Enterobacteriaceae*, including those expressing ESBLs and carbapenemases.

This study will evaluate the pharmacokinetics of LYS228, and the clinical response, safety, and tolerability of LYS228 in combination with vancomycin and metronidazole in patients with complicated intra-abdominal infections (cIAI).

The most relevant data for the present study are summarized in the sections below. For detailed information about LYS228, please refer to the Investigator's Brochure.

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1.4 Study purpose

The purpose of this non-confirmatory study is to evaluate the probability of LYS228 pharmacokinetic-pharmacodynamic (PK/PD) target attainment and clinical response in order to refine human efficacious dose selection for future confirmatory studies.

2 Objectives and endpoints

2.1 Primary objectives

Primary objectives	***Endpoints related to primary objectives***
• To evaluate the plasma pharmacokinetics of LYS228 in patients with cIAI	• Plasma PK on Day 5
• To evaluate the clinical response to LYS228 in combination with vancomycin and metronidazole compared to standard of care antibiotics for treating patients with cIAI	• Clinical success at 28 days after randomization (End of Study) determined by signs, symptoms, and the need for additional antibiotics, unplanned surgical procedures, or drainage.

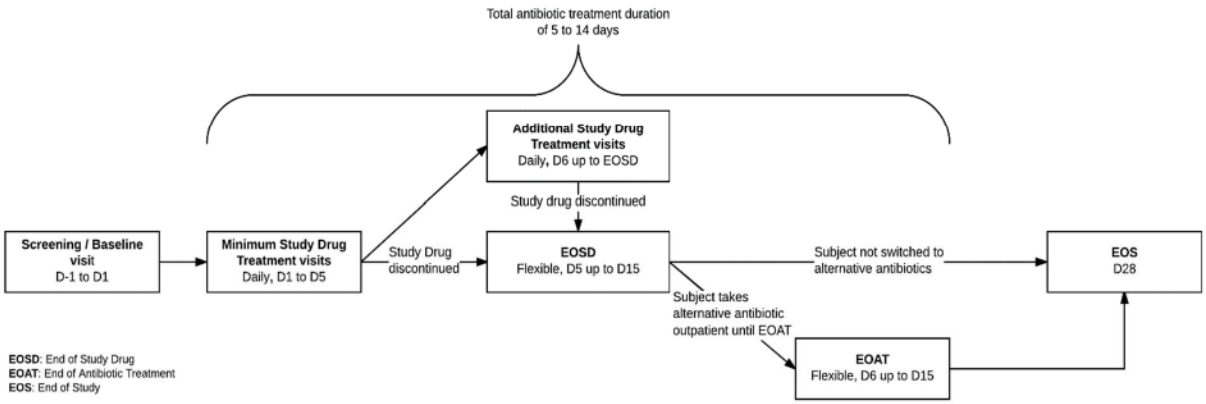
2.2 Secondary objectives

Secondary objectives	***Endpoints related to secondary objectives***
• To evaluate the safety and tolerability of LYS228 in combination with vancomycin and metronidazole in patients with cIAI	• Adverse events • Clinical laboratory tests • Vital signs
• To evaluate the microbiological response to LYS228 in combination with	• Microbiologic success at 28 days after randomization (End of Study) determined

Secondary objectives	*Endpoints related to secondary objectives*
vancomycin and metronidazole compared to standard of care antibiotics for treating patients with cIAI	by culture from the intra-abdominal focus of infection when available or presumed eradication based on clinical success

3 Investigational plan

3.1 Study design



This is a randomized, comparator-controlled, evaluator-blinded, non-confirmatory study to evaluate the pharmacokinetics, clinical response, safety and tolerability of intravenous LYS228 in patients with complicated intra-abdominal infection (cIAI). The study will enroll approximately 60 patients with cIAI who have not yet been treated for their current infection and will be randomized to receive LYS228 in combination with vancomycin and metronidazole or standard of care antibiotic therapy for cIAI for no less than 5 days and up to 14 days. The study treatment will be administered to patients on an open-label basis. There will be no placebo-treated patients. A blinded evaluator at each site will perform safety and clinical response assessments.

The study consists of a Screening and Baseline Period (D-1 to D1), a Treatment Period of variable duration (D1 to D5, and possibly up to D15), a Follow-up period of at least 14 days and an EOS visit on Day 28. A patients' flowchart is provided in Section 17.

Screening and Baseline Period

Patients who meet the eligibility criteria at screening and are admitted to an inpatient unit will be randomized to one of the 2 treatment arms: (1) LYS228 in combination with vancomycin and metronidazole, or (2) standard of care antibiotic therapy for the treatment of cIAI. Blood cultures will be obtained before administration of Study Drug and preferably before administration of any antibiotic therapy. In addition, cultures from the intra-abdominal focus of infection will be obtained during surgery or at the time of percutaneous drainage. The Screening/Baseline (D-1 to 1), and first dosing visits (D1) may occur on the same day. Patients can be screened and enrolled before or after surgery or percutaneous drainage of infection, as detailed in the inclusion criteria.

Treatment Period

The first dose of Study Drug will be administered on D1 after confirmation of eligibility, randomization and collection of blood cultures.

"Study Drug" and "study treatment" include: Intravenous LYS228 in combination with intravenous vancomycin and intravenous or oral metronidazole (Arm 1), or standard of care intravenous antibiotic therapy for the treatment of cIAI (Arm 2), administered between randomization and End of Study Drug. Study Drug will be administered for no less than 5 days and it is expected to be administered in an in-patient setting.

Change of treatment from Study Drug to alternative antibiotics is permissible in the following circumstances:

1. Change to oral antibiotics after completing 5 days of intravenous Study Drug if a patient is clinically improved and discharged, or
2. Patient requires additional therapy or change to rescue therapy in the case of clinical failure, or adverse event that merits discontinuation of Study Drug as detailed in Section 7.2.

Alternative antibiotics must be effective against the relevant clinical isolates. The total antibiotic treatment duration, including Study Drug and any alternative antibiotic, will be no less than 5 days and up to 14 days, unless discussed with sponsor.

During the treatment period all patients will complete a minimum of 5 visits (V101 –V105) and up to 15 visits (V101-V115), depending on the duration of hospitalization. All patients that change treatment to an alternative antibiotic must have an EOSD visit prior to antibiotic change.

Follow-up Period

The follow-up period spans the period after completion of antibiotic treatment (EOAT visit) up to 28 days after randomization, at which time all patients will have an EOS visit. This visit is considered the Test of Cure (TOC) visit for evaluation of clinical response. A post-study safety phone call will be conducted 30 days after the last administration of study treatment.

3.2 Rationale of study design

3.2.1 Study population

The study includes patients with complicated intra-abdominal infections that extend beyond the hollow viscus and local viscera into the peritoneal or retroperitoneal space, leading to either abscess formation or peritonitis, and associated with systemic symptoms of infection. It includes patients with a wide range of estimated creatinine clearances (>30 mL/min) to increase the understanding of LYS228 PK in cIAI patients with normal renal function, as well as mild and moderate renal dysfunction. It excludes patients that received previous effective antibiotic therapy for a continuous duration of more than 24 hours during the 72 hours prior to screening to prevent confounding of the clinical response endpoints. See Section 4.1 and Section 4.2 for the full inclusion and exclusion criteria.

3.2.2 Design

All patients with cIAI require treatment with antibiotics to prevent additional morbidity and mortality. Standard of care treatment for cIAI includes antibiotics effective against enteric Gram-negative, Gram-positive, and anaerobic bacteria. LYS228 is expected to be effective only against enteric Gram-negative bacteria, therefore it will be administered in combination with vancomycin and metronidazole to provide adequate treatment of Gram-positive and anaerobic bacteria. The combination will be compared to standard of care broad spectrum intravenous therapy for treatment of cIAI. Best available oral therapy is permitted only for documented infections caused by multiple-drug resistant bacteria where intravenous antibiotics are not effective.

The use of a control arm and evaluator-blinded design will mitigate potential bias in safety and clinical response assessments. For clinical response, bias may be introduced by the patients for some of the more subjective endpoints (symptoms), but many of the endpoints are much less likely to be subject to bias. These include fever and white blood cell counts. Patient blinding is not considered feasible or desirable given the need to adopt a double dummy design resulting in long infusion time and large volumes.

Expected clinical response rates can be derived from controlled studies of similar antibiotics to treat patients with cIAI and from regulatory guidance. With these data, the clinical response of LYS228 can be assessed using historical data as controls. Use of historical controls supports the 2:1 randomization ratio, which maximizes the number of patients to be enrolled in the LYS228 arm to assess clinical response.

3.2.3 Clinical response endpoints

The primary endpoint of clinical success, defined by improvement of clinical signs and symptoms of infection, has been validated in controlled studies of similar antibiotics to treat patients with cIAI. Bacterial cultures to document microbiologic eradication are not likely to be available for this infection, therefore presumed microbiologic success is derived from clinical success. The study uses clinical response endpoints that are consistent with the ones used by the historical controls to minimize confounders in the evaluation of clinical response.

3.3 Rationale for dose/regimen, route of administration and duration of treatment

The probability of microbiologic target attainment to achieve efficacy can be derived from existing knowledge of PK drivers of efficacy from other β-lactam antibiotics with similar mechanisms of action, Corporate Confidential Information

Initial LYS228 dose will be selected based on renal function as determined by estimated creatinine clearance (eCrCL) calculated at screening based on data obtained from local laboratories and the study qualified formula (Cockcroft Gault equation) as described in the SOM and Pharmacy Manual (PM). During the treatment course, dose must be adjusted based on the eCrCL, as indicated based on changes in serum creatinine. See Table 3-1 for the dosing of LYS228.

3.3.1 Efficacious dose

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3.3.2 Treatment duration

The current standard of care for the treatment of cIAI is 5 to 14 days of antibiotic therapy. The optimal duration of antimicrobial therapy for the treatment of acute symptomatic episodes has not been systematically studied. Furthermore, because of the wide variation in underlying abnormalities and clinical presentations, a uniform recommendation for treatment duration cannot be made. However, shorter courses of therapy appear to be sufficient, particularly among patients who demonstrate a positive response to treatment, show no signs of persistent leucocytosis or fever, and are able to resume oral diet. Patients who meet these criteria usually do not require more than 5 days of treatment.

All patients will receive intravenous LYS228 or standard of care therapy in an in-patient setting for a minimum of 5 days and up to 14 days. Extension of the treatment duration might be allowed with non-LYS228 antibiotics, and must be discussed with sponsor on a case by case basis.

A minimum of 5 days of therapy will allow analysis of safety, clinical response and LYS228 PK, at a time-point where steady state and clinical success is expected in this population. Continuation of therapy up to 14 days allows patients with more refractory infections to receive a longer course of antibiotics and have a higher probability of clinical and microbiological success.

Practice guidelines, patient convenience, and risks associated with an indwelling intravenous catheter make the administration of an intravenous antibiotic for the up to 14 day treatment duration difficult and impractical. Allowing a change of treatment from Study Drug to alternative oral antibiotics, if a patient is clinically improved, will facilitate outpatient administration to complete the required course of therapy. Oral therapy in many cases mimics the standard of care.

3.3.3 Dose or regimen adjustment

The sponsor may recommend an additional dose regimen for LYS228 based on the availability of new data. Longer infusion times result in increased fT%>MIC. Therefore, continuous infusion of LYS228 may be considered based on ongoing analysis of exposure, stability and sterility data, and/or analysis of the ongoing clinical studies of LYS228.

The investigator is expected to adjust the LYS228 dose, as required, based on individual patients' renal function during the treatment period or emerging new PK data. Please refer to the SOM for further details.

3.4 Rationale for choice of comparator

Using pre-specified local standard of care antibiotic therapy for cIAI, provides an adequate control with antibiotics that are used in clinical practice for the indication. The study implements a preferred standard of care regimen, piperacillin/tazobactam, in order to limit confounding factors when comparing treatment groups. Considering that LYS228 activity is limited to *Enterobacteriaceae*, and piperacillin/tazobactam has broad spectrum activity, the study also implements preferred antibiotics to be used in combination with LYS228 to provide adequate treatment of gram-positive and anaerobic bacteria, which are relevant pathogens in cIAI. Modifications to the preferred comparatorare allowed in order to optimize empiric antibiotic therapy based on host risk factors for bacterial drug resistance, local antibiograms, documented drug resistance, tolerability, or other relevant limitations to the preferred comparator. (See Section 6.1.1) Modification to the preferred antibiotic to be used for treatment of gram-positive bacteria (e.g vancomycin) is also allowed in instances of documented bacterial drug resistance, tolerability, or other relevant limitations.

3.5 Purpose and timing of interim analyses/design adaptations

LYS228 exposure across patients with varying renal function will be evaluated during the study to confirm that LYS228 concentrations are predicted to be adequate to treat the patient population. The PK exposure of the initial 8 patients will be analyzed. A second PK analysis with 20 patients will be conducted. This will enable an emerging data review of the required doses for patients in each renal function category and adjustment as necessary (see Section 11.8 for additional details).

3.6 Risks and benefits

Patients participating in the study. may benefit from receiving effective antibiotic therapy to treat cIAI.

The risk to patients in this trial will be minimized by adherence to the eligibility criteria, close clinical monitoring, and following study stopping rules. Corporate Confidential Information

3.6.1 Hypersensitivity reaction

Other β-lactam antibiotics have been associated with the development of hypersensitivity (Romano et al 2016). Rare cases of toxic epidermal necrolysis have been reported following administration of aztreonam and other β-lactam antibiotics (LeFrock et al 1987). These have usually been observed in patients undergoing bone marrow transplantation.

Patients will be closely monitored for evidence of rash or other forms of hypersensitivity. Patients will be excluded from the study if they have a history of β-lactam allergy or hypersensitivity.

3.6.2 Hepatotoxicity

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Markers of liver toxicity, including transaminases, will be monitored in the study.

3.6.3 Nephrotoxicity

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Nephrotoxicity is a known adverse effect of many β-lactam antibiotics (Lagacé-Wiens and Rubinstein 2012). Markers of renal toxicity, including serum creatinine, will be monitored and LYS228 dose will be adjusted based on the estimated creatinine clearance.

3.6.4 Local Reaction

Local reactions such as phlebitis or thrombophlebitis following intravenous administration of aztreonam occur at rates of 1.9% to 2.4% but may be higher depending on how they are assessed (Henry and Bendush 1985, Ray-Barruel et al 2014). In hospitalized patients, catheter-related phlebitis rates can be as high as 42% of catheters and the relative risk of phlebitis is 2.5 times higher with antibiotics (Maki and Ringer 1991). LYS228 administration may result in similar local reactions. To reduce the risk of such reactions, catheter placement and management

should follow local standard of care. Such local reactions will be monitored as detailed in the SOM.

3.6.5 Gastrointestinal discomfort

Administration of β-lactam antibiotics is associated with the development of abdominal pain or discomfort, diarrhea, nausea, and vomiting (Lagace-Wiens and Rubinstein 2012). These symptoms generally occur in less than 2% of patients receiving aztreonam (Newman et al 1985). Patients will be monitored for gastrointestinal symptoms.

3.6.6 *Clostridium difficile*–associated diarrhea

C. difficile-associated diarrhea has been reported following treatment with nearly all antibiotics, including aztreonam (Lagace-Wiens and Rubinstein 2012). Such diarrhea may range in severity from mild abdominal discomfort to severe colitis that may result in death. Patients suspected of having symptoms due to *C. difficile* should be tested for the presence of *C. difficile* antigen followed by a confirmatory test for the *C. difficile* toxin in the stool. Patients testing positive should be treated with appropriate standard of care.

3.6.7 Seizure

Seizures are associated with β-lactam antibiotics (Sutter et al 2015).

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Patients will be monitored for seizure activity.

3.6.8 Thrombocytopenia

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Cytopenias, including thrombocytopenia, have been observed in patients receiving β-lactam antibiotics (Lagace Wiens and Rubinstein 2012). Aztreonam administration demonstrated no adverse effects on platelet function in healthy subjects (Tartaglione et al 1986, Agnelli et al 1987).

Women of child bearing potential and sexually active males must be informed that taking the study treatment may involve unknown risks to the fetus if pregnancy were to occur during the study, and agree that in order to participate in the study they must adhere to the contraception requirements outlined in Section 4.2 (Exclusion Criteria). If there is any question that the patient will not reliably comply, they should not be entered or continue in the study.

Sexually active males must be informed of the requirement to wear a condom for the following reasons:

- Prevent pregnancy in a female partner

AND

Prevent potential delivery of investigational drug via seminal fluid to their partner as study treatment may involve unknown risks to the fetus if pregnancy were to occur

There may be unknown risks of LYS228 which may be serious.

3.7 Blood sample volumes

Approximately 250 mL of blood is planned to be collected over a period of approximately 28 days, from each patient as part of the study. Additional samples may be required for safety monitoring.

Timings of blood sample collection are outlined in the Assessment schedule (Section 8.1).

A summary blood log is provided in the SOM. Instructions for all sample collection, processing, storage and shipment information is also available in the SOM and Central Laboratory Manual.

See Section 8.9 regarding the potential use of residual samples.

4 Population

Approximately 60 patients will be randomized to LYS228 or a comparator in 2:1 ratio and are expected to be included in the microbiologic modified intention to treat (micro-mITT) population, as defined in Section 11.1.

Patients that do not meet criteria for inclusion in the efficacy analysis sets may be replaced as needed. Additionally, patients who discontinue the study for reasons other than safety may also be replaced.

The investigator or designee must ensure that all patients being considered for the study meet the following eligibility criteria. No additional criteria should be applied by the investigator, in order that the study population will be representative of all eligible patients.

Patient selection is to be established by checking through all eligibility criteria at screening. A relevant record (e.g. checklist) of the eligibility criteria must be stored with the source documentation at the study site.

Deviation from **any** entry criterion excludes a patient from enrollment into the study.

Additional patients may be enrolled in the event that the sponsor recommends changes or additional dosing regimens, or change in the infusion procedures based on emerging pharmacokinetics or safety data.

4.1 Inclusion criteria

1. Written informed consent must be obtained by one of the following mechanisms before any study assessment can be performed:
    a. A patient who has capacity to give informed consent, signs and dates the informed consent form;
    b. A patient who has capacity to give informed consent, is physically unable to sign and date the informed consent form (e.g. due to inability to write), but gives consent and an impartial witness signs and dates the consent form to indicate that consent was given .
    c. A patient who lacks capacity to give informed consent, is enrolled after a Legally Authorized Representative signs and dates the informed consent form, giving consent on the patient's behalf using the principle of substituted judgment.

Note: b and c applies only if permitted by local country regulations, and the governing ethics committee, which shall determine the qualifications of the Legally Authorized Representative.

2. Male and female patients 18 to 65 years inclusive, and after interim analysis of 8 patients (see Section 3.5 and Section 11.8) patients up to 85 years, inclusive, with visual confirmation (presence of pus within the abdominal cavity) of an intra-abdominal infection associated with peritonitis including at least 1 of the following diagnoses during surgical intervention (e.g. laparotomy, laparoscopic surgery, or percutaneous draining of an abscess):
   - Cholecystitis (including gangrenous cholecystitis) with rupture, perforation, or progression of the infection beyond the gallbladder wall;
   - Diverticulitis with perforation or abscess;
   - Appendicitis with appendiceal perforation or periappendiceal abscess;
   - Acute gastric or duodenal perforation, if operated on >24 hours after perforation;
   - Traumatic perforation of the intestine, if operated on >12 hours after perforation;
   - Secondary peritonitis or intra-abdominal abscess due to perforated viscus, or spread from other focus of infection

     And one of the following:

     a. Patient has undergone surgery for the inclusionary infection and pus has been visualized within the peritoneal cavity, either directly (i.e. by laparotomy or laparoscopy), or by aspiration.
     b. Surgical intervention is scheduled to take place within 24 hours of entry and all criteria below are met:
        - The patient has fever (>38ºC), hypothermia (with a core temperature <35 ºC), leukocytosis (>12,000/µL), or chills.
        - The patient has abdominal pain, nausea, vomiting, tenderness to palpation, rebound tenderness, or guarding.
        - Radiologic imaging findings support inclusionary diagnosis
3. Patients receiving antibiotic treatment of any duration judged to be inadequate due to persistent signs and symptoms of infection and documented bacterial resistance to the antibiotic received provided they meet all of the following criteria:
   a. The patient had a surgical intervention (e.g. laparotomy, laparoscopic surgery, or percutaneous draining of an abscess) that was completed within 24 hours prior to study entry or is scheduled to have such an intervention no more than 24 hours after study entry
   b. Findings of persistent infection were documented at the time of surgical intervention
   c. Specimens for bacterial cultures and susceptibility testing from the intra-abdominal focus of infection were taken during surgical intervention
   d. No further non-study antibiotic therapy is administered after randomization
4. Patients must weigh at least 40 kg to participate in the study, and must have a body mass index (BMI) within the range of 18 - 35 kg/m^2. BMI = Body weight (kg) / [Height (m)]^2

5. Expectation that, in the judgment of the Investigator, the patient will survive with effective antibiotic therapy and appropriate supportive care for the anticipated duration of the study.
6. Able to communicate well with the investigator, to understand and comply with the requirements of the study.

4.2 Exclusion criteria

1. Use of other investigational drugs at the time of enrollment, or within 5 half-lives of enrollment, or within 30 days, whichever is longer; or longer if required by local regulations.
2. Any of the following:
   - Abdominal wall abscess
   - Small bowel obstruction
   - Traumatic bowel perforation undergoing surgery within 12 hours
   - Perforation of gastroduodenal ulcer with surgery within 24 hours
   - An intra-abdominal process that is not likely caused by infection
   - Simple cholecystitis
   - Gangrenous cholecystitis without rupture
   - Simple appendicitis
   - Acute suppurative cholangitis
   - Infected necrotizing pancreatitis
   - Pancreatic abscess
   - Qualifying cIAI managed by staged abdominal repair
   - Source control of infection is not likely to be achieved
   - Perinephric abscess or peritonitis due to an indwelling peritoneal dialysis catheter
   - Qualifying cIAI diagnosis suspected to be due to fungus, parasites (e.g. amoebic liver abscess), virus, or tuberculosis.
3. Received prior antibiotics within 72 hours before the initiation of study therapy, with the following exceptions:
   - The patient received less than 24 hours of total prior antibiotic therapy (based on the dosing interval)
   - Patients receiving ongoing antibacterial drug prophylaxis that will be discontinued at study screening and until study completion
   - Patients who received antibacterial drugs for surgical prophylaxis and then developed cIAI
   - Patients who meet inclusion criteria number 4
4. Received long-acting antibiotic (≥24 hours dosing interval) intra- or post-operatively
5. Received any non-study antibiotic more than 6 hours post-procedure
6. Concomitant bacterial infection at time of enrollment requiring non-Study Drug antibiotics and that may interfere with the evaluation of clinical response to the study antibiotic

7. Non-abdominal bacterial infection diagnosed within 7 days prior to enrollmentincluding endocarditis, osteomyelitis, abscess, meningitis, or pneumonia.
8. Patient has APACHE II score > 30 (Knaus et al 1985) or is considered, in the judgement of the investigator, unlikely to survive 4 weeks (e.g. rapidly progressive terminal illness, including septic shock)
9. Patients who require vasopressors or other agents for hemodynamic support, or admission to an intensive care unit due to critical illness
10. Patient cannot tolerate administration of intravenous Study Drug (≥1 L of fluid per day)
11. Patients that meet sepsis criteria as defined by the quick sequential sepsis-related organ failure assessment (qSOFA) (Singer et al 2016)
    - Respiratory rate ≥ 22 breaths per minute AND
    - Altered mentation AND
    - Systolic BP ≤ 100 mmHg

Assessment can be repeated once after fluid resuscitation and treatment of fever

12. Pregnant or nursing (lactating) women, where pregnancy is defined as the state of a female after conception and until the termination of gestation, confirmed by a positive human chorionic gonadotropin (hCG) laboratory test
13. Women of child-bearing potential, defined as all women physiologically capable of becoming pregnant, unless they are using highly effective methods of contraception during dosing and for 7 days after stopping all study treatment. ***Highly effective contraception methods include:***
    - Total abstinence (when this is in line with the preferred and usual lifestyle of the patient. Periodic abstinence (e.g. calendar, ovulation, symptothermal, post-ovulation methods) and withdrawal are not acceptable methods of contraception.
    - Female sterilization (have had surgical bilateral oophorectomy with or without hysterectomy), total hysterectomy or tubal ligation at least six weeks before taking investigational drug. In case of oophorectomy alone, only when the reproductive status of the woman has been confirmed by follow up hormone level assessment
    - Male sterilization (at least 6 months prior to screening). For female patients on the study the vasectomized male partner should be the sole partner for that patient (see SOM regarding vasectomized partners)
    - Use of oral, injected or implanted hormonal methods of contraception or placement of an intrauterine device (IUD) or intrauterine system (IUS) or other forms of hormonal contraception that have comparable efficacy (failure rate <1%), for example hormone vaginal ring or transdermal hormone contraception
    - In case of use of oral contraception women should have been stable on the same pill for a minimum of 3 months before taking investigational drug

14. Sexually active males unwilling to use a condom during intercourse during treatment period and for 1 day (> 5* T1/2) after completion of treatment. A condom is required for all sexually active male participants, including vasectomized men, to prevent them from fathering a child AND to prevent potential delivery of the drug via seminal fluid.
15. Patients with an estimated creatinine clearance of < 30mL/min calculated based on the study qualified formula or who require renal replacement therapy [hemodialysis, peritoneal dialysis or continuous veno-venous hemofiltration (CVVH)]
16. Any of the following lab abnormalities at screening:
    - ALT, γ-GT, and / or AST ≥ 3 x the upper limit of normal (ULN)
    - Serum bilirubin ≥ 3 mg/dL
    - Hemoglobin levels ≤ 9 g/dL
    - Thrombocytopenia as defined by ≤ 100,000 platelets/μL
    - Neutropenia as defined by ≤ 1,000 polymorphonuclear leukocytes /μL
17. History or current diagnosis of ECG abnormalities indicating significant safety risk for participation in the study, such as:
    - Concomitant clinically significant cardiac arrhythmias (e.g. sustained ventricular tachycardia and clinically significant second or third degree AV block without a pacemaker)
    - History of familial long QT syndrome or known family history of Torsades de Pointes
18. History of seizures requiring current treatment with an anti-seizure medication.
19. History of adrenal disease that requires the use of steroids (≥ 60 mg prednisone or equivalent).
20. History of hypersensitivity to, Study Drug or beta-lactam antibiotics (e.g. penicillins, cephalosporins, carbapenems, or monobactams), metronidazole
21. History of malignancy of any organ system (other than localized basal cell carcinoma of the skin), unless considered cured or in remission (not receiving therapy and no evidence of local recurrence or metastases that require therapy).

22. History of positive HIV (confirmatory testing) except patients receiving effective antiretroviral therapy and with documented virologic control (HIV RNA < 500 copies/mL) and an absolute CD4 1 cell count of ≥350 cells/mm^3 within the preceding 6 months
23. Patients with estimated glomerular filtration rate <30mL/min calculated based on study qualified formula or who require renal replacement therapy (hemodialysis, peritoneal dialysis, or continuous veno-venous hemofiltration
24. Taking medications prohibited by the protocol [as detailed in Section 5.2 (Prohibited treatment)].

    No additional exclusions may be applied by the investigator, in order to ensure that the study population will be representative of all eligible patients.

5 Restrictions for Study Patients

For the duration of the study, the patients should be informed and reminded of the restrictions outlined in this section.

5.1 Contraception requirements

Women of child bearing potential must be informed that taking the study treatment may involve unknown risks to the fetus if pregnancy were to occur during the study, and agree that in order to participate in the study they must adhere to the contraception requirement outlined in the Section 4.2 (Exclusion Criteria).

Sexually active males should be reminded of the requirement to wear a condom for the following reasons:

Prevent pregnancy in a female partner

AND

Prevent delivery of investigational drug via seminal fluid to their partner as study treatment may involve unknown risks to the fetus if pregnancy were to occur

If there is any question that the patient will not reliably comply in the judgement of the investigator, the patient should not be entered or continue in the study. Male patients should be informed that if a female partner becomes pregnant while he is enrolled in the study, contact with the female partner will be attempted to request her consent to collect pregnancy outcome information. Please refer to Section 4.2 (Exclusion criteria) for details of contraception requirements for the study.

5.2 Prohibited treatment

The use of effective concomitant systemic antibiotics with activity against Gram-negative bacteria (oral, IV, or intramuscular) in addition to those designated in the two study groups is prohibited.

Antibiotic therapy for documented clinical or microbiologic failures (rescue antibiotic therapy) or required to treat adverse events are permitted. Antifungals or antivirals are allowed to treat concomitant non-bacterial infections.

No other clinically relevant drug-drug interactions are expected.

5.3 Dietary restrictions and smoking

There is no restriction on diet or on smoking status.

5.4 Other restrictions

There are no other restrictions

6 Treatment

6.1 Study treatment

LYS228 (Arm 1) will be provided by the sponsor

Standard of care intravenous antibiotic therapy (Arm 2) and alternative antibiotics used in the event of change from Study Drug will be provided locally.

Details on the requirements for storage and management of study treatment, and instructions to be followed for patient numbering, prescribing/dispensing and taking study treatment are outlined in the SOM and the PM.

6.1.1 Investigational treatment and control drugs

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LYS228 will be packed and labeled under responsibility of Novartis and will be supplied to the Investigator sites as open label bulk supply. LYS228 preparation instructions will be provided in the PM.

Antibiotics to be used in combination with LYS228 for treatment of Gram-positive and anaerobic bacteria (vancomycin and metronidazole) will be provided locally and prepared consistent with local practices.

The preferred standard of care antibiotic to be used as an active comparator for LYS228 is piperacillin / tazobactam, the active comparator will be provided locally and prepared consistent with local practices. Every reasonable effort should be made to ensure that the preferred standard of care antibiotic is used (piperacillin/tazobactam). In instances such as documented drug resistance, poor tolerability, or drug shortages, the preferred standard of care control can be modified to meropenem (see Section 3.4).

Alternative antibiotics with Gram-positive activity only (daptomycin or linezolid) are not considered prohibited medications, and may be used based on microbiologic susceptibility data. Alternative antibiotics used as rescue therapy or in the event of change from Study Drug will be provided locally.

Table 6-1 Overview of study medication

Study drug name	Formulation	Unit dose	Packaging	PSourcing
LYS228	Corporate Confidential Information		Open label patient specific kits	Provided by Novartis
Vancomycin	Commercially available presentation			Locally
Metronidazole	Commercially available presentation			Locally
Piperacillin/Tazobactam Alternatively to Piperacillin/Tazobactam*: Meropenem	Commercially available presentation			Locally

*Alternative standard of care antibiotic regimen (meropenem) may only be used if Piperacilling/Tazobactam treatment regimen cannot be used (e.g. in the case of resistance)

6.1.2 Alternative and rescue antibiotic treatment

Alternative oral antibiotics allowed after completion of intravenous Study Drug or rescue antibiotics used during the treatment period, as described in Section 3, will be provided locally by the sites.

6.2 Treatment arms

Patients will be randomized to one of the following two treatment arms in a ratio of 2:1.

- Arm 1: LYS228 (n=40). LYS228 dose for each patient will be based on the renal function as described in Section 3.3. Vancomycin and metronidazole will be given in addition to LYS228 to treat Gram-positive and anaerobic spectra of bacteria based on local standard of care procedures.
- Arm 2: Standard of care intravenous antibiotic for the treatment of cIAI (n=20) with activity against Gram-negative, Gram-positive and anaerobic bacteria based on local standard of care procedures.

Adjustment of the non-LYS228 Study Drug components of treatment arms may be permitted based on microbiologic data as detailed in the SOM

6.3 Treatment assignment and randomization

The randomization numbers will be assigned using an Interactive response Technology (IRT). Please refer to the SOM for the process and timing of treatment assignment and randomization of patients.

6.4 Treatment blinding

This is an evaluator-blinded study. The physician performing the safety and clinical response assessments will remain blinded to study treatment throughout the study, except where indicated below.

This will require the investigator site staff to apply certain restrictions to ensure the evaluator physician does not have access to study documents, and does not observe the drug administration because the Study Drugs are provided in an open-label fashion. A double-dummy design is not feasible, as explained in Section 3.3. Further details can be found in the SOM

With the exception of the blinded evaluator, all site staff (including study investigator and study nurse) will be unblinded to study treatment during treatment allocation and patient dosing.

The Sponsor staff will be unblinded in this study.

Table 6-2 Blinding and unblinding plan

Role	Time or Event		
	Randomization list generated	Treatment allocation and administration	Safety emergency event (unblinding of a single patient)
Patients	B	UI	UI
Evaluator physician	B	B	UI
Unblinded site staff (see text for details)	B	UI	UI
Drug supply and randomization office	UI	UI	UI
Unblinded sponsor staff	B	UI	UI
Statistician/statistical programmer/data analysts	B	UI	UI
Independent committees used for assessing interim results	NA	NA	NA

B Remains blinded

NA Not applicable

UI Allowed to unblinded on individual patient level

6.5 Treating the patient

See the SOM and the PM for further details on LYS228 administration. All other antibiotics will be administered according to local practices.

Sponsor qualified medical personnel will be readily available to advise on trial related medical questions or problems.

6.6 Permitted dose adjustments and interruptions of study treatment

Dose adjustments are permitted during the study to adjust for changes in renal function.

As described in Section 3.3 and the SOM, the dose of LYS228 is selected according to estimated CrCl at screening. Patients with an estimated CrCl $\geq$ 90 ml/min will receive every 6 hours and patients with an estimated CrCl < 90/mL/min will receive every 6 hours. During the treatment period, if changes in estimated CrCl occur, the LYS228 dose must be adjusted accordingly with the study formula as described in the PM. In case of notable adverse events, safety concerns, or emerging pharmacokinetic data, administration of an LYS228 dose below the initial dose, or adjustments to the LYS228 infusion may be considered in consultation with the sponsor or by the sponsor. Other study treatment dose adjustments and/or interruptions are not permitted.

These changes will be recorded on the Dosage Administration Record CRF.

6.7 Emergency breaking of assigned treatment code

Not applicable. The Study Drugs will be administered in an open-label fashion. The blinded evaluator designated by the investigator may have access to the treatment assignment if needed.

6.8 Treatment exposure and compliance

IV infusions will be administered at the study site. The site staff will instruct patients to take any medications administered in an out-patient setting as prescribed. LYS228 concentrations will be measured in all patients treated with LYS228, as detailed in Section 8.5.

6.9 Recommended treatment of adverse events

Adverse events should be treated per local standard of care. Corporate Confidential Information Medications used to treat AEs must be recorded on the Concomitant medications/Significant non-drug therapies CRF.

6.10 Rescue antibiotic medication

The use of rescue antibiotics is at the discretion of the investigator, and should be limited to instances of clinical failure as defined in Section 11.4.1 including persistence or worsening of symptoms of cIAI. Symptoms of cIAI and clinical failures must be recorded in the CRF. Use of rescue medication must be recorded in the CRF.

6.11 Concomitant treatment

The investigator must instruct the patient to notify the study site about any new medications he/she takes after the patient was enrolled into the study.

All prescription medications, over-the-counter drugs and significant non-drug therapies (including physical therapy and blood transfusions) administered or taken within the timeframe defined in the entry criteria prior to the start of the study and during the study, must be recorded on the Concomitant medications/Significant non-drug therapies CRF. Administration of supportive intravenous fluids, enteric or parenteral nutrition, and therapies for local wound care will be captured only in the source documents.

Medication entries should be specific to trade name, the single dose and unit, the frequency and route of administration, the start and discontinuation date and the reason for therapy.

Each concomitant drug must be individually assessed against all exclusion criteria/prohibited medications. If in doubt, the investigator should contact Novartis before randomizing a patient or, if the patient is already enrolled, to determine if the patient should continue participation in the study.

7 Study completion and discontinuation

7.1 Study completion and post-study treatment

Each patient will be required to complete the study in its entirety and thereafter no further study treatment will be made available to them.

Study completion (end of trial) is defined as when the last patient completes their EOS visit, and any repeat assessments associated with this visit have been documented and followed-up appropriately by the Investigator, or in the event of an early study termination decision, the date of that decision.

All treated patients should have a safety follow-up call conducted 30 days after End of Study Drug. The information collected is kept as source documentation. All SAEs reported during this time period must be reported as described in Section 9.2 and the SOM. Documentation of attempts to contact the patient should be recorded in the source documentation.

7.2 Discontinuation of study treatment

Discontinuation of the study treatment for a patient occurs when study treatment is stopped earlier than the protocol planned duration.

Study treatment for an individual patient must be discontinued under the following circumstances:

- Patient/guardian decision - patients may choose to discontinue study treatment for any reason at any time.
- The investigator believes that continuation would negatively impact the safety of the patient or the risk/benefit ratio of trial participation. This might include patients that have bacterial isolates in blood or cIAI focus of infection cultures at baseline that are not susceptible to study treatment
- Any protocol deviation that results in a significant risk to the patient's safety.
- Pregnancy [see Section 8.6 (Safety) and Section 9.6 (Pregnancy reporting)]

Study Drug treatment for an individual patient must be placed on hold until a full review of the safety data occurs for any of the following events/findings:

- Use of prohibited treatment as outlined in Section 5.2
- Emergence of the following adverse events:
  - One AE of grade 3 or higher by Common Terminology Criteria for Adverse Events (CTCAE), version 5.0, (2017) criteria requiring intervention considered to be related to Study Drug or SAE believed to be related to Study Drug.
  - Creatinine increase to 2.0 x baseline considered to be related to Study Drug
  - $\geq$ Grade 2 decrease in platelet count ($<70x10^9$/L) considered to be related to Study Drug
  - Any evidence (clinical signs and symptoms) of seizure activity. NOTE: Patients can continue Study Drug if an alternative (not Study Drug- related) cause is clear and likely.
  - $\geq$ Grade 2 hypersensitivity reaction as defined by flushing or rash, temperature >38°C or therapeutic intervention required. Immediate interruption of the study treatment infusion is required in such cases. NOTE: Patients can continue Study Drug if an alternative cause (not Study Drug-related) is clear and likely.
- Clinical or microbiologic failure as defined in Section 11.4.2.
- If a liver or renal event occurs, follow guidelines outlined in Appendix 1 and Appendix 2 regarding discontinuation of study treatment

- Any medical condition, laboratory abnormalities, or adverse event that in the judgment of the investigator and the sponsor justify dosing discontinuation for the individual patient, taking into consideration the patient's overall status.

Patients that miss one or more doses of Study Drug may continue in the study if the investigator feels that this is the appropriate course of therapy, unless the patient missed doses because of an adverse event related to administration of the Study Drug. If an alternative cause is identified, such patients may continue Study Drug.

If discontinuation of study treatment occurs, the investigator must determine the primary reason for the patient's premature discontinuation of study treatment and record this information on the Dosage Administration CRF.

Patients who discontinue study treatment or who decide they do not wish to participate in the study further should NOT be considered withdrawn from the study UNLESS they withdraw their consent (see Section 7.3, Withdraw of Informed Consent). If they fail to return for the EOS assessments for unknown reasons, every effort (e.g. telephone, e-mail, letter) should be made to contact the patient/pre-designated contact as specified in Section 7.4 (Lost to follow-up). This contact would preferably be done according to the study visit schedule.

After study treatment discontinuation, at a minimum, in abbreviated visits, the following data should be collected at clinic visits or via telephone/email contact:

- new / concomitant treatments
- adverse events/Serious Adverse Events

The investigator must also contact the IRT to register the patient's discontinuation from study treatment. Please refer to the SOM and IRT Manual for details.

7.3 Withdrawal of informed consent

Patients may voluntarily withdraw consent to participate in the study for any reason at any time.

Withdrawal of consent occurs only when a patient:

- Does not want to participate in the study anymore and
- Does not allow further collection of personal data

In this situation, the investigator must make every effort (e.g. telephone, e-mail, letter) to determine the primary reason for the patient's decision to withdraw his/her consent and record this information. Study treatment must be discontinued and no further assessments conducted, and the data that would have been collected at subsequent visits will be considered missing. Further attempts to contact the patient are not allowed unless safety findings require communicating or follow-up. All efforts should be made to complete the assessments prior to study withdrawal. A final evaluation at the time of the patient's study withdrawal should be made as detailed in the assessment table.

Novartis/*sponsor* will continue to keep and use collected study information (including any data resulting from the analysis of a patient's samples until their time of withdrawal) according to applicable law.

7.4 Lost to follow-up

For patients whose status is unclear because they fail to appear for study visits without stating an intention to discontinue or withdraw, the investigator should show "due diligence" by documenting in the source documents steps taken to contact the patient, e.g. dates of telephone calls, registered letters, etc. A patient cannot be formally considered lost to follow-up until his/her scheduled end of study visit would have occurred.

7.5 Study Stopping rules

The investigators and the sponsor will continually review adverse events and laboratory findings during the study.

The study may be stopped (i.e. stop new enrollment and stop dosing for all patients) in case of safety concerns that are suspected to be related to Study Drug, including:

- ≥ 5 patients have a Study Drug-related Grade 2 increase in serum creatinine (≥ 1.5 x baseline)
- ≥3 patients have a Study Drug-related similar adverse event of Grade 3 or higher (per CTCAE scoring)
- ≥2 patients have a Study Drug-related seizures
- The principal investigator or the sponsor consider that the number and/or severity and/or system organ class of adverse events justify discontinuation of drug administration
- The sponsor unilaterally requests it

The severity of adverse events will be graded by the study site investigator (or his designee) based on clinical judgment using CTCAE v. 5.0. CTCAE grades will be used in the source documents to quantify events that may lead to patient's discontinuation. Safety reviews will be conducted jointly between medically qualified representatives of the sponsor and Investigators. Dosing can be reviewed provided the safety data supports this.

7.6 Early study termination by the sponsor

The study can be terminated by Novartis at any time for any reason. This may include reasons related to the benefit/risk assessment of participating in the study, practical reasons (including slow enrollment), or for regulatory or medical reasons. Should this be necessary, patients must be seen as soon as possible and treated as a prematurely discontinued patient. The investigator may be informed of additional procedures to be followed in order to ensure that adequate consideration is given to the protection of the patients' interests. The investigator will be responsible for informing IRBs/IECs of the early termination of the trial.

8 Procedures and assessments

8.1 Assessment schedule

Patients should be seen for all visits/assessments as outlined in the assessment schedule or as close to the designated day/time as possible.

Missed or rescheduled visits should not lead to automatic discontinuation. Patients who prematurely discontinue the study for any reason should be scheduled for a visit as soon as possible, at which time all of the assessments listed for the final visit will be performed. At this final visit, all dispensed investigational product should be reconciled, and the adverse event and concomitant medications recorded on the CRF.

Table 8-1 Assessment schedule

Study phase	Screening/ Baseline	Treatment															Follow up
Visit Name	Screening/ Baseline	Minimum Study Drug treatment visits					Additional Study Drug treatment visits[2,3,4]									End of Study Drug (EOSD) visit[2,3,4]	End of Antibiotic Treatment (EOAT) visit[3,4,5]	End of Study (EOS) visit
Visit Number[1]	V1	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	199
Days	-1 to 1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Flexible, 5 up to 15	Flexible, 6 up to 15	28
Informed consent	X																	
Inclusion / Exclusion criteria	S[6]																	
Medical history/current medical conditions	X																	
Demography	X																	
Clinical evaluation of IAI	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Corporate Confidential Information	X[7]	As Required[7]																
	X	As Required[8]																

Study phase	Screening/ Baseline	Treatment															Follow up
Visit Name	Screening/ Baseline	Minimum Study Drug treatment visits					Additional Study Drug treatment visits[2,3,4]								End of Study Drug (EOSD) visit[2,3,4]	End of Antibiotic Treatment (EOAT) visit[3,4,5]	End of Study (EOS) visit
Visit Number[1]	V1	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	199
Days	-1 to 1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Flexible, 5 up to 15	Flexible, 6 up to 15	28
Estimated creatinine clearance[9]	X	As Required																X
Physical Examination	S[6]	As Required (S)[6]																
Pregnancy and assessments of fertility	X																	X
Body Height	X																	
Body Weight	X																	X
Body Temperature	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood Pressure	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Pulse rate	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Urinalysis	X	X[10]			X											X	X	X
Hematology	X	X[10]			X			X			X			X		X	X	X
Blood chemistry	X	X[10]			X			X			X			X		X	X	X
ECG evaluation	X															X		X
Study Drug administration[11]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X		
Switch to alternative (oral) antibiotic																As Required		
Alternative (oral) antibiotic compliance																	X	
Rescue Antibiotic	As Required																	

Study phase	Screening/ Baseline	Treatment															Follow up
Visit Name	Screening/ Baseline	Minimum Study Drug treatment visits					Additional Study Drug treatment visits[2,3,4]									End of Study Drug (EOSD) visit[2,3,4]	End of Antibiotic Treatment (EOAT) visit[3,4,5]	End of Study (EOS) visit
Visit Number[1]	V1	101	102	103	104	105	106	107	108	109	110	111	112	113	114	115	116	199
Days	-1 to 1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Flexible, 5 up to 15	Flexible, 6 up to 15	28
PK blood collection[12]		X[13]	X[14]	X[14]	X[14]	X[2,15]		X[14]		X[14]		X[14]		X[14]		X[14]		
Corporate Confidential Information	X																	
		X																
Adverse Events	As Required																	
Serious Adverse Events[17]	As Required																	
Concomitant therapies	As Required																	
Comments	As Required																	
Safety Follow up Call																		S[6,18]

[1]Visit structure given for internal programming purpose only

[2]Upon discontinuing Study Drug, patients must undergo an End of Study Drug visit. That day's scheduled Study Drug treatment visit/assessments will be replaced by the End of Study Drug visit/assessments; with the exception that, if an End of Study Drug visit takes place on Day 5, PK blood collection must still be performed on Day 5

[3]After completing an End of Study Drug visit, patients who are switched to alternative antibiotics are only required to complete the End of Antibiotic Treatment and End of Study Visits. All other treatment visits and their relevant assessments are no longer required

[4]After completing an End of Study Drug visit, patient who discontinue antibiotic therapy (i.e. are not switched to alternative antibiotics) are only required to complete the End of Study Visit. All other visits and their relevant assessments are no longer required

[5]End of Antibiotic Treatment visit only applies to patients who are switched to alternative antibiotics. This visit must take place after discontinuing alternative antibiotics.

[6]S= assessment will be recorded on source documentation only and will not be entered into the CRF

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[9] Calculated based on SCr analyzed by local lab.

[10]If Screening visit occurs within 24 hours of Day 1, the urinalysis, hematology, and blood chemistry assessments do not need to be performed on Day 1

[11]Study Drug will be administered for a minimum of 5 days and up to 14 days. Study Drug no longer administered after a switch to alternative antibiotics

[12]PK samples only to be collected for patients randomized to receive LYS228.

[13]Samples to be collected at 1, 2, 4 hours after the start of any ONE Day 1 LYS228 infusion.

[14]One pre-dose sample to be collected before any ONE LYS228 dose on Days 2, 3, 4, 7, 9, 11, 13 and EOSD visit.

[15]Samples to be collected Pre-dose and at 1, 2, 3, 4, and 6 hours after the start of any ONE Day 5 LYS228 infusion.

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[17]Serious Adverse Events (SAEs) must be reported according to the timeframe specified in the protocol

[18]Post study safety contact is via phone or email and will take place 30 days after End of Study Drug. Information collected will be kept as source documentation only; Exception: in the case of a reported SAE, follow instructions for SAE reporting outlined in the protocol and SOM

8.2 Informed consent procedures

As described in Section 4.1 written consent is preferred and may be the only method acceptable according to certain local laws and regulations. If verbal assent is obtained (where permissible), a patient's representative must be present and written consent by the patient must be obtained as soon as the patient is capable of providing it. Informed consent/assent must be witnessed, where required by law or regulation and using an IRB/IEC-approved document.

If applicable, in cases where the subject's representative gives consent (if allowed according to local requirements), the patient must be informed about the study to the extent possible given his/her understanding. If the patient is capable of doing so, he/she must indicate assent by personally signing and dating the written informed consent document or a separate assent form.

Informed consent must be obtained before conducting any study-specific procedures (e.g. all of the procedures described in the protocol). The process of obtaining informed consent must be documented in the patient source documents.

Novartis will provide to investigators a proposed informed consent form that complies with the ICHE6 GCP guideline and regulatory requirements and is considered appropriate for this study. The informed consent form will also include a section related to optional future research which will require a separate signature if the patient agrees to future research. The procedures set out in the main consent form concerning the storage, maintenance of privacy, and release of the data or specimens for the main study will also be adhered to for any future research. Any changes to the proposed consent form suggested by the investigator must be agreed to by Novartis before submission to the IRB/IEC.

Information about common side effects already known about the investigational drug can be found in the Investigator's Brochure (IB). This information will be included in the patient informed consent and should be discussed with the patient during the study as needed. Any new information regarding the safety profile of the investigational drug that is identified between IB updates will be communicated as appropriate, for example, via an Investigator Notification or an Aggregate Safety Finding. New information might require an update to the informed consent and then must be discussed with the patient.

Ensure patients are informed of the contraception requirements outlined in the Section 4.2 (Exclusion criteria) and in Section 5.1 (Contraception requirements).

A copy of the approved version of all consent forms must be provided to the Novartis monitor after IRB/IEC approval.

Refer to the SOM for a complete list of Informed Consent Forms included in this study.

8.3 Patient screening

It is permissible to re-screen a patient one time only if s/he fails the initial screening; however, each case must be discussed and agreed with the Sponsor on a case-by-case basis. In the case of the laboratory assessment at screening is outside of the range, the assessment may be repeated once prior to enrollment. If the repeated value remains outside of the specified ranges, the patient will be excluded from the study.

Information on what data should be collected for screening failures is outlined in the SOM.

8.4 Patient demographics/other baseline characteristics

Patient demographic and baseline characteristic data will be collected on all patients. Relevant medical history/current medical conditions data will also be collected until signature of informed consent. Details are outlined in the Site Operations Manual.

Investigators have the discretion to record abnormal test findings on the medical history CRF, if in their judgment, the test abnormality occurred prior to the informed consent signature.

8.5 Efficacy / Pharmacodynamics

8.5.1 Clinical evaluation of cIAI infection

A blinded evaluator will conduct the clinical evaluation necessary to determine clinical response at the time-points defined in the Assessment schedule (Section 8.1). For further details on evaluator blinding, please refer to the SOM.

The following data will be recorded in the CRF as part of the clinical evaluation of cIAI infection:

- Underlying inclusionary diagnosis (Inclusion criterion 2)
- Signs and symptoms of intra-abdominal infection: Onset, resolution or improvement, determined by CTCAE grading criteria. Signs and symptoms will be recorded daily during hospitalization and at the EOSD, EOAT, and EOS visits.
    - Present at inclusion, or before surgical intervention if enrolled post-operatively: fever (defined as body temperature > 38ºC), hypothermia with a core body temperature <35ºC, elevated WBC count (>12,000/µL), chills, abdominal pain, nausea, vomiting, tenderness to palpation, rebound tenderness, and guarding
    - New signs and symptoms attributed to progression of cIAI. Progression of infection other than cIAI (e.g. sepsis) will be captured as an SAE as detailed in Section 9.2
- Surgical intervention (e.g. laparotomy, laparoscopic surgery, or percutaneous draining of an abscess)
    - Primary intervention for baseline (index) cIAI
    - Re-intervention for progression or relapse of cIAI if required 96 hours after the start of Study Drug.

8.6 Safety

Safety assessments are specified below. Methods for assessment and recording are specified in the SOM, with the Assessment schedule (Section 8.1) detailing when each assessment is to be performed.

8.6.1 Physical examination

A complete physical examination will include the examination of general appearance, skin, neck (including thyroid), eyes, ears, nose, throat, lungs, heart, abdomen, back, lymph nodes, extremities, vascular and neurological. If indicated based on medical history and/or symptoms, rectal, external genitalia, breast, and/or pelvic exams may be performed.

Information for all physical examinations must be included in the source documentation at the study site and will not be recorded in the CRF. Significant findings that are present prior to informed consent are included in the Medical History CRF. Significant findings observed after informed consent signature which meet the definition of an Adverse Event must be appropriately recorded on the Adverse Event CRF.

8.6.2 Vital signs

Vital signs include blood pressure (BP), body temperature, and pulse measurements. After the patient has been sitting/supine position for 3 minutes, systolic and diastolic BP will be measured using an automated validated device, with an appropriately sized cuff. In case the cuff sizes available are not large enough for the patient's arm circumference, a sphygmomanometer with an appropriately sized cuff may be used.

8.6.3 Body height and body weight

Height in centimeters (cm) and body weight (to the nearest 0.1 kilogram [kg] in indoor clothing, but without shoes) will be measured.

Body mass index (BMI) will be calculated using the following formula:

- BMI = Body weight (kg) / [Height (m)]2

8.6.4 Laboratory evaluations

Clinically relevant deviations of laboratory test results occurring during or at completion of the study must be reported and discussed with Novartis personnel. The results should be evaluated for criteria defining an adverse event and reported as such if the criteria are met.

Repeated evaluations are mandatory until normalization of the result(s) or until the change is no longer clinically relevant. In case of doubt, Novartis personnel should again be contacted.

Hematology

Hemoglobin, hematocrit, red blood cell count, white blood cell count with differential (e.g., neutrophils, basophils, eosinophils, monocytes, lymphocytes) and platelet count will be measured. Coagulation testing including prothrombin time (PT) also reported as INR and activated partial thromboplastin time (aPTT) will be measured.

Blood chemistry

Albumin, alkaline phosphatase, total bilirubin, bicarbonate/carbon dioxide (CO2), calcium, cholesterol, chloride, creatinine, creatinine kinase (CK), GGT, glucose, lactic dehydrogenase (LDH), inorganic phosphorus, lipase, amylase, magnesium, potassium, total protein, AST, ALT, sodium, triglycerides, blood urea nitrogen (BUN) uric acid. If the total bilirubin concentration is increased above 1.5 times the upper limit of normal, direct and indirect reacting bilirubin should be differentiated.

Urinalysis

A midstream urine sample (approximately 30 mL) will be obtained from patients with no urinary catheters, in order to avoid contamination with epithelial cells and sediments, and allow proper assessments. A semi-quantitative “dipstick” evaluation for the following parameters will be performed: specific gravity, pH, glucose, protein, bilirubin, ketones, nitrite, leukocytes and blood. If the dipstick result is positive for protein, nitrite, leucocytes and/or blood, the sample will be sent for microscopic analysis of WBC, RBC and casts.

8.6.5 Electrocardiogram (ECG)

Full details of all procedures relating to the ECG collection and reporting are contained in the SOM.

PR interval, QRS duration, heart rate, RR interval, QT interval, QTc.

The Fridericia QT correction formula (QTcF) must be used for clinical decisions.

As applicable, QTcF and QTcB may be calculated in-house. Unless auto-calculated by the ECG machine, the investigator must calculate QTcF at the Screening visit to assess eligibility. See the Site Operations Manual for additional details.

Clinically significant abnormalities must be reported in the AE CRF.

8.6.6 Pregnancy and assessments of fertility

Pregnancy

All pre-menopausal women who are not surgically sterile will have pregnancy testing. See the Assessment schedule (Section 8.1), for timing of the protocol required pregnancy testing; additional pregnancy testing may be performed to meet local requirements*. A positive urine pregnancy test requires immediate halt of study treatment until serum β-hCG is performed and found to be negative.

*Additional pregnancy testing might be performed if requested per local requirements.

Refer to Section 9.6 for details on Reporting Pregnancy.

Assessments of Fertility

Refer to Section 4.2 for criteria to determine women that are not of child bearing potential.

Medical documentation of oophorectomy, hysterectomy, or tubal ligation must be retained as source, if available. Subsequent hormone level assessment to confirm the woman is not of child bearing potential must also be available as source documentation in the following cases:

- surgical bilateral oophorectomy without a hysterectomy
- reported 12 months of natural (spontaneous) amenorrhea with an appropriate clinical profile.

If a female patient cannot be confirmed to be of non-childbearing potential, highly effective contraception methods should be used as described in Section 4.2.

8.7 Pharmacokinetics

Pharmacokinetic (PK) samples will be collected at the time points defined in the Assessment schedule (Section 8.1). Follow instructions outlined in the Site Operations Manual regarding sample collection, numbering, processing and shipment. See Section 8.9 for information regarding the potential use of residual samples.

In order to better define the PK profile, the timing of the PK sample collection may be altered based on emergent data. The number of samples/blood draws and total blood volume collected will not exceed those stated in the protocol.

Pharmacokinetic (PK) samples will be obtained and evaluated in all LYS228 treated patients.

LYS228 will be determined by a validated LC-MS/MS method; Corporate Confidential Information

Concentrations will be expressed in mass per volume units.

Concentrations below the LLOQ will be reported as "zero" and missing data will be labeled as such in the Bioanalytical Data Report.

For standard pharmacokinetic abbreviations and definitions see the list provided at the beginning of this protocol.

The following pharmacokinetic parameters on the last day of LYS228 dosing will be determined using the actual recorded sampling times and non-compartmental method(s) with Phoenix WinNonlin (Version 6.4 or higher): Cmax, Tmax, AUCTau, , T1/2, %fT>MIC and CL, Vss from the plasma concentration-time data.

The linear trapezoidal rule will be used for AUC calculation. Regression analysis of the terminal plasma elimination phase for the determination of T1/2 will include at least 3 data points after Cmax. If the adjusted R^2 value of the regression analysis of the terminal phase will be less than 0.75, no values will be reported for T1/2.

9 Safety monitoring

9.1 Adverse events

An adverse event (AE) is any untoward medical occurrence (i.e., any unfavorable and unintended sign [including abnormal laboratory findings], symptom or disease) in a patient after *providing written informed consent* for participation in the study until the end of study visit. Therefore, an AE may or may not be temporally or causally associated with the use of a medicinal (investigational) product.

In addition, all reports of intentional misuse and abuse of the study treatment are also considered an adverse event irrespective if a clinical event has occurred. See Section 9.5 for an overview of the reporting requirements.

The occurrence of adverse events must be sought by non-directive questioning of the patient at each visit during the study. Adverse events also may be detected when they are volunteered by the patient during or between visits or through physical examination finding, laboratory test finding, or other assessments.

Abnormal laboratory values or test results constitute adverse events only if they fulfill at least one of the following criteria:

- They induce clinical signs or symptoms,
- They are considered clinically significant,
- They require therapy.

Clinically significant abnormal laboratory values or test results should be identified through a review of values outside of normal ranges/clinically notable ranges, significant changes from baseline or the previous visit, or values which are considered to be non-typical in patients with underlying disease. Investigators have the responsibility for managing the safety of individual patient and identifying adverse events. Alert ranges for liver and kidney related events are included in Appendix 1 and Appendix 2, respectively.

Adverse events should be recorded on the Adverse Events CRF under the signs, symptoms or diagnosis associated with them, and accompanied by the following information:

1. The severity grade: the Common Toxicity Criteria (CTC) AE grade 5.0 CTCAE grading will be provided separately. If CTCAE grading does not exist for an adverse event, use:

   1=mild,

   2=moderate,

   3=severe

   4=life threatening* (see Section 9.2 for definition of a serious adverse event (SAE))

   *Note: There may be cases where a CTCAE with a grade of 4 (life -threatening) may not necessarily be an SAE (e.g. certain laboratory abnormalities in the absence of meeting other seriousness criteria).

   CTC-AE grade 5 (death) is not used, but is collected as a seriousness criteria and also collected in other CRFs (e.g. Study Completion, Death/Survival).

2. its relationship to the study treatment:
   - Yes or No

3. its duration (start and end dates) or if the event is ongoing an outcome of not recovered/not resolved must be reported.
4. whether it constitutes a serious adverse event (see Section 9.2 for definition of SAE) and which seriousness criteria have been met
5. action taken regarding investigational treatment.

   All adverse events must be treated appropriately. Treatment may include one or more of the following:

   - no action taken (e.g. further observation only)
   - investigational treatment dosage increased/reduced
   - investigational treatment interrupted/withdrawn
   - concomitant medication or non-drug therapy given
   - hospitalization/prolonged hospitalization (see Section 9.2 for definition of SAE)
   - its outcome (not recovered/not resolved; recovered/resolved; recovering/resolving, recovered/resolved with sequelae; fatal; or unknown).

Information about common side effects already known about the investigational drug can be found in the Investigator's Brochure (IB) or will be communicated between IB updates in the form of Investigator Notifications. Once an adverse event is detected, it must be followed until its resolution or until it is judged to be permanent, and assessment should be made at each visit (or more frequently, if necessary) of any changes in severity, the suspected relationship to the investigational drug, the interventions required to treat it, and the outcome.

The investigator must also instruct each patient to report any new adverse event (beyond the protocol observation period) that the patient, or the patient's personal physician, believes might reasonably be related to study treatment. This information must be recorded in the investigator's source documents; however, if the AE meets the criteria of an SAE, it must be reported to Novartis.

Progression of signs and symptoms associated with baseline (index) cIAI will be reported as efficacy endpoints (detailed in Section 8.5) and not as adverse events. Progression of infection other than baseline cIAI (e.g. surgical wound infection) will be reported as AE.

*Refer to the Site Operations Manual for data capture methodology regarding AE collection for patients that fail screening.

9.2 Serious adverse event reporting

9.2.1 Definition of SAE

An SAE is defined as any adverse event (appearance of (or worsening of any pre-existing) undesirable sign(s), symptom(s) or medical conditions(s)) which meets any one of the following criteria:

- is fatal or life-threatening
- results in persistent or significant disability/incapacity
- constitutes a congenital anomaly/birth defect

- requires inpatient hospitalization or prolongation of existing hospitalization, unless hospitalization is for:
  - routine treatment or monitoring of the studied indication, not associated with any deterioration in condition
  - elective or pre-planned treatment for a pre-existing condition (that is unrelated to the indication under study and has not worsened since the start of Study Drug
  - treatment on an emergency outpatient basis for an event not fulfilling any of the definitions of a SAE given above and not resulting in hospital admission
  - social reasons and respite care in the absence of any deterioration in the patient's general condition
  - is medically significant, e.g. defined as an event that jeopardizes the patient or may require medical or surgical intervention.

All malignant neoplasms will be assessed as serious under "medically significant" if other seriousness criteria are not met.

Life-threatening in the context of a SAE refers to a reaction in which the patient was at risk of death at the time of the reaction; it does not refer to a reaction that hypothetically might have caused death if it were more severe (please refer to ICH-E2D Guideline 2004).

Medical and scientific judgment 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 (please refer to ICH-E2D Guideline 2004).

Any suspected transmission via a medicinal product of an infectious agent is also considered a serious adverse reaction.

All AEs (serious and non-serious) are captured on the CRF; SAEs also require individual reporting to the local Novartis Chief Medical Office and Patient Safety (CMO & PS) as per Section 9.2.2.

Progression of baseline to life threatening infection (e.g. sepsis, septic shock) will be reported as SAE.

9.2.2 SAE reporting

Screen Failures

Note the following requirement for Screen Failures: SAEs occurring after the patient has provided informed consent until the time the patient is deemed a Screen Failure must be reported to Novartis.

Treated Patients

To ensure patient safety, every SAE, regardless of causality, occurring after the patient has provided informed consent and until 30 days after the last administration of study treatment must be reported to Novartis within 24 hours of learning of its occurrence as described below.

Any SAEs experienced after this period should only be reported to Novartis if the investigator suspects a causal relationship to study treatment.

All follow-up information for the SAE including information on complications, progression of the initial SAE and recurrent episodes must be reported as follow-up to the original episode within 24 hours of the investigator receiving the follow-up information. An SAE occurring at a different time interval or otherwise considered completely unrelated to a previously reported one must be reported separately as a new event.

Follow- up information provided must describe whether the event has resolved or continues, if and how it was treated, whether the blind was broken or not (if applicable) and whether the patient continued or withdrew from study participation. Each re-occurrence, complication, or progression of the original event must be reported as a follow-up to that event regardless of when it occurs.

If the SAE is not previously documented in the Investigator's Brochure or Package Insert (new occurrence) and is thought to be related to the study treatment a CMO& PS Department associate may urgently require further information from the investigator for Health Authority reporting. Novartis may need to issue an Investigator Notification (IN) to inform all investigators involved in any study with the same study treatment that this SAE has been reported. Suspected Unexpected Serious Adverse Reactions (SUSARs) will be collected and reported to the competent authorities and relevant ethics committees in accordance with EU Guidance 2011/C 172/01 or as per national regulatory requirements in participating countries.

Follow the detailed instructions outlined in the SOM regarding the submission process for reporting SAEs to Novartis. Note: SAEs must be reported to Novartis within 24 hours of the investigator learning of its occurrence/receiving follow-up information.

9.3 Liver safety monitoring

To ensure patients' safety and enhance reliability in determining the hepatotoxic potential of an investigational drug, a standardized process for identification, monitoring and evaluation of liver events has to be followed.

Please refer to Table 15-1-Appendix 1 for complete definitions of liver events.

Follow-up of liver events

Every liver event defined in Table 15-1-Appendix 1 should be followed up by the investigator or designated personnel at the trial site, as summarized below. Additional details on actions required in case of liver events are outlined in Table 15-2 - Appendix 1.

- Repeating liver chemistry tests (ALT, AST, TBL, PT/INR, ALP and G-GT) to confirm elevation within 48-72 hours.

These liver chemistry repeats should be performed using the local laboratory used by the site. Repeat laboratory test results must be reported as appropriate via an electronic data transfer (if applicable), or entered on the appropriate unscheduled local laboratory CRF.

- If the initial elevation is confirmed, close observation of the patient will be initiated, including consideration of treatment halt if deemed appropriate.

- Discontinuation of the investigational drug (refer to Section 7.2 (Discontinuation of study treatment), if appropriate
- Hospitalization of the patient if appropriate
- Causality assessment of the liver event
- Thorough follow-up of the liver event should include
- Repeating liver chemistry tests two or three times weekly. Testing should include ALT, AST, ALP, PT/INR, and GGT. If total bilirubin is elevated > 2 x ULN, fractionation into direct and indirect bilirubin is required. To rule out muscular origin of transaminase elevations, CPK should be measured along with liver chemistry tests. Frequency of retesting can decrease to once a week or less if abnormalities stabilize or the Study Drug has been discontinued and the patient is asymptomatic. Retesting should be continued up to resolution.
- Obtaining a more detailed history of symptoms and prior or concurrent diseases.
- Obtaining a history of concomitant drug use (including nonprescription medications and herbal and dietary supplement preparations), alcohol use, recreational drug use, and special diets.
- Exclusion of underlying liver disease, as specified in Table 15-3.
- Imaging such as abdominal US, CT or MRI, as appropriate
- Obtaining a history of exposure to environmental chemical agents.
- Considering gastroenterology or hepatology consultations.

All follow-up information, and the procedures performed must be recorded as appropriate in the CRF. Refer to the SOM for additional details.

9.4 Renal safety monitoring

Every renal laboratory trigger or renal event must be followed up by the investigator or designated personnel at the trial site. Recommended follow-up assessments are listed in Appendix 2.

All follow-up information, and the procedures performed must be recorded as appropriate in the CRF. Refer to the Site Operations Manual for additional details

9.5 Reporting of study treatment errors including misuse/abuse

Medication errors are unintentional errors in the prescribing, dispensing, administration or monitoring of a medicine while under the control of a healthcare professional, patient/subject or consumer.

Misuse refers to situations where the medicinal product is intentionally and inappropriately used not in accordance with the protocol.

Abuse corresponds to the persistent or sporadic, intentional excessive use of a medicinal product, which is accompanied by harmful physical or psychological effects.

All study treatment errors and uses outside of what is foreseen in the protocol will be collected in the dose administration record (DAR) CRF. Study treatment errors are only to be reported to Chief Medical Office and Patient Safety (CMO& PS) department if the treatment error is associated with an SAE.

All instances of misuse or abuse must be documented in the adverse event (AE) CRF irrespective of the misuse/abuse being associated with an AE/SAE. In addition, all instances of misuse or abuse must be reported to Novartis Chief Medical Office and Patient Safety (CMO& PS). As such, instances of misuse or abuse are also to be reported using the SAE form/CRF. Table 9-1 summarizes the reporting requirements.

Table 9-1 Guidance for capturing study treatment errors

Treatment error type	Document in Dose Administration (DAR) CRF	Document in AE CRF	Complete SAE form/CRF
Unintentional study treatment error	Yes	Only if associated with an AE	Only if associated with an SAE
Misuse/Abuse	Yes	Yes	Yes, even if not associated with a SAE

For more information on AE and SAE definition and reporting requirements, please see Section 9.1 and Section 9.2, respectively.

9.6 Pregnancy reporting

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no guidelines on therapeutic recommendations in case of pregnancy are available. This study enrolls women who are considered to be of non-child-bearing potential or receiving highly effective methods of contraception thus pregnancy is not an expected outcome for any female study participant. However, in the case that a pregnancy in a female study participant should occur, please follow the below reporting guidelines.

To ensure patient safety, each pregnancy occurring after signing the informed consent must be **reported to Novartis within 24 hours** of learning of its occurrence. The pregnancy should be followed up to determine outcome, including spontaneous or voluntary termination, details of the birth, and the presence or absence of any birth defects, congenital abnormalities, or maternal and/or newborn complications.

Pregnancy must be recorded on the Pharmacovigilance Pregnancy Form and reported by the investigator to the local Novartis Chief Medical Office and Patient Safety (CMO& PS) Department. Pregnancy follow-up should be recorded on the same form and should include an assessment of the possible relationship to the study treatment.

Any SAE experienced during the pregnancy and unrelated to the pregnancy must be reported on a SAE form.

The Study Drug must be discontinued, though the patient may stay in the study, if she wishes to do so. All assessments that are considered as a risk during pregnancy must not be performed. The patient may continue all other protocol assessments.

Pregnancy outcomes should be collected for the female partners of any males who took study treatment in this study. Consent to report information regarding these pregnancy outcomes should be obtained from the mother.

9.7 Early phase safety monitoring

The Investigator will monitor adverse events in an ongoing manner and inform the Sponsor of any clinically relevant observations. Any required safety reviews will be made jointly between medically qualified personnel representing the Sponsor and Investigator. Such evaluations may occur verbally, but the outcome and key discussion points will be summarized in writing (e-mail) and made available to both Sponsor and all Investigator(s). Criteria pertaining to stopping the study/treatment or adapting the study design are presented above.

When two or more clinical site(s) are participating in the clinical study, the Sponsor will advise the Investigator(s) at all sites in writing (e-mail) (and by telephone if possible) of any new, clinically relevant safety information reported from another site during the conduct of the study in a timely manner.

10 Data review and database management

10.1 Site monitoring

Before study initiation, at a site initiation visit or at an investigator's meeting, a Novartis representative will review the protocol and data capture requirements (i.e. eCRFs) with the investigators and their staff. During the study Novartis employs several methods of ensuring protocol and GCP compliance and the quality/integrity of the sites' data. The monitor will visit the site to check the completeness of patient records, the accuracy of data capture / data entry, the adherence to the protocol and to Good Clinical Practice, the progress of enrollment, and ensure that Study Drug is being stored, dispensed, and accounted for according to specifications. Key study personnel must be available to assist the monitor during these visits.

The investigator must maintain source documents for each patient in the study, consisting of case and visit notes (hospital or clinic medical records) containing demographic and medical information, laboratory data, electrocardiograms, and the results of any other tests or assessments. All information on CRFs must be traceable to these source documents in the patient's file. The investigator must also keep the original informed consent form signed by the patient (a signed copy is given to the patient).

The investigator must give the monitor access to all relevant source documents to confirm their consistency with the data capture and/or data entry. Novartis monitoring standards require full verification for the presence of informed consent, adherence to the inclusion/exclusion criteria, documentation of SAEs, and the recording of data that will be used for all primary and safety variables. Additional checks of the consistency of the source data with the CRFs are performed according to the study-specific monitoring plan. No information in source documents about the identity of the patients will be disclosed.

10.2 Data collection

Designated investigator staff will enter the data required by the protocol into the Electronic Case Report Forms using fully validated software that conforms to 21 CFR Part 11 requirements. Designated investigator site staff will not be given access to the EDC system until they have been trained. Automatic validation programs check for data discrepancies and, by generating appropriate error messages, allow the data to be confirmed or corrected before transfer of the data to the CRO working on behalf of Novartis. The Investigator must certify that the data entered into the Electronic Case Report Forms are complete and accurate. After database lock, the investigator will receive copies of the patient data for archiving at the investigational site.

Data not requiring a separate written record will be defined in the Site Operations Manual and Assessment schedule (Section 8.1) and can be recorded directly on the CRFs. All other data captured for this study will have an external originating source (either written or electronic) with the CRF not being considered as source.

All data should be recorded, handled and stored in a way that allows its accurate reporting, interpretation and verification.

10.3 Database management and quality control

CRO working on behalf of Novartis review the data entered into the CRFs by investigational staff for completeness and accuracy and instruct the site personnel to make any required corrections or additions. Queries are sent to the investigational site using an electronic data query. Designated investigator site staff is required to respond to the query and confirm or correct the data. If the electronic query system is not used, a paper Data Query Form will be faxed to the site. Site personnel will complete and sign the faxed copy and fax it back to the data management team who will make the correction to the database. The signed copy of the Data Query Form is kept at the investigator site.

Concomitant medications entered into the database will be coded using the WHO Drug Reference List, which employs the Anatomical Therapeutic Chemical classification system. Medical history/current medical conditions and adverse events will be coded using the Medical dictionary for regulatory activities (MedDRA) terminology.

Laboratory samples will be processed centrally and the results will be sent electronically to Novartis (or a designated CRO).

Randomization codes and data about all Study Drug(s) dispensed to the patient and all dosage changes will be tracked using an Interactive Response Technology (IRT). The system will be supplied by a vendor, who will also manage the database. The database will be sent electronically to Novartis (or a designated CRO).

Each occurrence of a code break via IRT will be reported to the clinical team and monitor. The code break functionality will remain available until study shut down or upon request of Novartis.

The occurrence of relevant protocol deviations will be determined. After these actions have been completed and the database has been declared to be complete and accurate, it will be locked and the treatment codes will be unblinded and made available for data analysis. Any changes to the database after that time can only be made after written agreement by Novartis management.

10.4 Data Monitoring Committee

Not required.

10.5 Adjudication Committee

Not required.

11 Data analysis

The analysis will be conducted on all patient data at the time the trial ends. Any data analysis carried out independently by the investigator should be submitted to Novartis before publication or presentation.

11.1 Analysis sets

For all analysis sets, patients will be analyzed according to the study treatment(s) received.

The following are considered the definitions for the statistical analysis populations:

- Safety (or modified intent-to-treat, mITT) population: All patients that received at least one dose of Study Drug.
- Pharmacokinetics (PK) population: All patients in mITT that have at least one available valid (i.e. not flagged for exclusion) PK concentration measurement, and experienced no protocol deviations with relevant impact on PK data.
- Microbiological modified intent-to-treat (micro-mITT) population: All patients in mITT who have a baseline bacterial pathogen known to cause cIAI.
- Clinically evaluable (CE) population: All patients in mITT with cIAI confirmed by operative or percutaneous drainage findings, with sufficient information to determine clinical outcome at the TOC visit (EOS), and have no other protocol deviation with relevant impact on clinical data. Patients who received < 5 days of Study Drug due to clinical failure will NOT be excluded from the clinically evaluable population.

- Microbiologically evaluable (ME) population: All patients in micro-mITT who also have at least one clinically relevant pathogen susceptible to the study treatment isolated in the initial specimen collected from the intra-abdominal focus of infection.

11.2 Subject demographics and other baseline characteristics

All data for background and demographic variables will be listed by treatment arm and patient. Summary statistics will be provided by treatment.

Relevant medical history, current medical conditions, results of laboratory screens, drug tests and any other relevant information will be listed by treatment and patient.

11.3 Treatments

Data for study drug administration, rescue medication, and concomitant therapies will be listed by treatment and patient.

11.4 Analysis of the primary variable(s)

The primary aim of this study is to evaluate the LYS228 pharmacokinetic exposures for Pharmacokinetic/Pharmacodynamic (PK/PD) target attainment assessment and to evaluate the clinical response rate of LYS228 in combination with vancomycin and metronidazole in patients with complicated intra-abdominal infections (cIAI).

11.4.1 Primary Variable(s)

The primary PK variables are LYS228 plasma concentration and the following PK parameters:

- AUCtau, Cmax, Tmax, CL, Vss, T1/2, %fT > MIC (=2 ug/mL) in plasma on Day 5.

The primary clinical response variable is clinical success at 28 days after randomization (EOS visit). Clinical response is defined as follows:

- Clinical success is defined as resolution, or substantial improvement (i.e. reduction of severity of all baseline signs and symptoms and worsening of none) of all or most baseline signs and symptoms of cIAI infection without the need for additional antibiotic therapy other than any oral antibiotics given to complete treatment at home following discontinuation of Study Drug and no drainage or surgical reintervention required 96 hours after the start of Study Drug. Clinical success will be evaluated in the micro-mITT and CE populations.
- Clinical failure is defined as lack of response to study therapy as documented by persistence or worsening of baseline symptoms and/or signs of cIAI infection, such that alternative or additional antibiotic therapy or surgical interventions are required. Failure also includes the following: death related to the cIAI at any time, persisting or recurrent infection within the abdomen documented by findings from percutaneous or operative reintervention 96 hours after the start of Study Drug, relapse, and use of additional antibiotics for baseline or new cIAI. Patients that require replacement or reinsertion of percutaneous drain due to mechanical failure (obstruction of catheters, misplaced catheters) will not be considered clinical failures.

These primary variables will be assessed separately.

11.4.2 Statistical model, hypothesis, and method of analysis

Pharmacokinetics

Noncompartmental PK analysis will be performed on the LYS228 plasma concentration-time profiles and the following PK parameters characterizing the disposition of LYS228 will be generated using WinNonlin Phoenix (Version 6.4 or higher, Pharsight, Mountain View, CA):

- AUCtau, Cmax, Tmax, CL, Vss, T1/2, %fT>MIC.
- Additional PK parameters may be determined and compartmental PK modeling may be performed where appropriate.

LYS228 plasma concentration data will be listed by patient and visit/sampling time-point. Descriptive summary statistics will be provided by visit/sampling time-point. Descriptive summary statistics will include mean (arithmetic and geometric), SD, CV (arithmetic and geometric), median, minimum and maximum values. Graphical presentation will be used to display the trough concentrations versus the study visit days. Concentrations below the lower level of quantification (LLOQ) of the assay will be treated as zero in summary statistics. A geometric mean will not be reported if the dataset includes zero values.

PK parameters will be calculated as described above and will be listed by patient, and will be summarized by dose and renal category. Descriptive summary statistics will include mean (arithmetic and geometric), SD, CV (arithmetic and geometric), median, minimum and maximum values. An exception to this is Tmax where median, minimum and maximum will be presented.

Additionally, 95% confidence intervals will be computed for log-transformed primary PK parameters (AUCtau and Cmax). These intervals will be reported on the original exponential scale.

A population PK/PD analysis combining all data from this study and from other studies will be performed for target attainment analysis and will be reported separately.

Graphical presentation will be used to explore the relationship between the quartile exposure parameters and clinical response. Individual clinical response vs AUCtau, Cmax, and %fT>MIC will be explored, respectively.

Pharmacodynamics/clinical response

The proportion of patients who achieved clinical success or failure will be computed for each treatment and provided along with an 80% two-sided confidence interval based on the Clopper-Pearson method. The 80% confidence interval for the difference in clinical success between treatment s will be computed according to the Wilson score method without continuity correction.

Clinical success or failure will be evaluated at the end of Study Drug therapy (EOSD), at the end of all antibiotic therapy (EOAT), and at 28 days post-randomization (EOS).

Bayesian methods will be implemented based on the Beta-Binomial model to compare the clinical success rate in micro-mITT population at 28 days post-randomization. Informative

prior on LYS228 and control will be used as follows: Meta-analytic-predictive (MAP) informative prior (Spiegelhalter et al 2004, Neuenschwander et al 2010 and Schmidli et al 2014) for the clinical success rate in micro-mITT population on active control was obtained based on the evaluable historical data (FDA guidance, 2015): 0.67*Beta(172,38)+0.23*Beta(9,3)+0.1*Beta(1, 1), where median=0.81, sd=0.144, 95% CI=[0.25, 0.93] and the quantified information is equivalent to an effective sample size of 7.

An 80% posterior credible interval of the clinical success rate at EOS visit in the micro-mITT population will be provided for each treatment group as well as the treatment difference.

11.4.3 Handling of missing values/censoring/discontinuations

For PK analysis, all concentrations below the limit of quantification or missing data will be labeled as such in the concentration data listings. Concentrations below the lower limit of quantification (LLOQ) will be treated as zero in summary statistics and for the calculation of PK parameters.

Patients with missing or indeterminate outcome data will be classified as clinical failure in the micro-mITT population and will be excluded from the CE population.

11.4.4 Sensitivity analyses

Additional sensitivity analyses may be performed in which adjustments are made for factors that cannot be controlled at randomization.

Sensitivity analyses to explore the homogeneity of treatment effects across different centers, and to reflect the effect of the choice of the control group treatment on the treatment effect of LYS228, may be done if the sample sizes are adequate to do so. For the former analysis, a fixed effects model with treatment, center and treatment-by-center interaction as fixed effects will be used. For the latter analysis, centers using the same standard of care will be pooled in one group, and the analysis will be conducted by using a fixed effects model with treatment, center group, and treatment-by-center group as fixed effects.

11.5 Analysis of secondary variables

11.5.1 Efficacy / Pharmacodynamics

The proportion of patients who achieved microbiological success will be computed for each treatment and provided along with an 80% two-sided confidence interval based on the Clopper-Pearson method. The 80% confidence interval for the difference between treatment s will be computed according to the Wilson score method without continuity correction.

- Microbiological success is defined as documented microbiological eradication of all baseline intra-abdominal pathogens following at least 5 days of Study Drug, or presumed microbiological eradication is defined as a clinical success (see above). Microbiological success will be evaluated in micro-mITT and ME populations.

- Microbiological failure is defined as i) culture of the intended to treat baseline Gram-negative pathogen from a specimen obtained from the site of abdominal infection following at least 5 days of Study Drug, or ii) presumed microbiological failure if no cultures obtained and there is clinical failure.

Microbiological success will be evaluated at the EOSD, at the EOAT, and at 28 days post-randomization (EOS).

Subgroup summaries will also be provided by bacterial genetic characterization and in a subset of patients with cIAI caused by drug resistance mechanisms including CRE.

11.5.2 Safety

Vital signs

All vital signs data will be listed by treatment, patient, and visit/time and if ranges are available abnormalities will be flagged. Summary statistics will be provided by treatment and visit/time.

ECG evaluations

All ECG data will be listed by treatment, patient and visit/time, abnormalities will be flagged. Summary statistics will be provided by treatment and visit/time.

Clinical laboratory evaluations

All laboratory data will be listed by treatment, patient, and visit/time and if normal ranges are available abnormalities will be flagged. Summary statistics will be provided by treatment arm and visit/time.

Adverse events

All information obtained on adverse events will be displayed by treatment arm and patient.

The number and percentage of patients with adverse events will be tabulated by body system and preferred term with a breakdown by treatment. A patient with multiple adverse events within a body system is only counted once towards the total of this body system.

11.5.3 Pharmacokinetic - exposure relationships

Graphical presentation may be used to explore the relationship between the quartile exposure parameters AUCtau, Cmax, and %fT>MIC and clinical response. The analysis results will be reported in a separate document.

11.7 Sample size calculation

The sample size calculation takes into account the likely distribution of renal function and PK variability in the study population

In addition, with 40 patients on LYS228 vs. 20 patients on active control will produce a two-sided 80% frequentist CI for the difference in population proportions of (-0.13, 0.15) if the observed clinical success rates in the micro-mITT population for both treatment groups are 80%. For the same sample sizes of 40 vs 20, the two-sided 80% posterior credible interval for the difference in clinical success rate is (-0.059, 0.062) if the observed clinical success rates in micro-mITT population for both treatment groups are 80%.

11.8 Power for analysis of key secondary variables

Not applicable.

12 Ethical considerations

12.1 Regulatory and ethical compliance

This clinical study was designed and shall be implemented, executed and reported in accordance with the ICH Harmonized Tripartite Guidelines for Good Clinical Practice, with applicable local regulations (including European Directive 2001/20/EC, US Code of Federal Regulations Title 21, and Japanese Ministry of Health, Labor, and Welfare), and with the ethical principles laid down in the Declaration of Helsinki.

12.2 Responsibilities of the investigator and IRB/IEC

Before initiating a trial, the investigator/institution must obtain approval/favorable opinion from the Institutional Review Board/Independent Ethics Committee (IRB/IEC) for the trial protocol, written informed consent form, consent form updates, patient recruitment procedures (e.g. advertisements) and any other written information to be provided to patients. Prior to study start, the investigator is required to sign a protocol signature page confirming his/her agreement to conduct the study in accordance with these documents and all of the instructions and procedures found in this protocol and to give access to all relevant data and records to Novartis monitors, auditors, Novartis Quality Assurance representatives, designated agents of Novartis, IRBs/IECs, and regulatory authorities as required. If an inspection of the clinical site is requested by a regulatory authority, the investigator must inform Novartis immediately that this request has been made.

For multi-center trials, a Coordinating Investigator will be selected by Novartis by the time of Last Patient Last Visit to be a reviewer and signatory for the clinical study report.

12.3 Publication of study protocol and results

The key design elements of this protocol will be posted in a publicly accessible database such as clinicaltrials.gov. In addition, upon study completion and finalization of the study report the results of this trial will be either submitted for publication and/or posted in a publicly accessible database of clinical trial results.

12.4 Quality Control and Quality Assurance

Novartis maintains a robust Quality Management (QM) system that includes all activities involved in quality assurance and quality control, including the assignment of roles and responsibilities, the reporting of results, and the documentation of actions and escalation of issues identified during the review of quality metrics, incidents, audits and inspections.

Audits of investigator sites, vendors, and Novartis systems are performed or overseen by Novartis Pharma Auditing and Compliance Quality Assurance (or CRO working on behalf of Novartis), a group independent from those involved in conducting, monitoring or performing quality control of the clinical trial. The clinical audit process uses a knowledge/risk based approach.

Audits are conducted to assess GCP compliance with global and local regulatory requirements, protocols and internal SOPs, and are performed according to written Novartis processes.

13 Protocol adherence

This protocol defines the study objectives, the study procedures and the data to be collected on study participants. Additional assessments required to ensure safety of patients should be administered as deemed necessary on a case by case basis. Under no circumstances is an investigator allowed to collect additional data or conduct any additional procedures for any research related purpose involving any investigational drugs under the protocol.

Investigators ascertain they will apply due diligence to avoid protocol deviations. If an investigator feels a protocol deviation would improve the conduct of the study this must be considered a protocol amendment, and unless such an amendment is agreed upon by Novartis and approved by the IRB/IEC and health authorities, where required, it cannot be implemented.

13.1 Protocol Amendments

Any change to the protocol can only be made in a written protocol amendment that must be approved by Novartis, Health Authorities where required, and the IRB/IEC prior to implementation.

Amendments that are intended to eliminate an apparent immediate hazard to patients may be implemented immediately, provided the Health Authorities are subsequently notified by protocol amendment and the reviewing IRB/IEC is notified.

Notwithstanding the need for approval of formal protocol amendments, the investigator is expected to take any immediate action required for the safety of any patient included in this study, even if this action represents a deviation from the protocol. In such cases, the reporting requirements identified in Section 9 (Safety Monitoring) must be followed and the Study Lead informed.

14 References

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15 Appendix 1: Liver Event Definitions and Follow-up Requirements

Table 15-1 Liver Event Definitions

Definition	Thresholds
Potential Hy's law cases	• ALT or AST > 3 × ULN and TBL > 2 × ULN without initial increase in ALP to > 2 × ULN
ALT or AST elevation with coagulopathy	• ALT or AST > 3 × ULN and INR > 1.5 (in the absence of anticoagulation)
ALT or AST elevation accompanied by symptoms	• ALT or AST > 3 × ULN accompanied by (general) malaise, fatigue, abdominal pain, nausea, or vomiting, or rash, or eosinophilia
Isolated ALT or AST elevation	• ALT or AST > 8 × ULN • 5 x ULN < ALT/AST ≤ 8 x ULN • 3 x ULN < ALT/AST ≤ 5 x ULN
Isolated ALP elevation	• ALP > 2 × ULN (in the absence of known bone pathology)
Others	• Any clinical event of jaundice (or equivalent term) • Any adverse event potentially indicative of liver toxicity

Table 15-2 Actions required for Liver Events

Criteria	Actions required
Potential Hy's Law case ALT or AST elevation with coagulopathy ALT or AST elevation accompanied by symptoms Isolated ALT or AST elevation > 8 × ULN Jaundice	• Discontinue the study treatment immediately • Hospitalize, if clinically appropriate • Establish causality • Complete CRFs per liver event guidance*
Isolated ALT or AST elevation > 5 to ≤ 8 × ULN	• If confirmed, consider interruption or discontinuation of Study Drug • If elevation persists for more than 2 weeks, discontinue the Study Drug • Establish causality • Complete CRFs per liver event guidance*
Isolated ALT or AST elevation > 3 to ≤ 5 × ULN (patient is asymptomatic)	• Monitor liver chemistry tests two or three times weekly
Isolated ALP elevation	• Repeat liver chemistry tests within 48-72 hours • If elevation is confirmed, measure fractionated ALP; if >50% is of liver origin, establish hepatic causality • Complete CRFs per liver event guidance*
Any AE potentially indicative of liver toxicity	• Consider study treatment interruption or discontinuation • Hospitalize if clinically appropriate • Complete CRFs per liver event guidance*

*Liver event guidance for CRF completion is available in the Site Operations Manual

Table 15-3 Exclusion of underlying liver disease

Disease	Assessment
Hepatitis A, B, C, E	• IgM anti-HAV; HBSAg, IgM anti-HBc, HBV DNA; anti-HCV, HCV RNA, IgM & IgG anti-HEV, HEV RNA
CMV, HSV, EBV infection	• IgM & IgG anti-CMV, IgM & IgG anti-HSV; IgM & IgG anti-EBV
Autoimmune hepatitis	• ANA & ASMA titers, total IgM, IgG, IgE, IgA
Alcoholic hepatitis	• Ethanol history, GGT, MCV, CD-transferrin
Nonalcoholic steatohepatitis	• Ultrasound or MRI
Hypoxic/ischemic hepatopathy	• Medical history: acute or chronic CHF, hypotension, hypoxia, hepatic venous occlusion. Ultrasound or MRI.
Biliary tract disease	• Ultrasound or MRI, ERCP as appropriate.
Wilson disease	• Caeruloplasmin
Hemochromatosis	• Ferritin, transferrin
Alpha-1-antitrypsin deficiency	• Alpha-1-antitrypsin

16 Appendix 2: Specific Renal Alert Criteria and Actions

Table 16-1 Specific Renal Alert Criteria and Actions

Criteria	Action required
Serum creatinine (sCr) increase 25 – 49% compared to baseline	• Consider causes and possible interventions • Follow up within 2-5 days
Serum creatinine increase > 50%	• Consider causes and possible interventions • Repeat assessment within 24-48h if possible • Consider drug interruption or discontinuation unless other causes are diagnosed and corrected • Consider hospitalization and specialized treatment
Protein-creatinine or albumin-creatinine ratio increase ≥ 2-fold or new onset dipstick proteinuria ≥ 1+ or Albumin-creatinine ratio (ACR) ≥ 30 mg/g or ≥ 3 mg/mmol; or Protein-creatinine ratio (PCR)≥ 150 mg/g or >15 mg/mmol	• Consider causes and possible interventions • Assess serum albumin & serum protein • Repeat assessment to confirm • Consider drug interruption or discontinuation unless other causes are diagnosed and corrected
New onset glucosuria on urine dipstick (unless related to concomitant treatment, diabetes)	Assess & document: • Blood glucose (fasting) • Serum creatinine • Urine albumin-creatinine ratio
New hematuria on dipstick	Assess & document: • Urine sediment microscopy • Assess sCr and urine albumin-creatinine ratio • Exclude infection, trauma, bleeding from the distal urinary tract/bladder, menstruation • Consider bleeding disorder

Additional specialized assessments are available to assess renal function or renal pathology. (Note: In exceptional cases when a nephrologist considers a renal biopsy, it is strongly recommended to make specimen slides available for evaluation by Novartis to potentially identify project-wide patterns of nephrotoxicity.)

Whenever a renal event is identified, a detailed patient history and examination are indicated to identify, document and potentially eliminate risk factors that may have initiated or contributed to the event:

- Blood pressure assessment (after 5 min rest, with an appropriate cuff size)
- Signs and symptoms such as fever, headache, shortness of breath, back or abdominal pain, dysuria, hematuria, dependent or periorbital edema
- Changes in blood pressure, body weight, fluid intake, voiding pattern, or urine output
- Concomitant events or procedures such as trauma, surgical procedures, cardiac or hepatic failure, contrast media or other known nephrotoxin administration, or other potential causes of renal dysfunction, e.g., dehydration, hemorrhage, tumor lysis

Table 16-2 Follow-up of renal events

Action	Follow up
Assess*, document and record in the Case Report Form (CRF) or via electronic data load. Review and record possible contributing factors to the renal event (co-medications, other co-morbid conditions) and additional diagnostic procedures (MRI etc) in the CRF.	• Urine dipstick and sediment microscopy • Blood pressure and body weight • Serum creatinine, electrolytes (sodium, potassium, phosphate, calcium), bicarbonate and uric acid • Urine output
Monitor patient regularly (frequency at investigator's discretion) until:	• Event resolution: (sCr within 10% of baseline or protein-creatinine ratio within 50% of baseline) or • Event stabilization: sCr level with ±10% variability over last 6 months or protein-creatinine ratio stabilization at a new level with ±50% variability over last 6 months.

* Urine osmolality: in the absence of diuretics or chronic kidney disease this can be a very sensitive metric for integrated kidney function that requires excellent tubular function. A high urinary osmolality in the setting of an increase in sCr will point toward a "pre-renal" cause rather than tubular toxicity.