Cadazolid/ACT-179811

Protocol AC-061A301

A multi-center, randomized, double-blind study to compare the efficacy and safety of cadazolid versus vancomycin in subjects with *Clostridium difficile*-associated diarrhea (CDAD)

NCT Number	NCT01987895
Document Version, Date	Final Version 3, 22 October 2015
Document History:	
- Original Version	29 August 2013
- Amendment 1	11 December 2014
- Amendment 2	22 October 2015

Main reason for Amendment 1

The main analysis of the primary endpoint Clinical Cure will be conducted on two co-primary analysis sets (i.e., modified intent-to-treat [mITT] analysis set and Per-protocol analysis set [PPS]) instead of sequential analysis.

Further changes include the addition of an emerging hypervirulent *Clostridium difficile* strain, the addition of endpoints related to susceptibility testing of *C. difficile* and vancomycin-resistant enterococci, and general clarifications of eligibility criteria and statistical analyses including a modification to the definition of recurrence for analyses of secondary variable sustained cure rate.

Main reason for Amendment 2

To remove the interim analysis originally planned after the randomization of 67% of the subjects.

SPONSOR CONTACT DETAILS

SPONSOR	ACTELION Pharmaceuticals Ltd Gewerbestrasse 16 CH-4123 Allschwil Switzerland
Clinical Trial Physician	[redacted]

MEDICAL HOTLINE
Toll phone number: +41 61 227 05 63
Site-specific toll telephone numbers and toll-free numbers for the Medical Hotline can be found in the Investigator Site File.

ACTELION CONTRIBUTORS TO THE PROTOCOL

Clinical Trial Physician	[redacted]
Clinical Science Program Head	[redacted]
Clinical Trial Scientist	[redacted]
Trial Statistician	[redacted]
Clinical Trial Pharmacologist	[redacted]
Drug Safety Physician	[redacted]

COORDINATING INVESTIGATOR

Name / Title	Address

CONTRACT RESEARCH ORGANIZATIONS INFORMATION

CENTRAL LABORATORY	[redacted]
CENTRAL RANDOMIZATION	
ECG reading (some sites)	
Independent Statistical Data Analysis Center	
CONTRACT RESEARCH ORGANIZATIONS (CRO)	

A list of site-specific contact details for CROs can be found in the Investigator Site File.

SIGNATURE PAGE FOR ACTELION PHARMACEUTICALS LTD

Hereinafter called Actelion

Drug name / number

Cadazolid / ACT-179811

Indication

Clostridium difficile-associated diarrhea

Protocol number, study title

AC-061A301, a multi-center, randomized, double-blind study to compare the efficacy and safety of cadazolid versus vancomycin in subjects with *Clostridium difficile*-associated diarrhea (CDAD).

I approve the design of this study.

TITLE	NAME	DATE	SIGNATURE
Clinical Trial Physician	[redacted]	[redacted]	
Clinical Trial Statistician	[redacted]	[redacted]	

INVESTIGATOR SIGNATURE PAGE

Drug name / number

Cadazolid / ACT-179811

Indication

Clostridium difficile-associated diarrhea

Protocol number, study title

AC-061A301, a multi-center, randomized, double-blind study to compare the efficacy and safety of cadazolid versus vancomycin in subjects with *Clostridium difficile*-associated diarrhea (CDAD).

I agree to the terms and conditions relating to this study as defined in this protocol, the Case Report Form (CRF), and any other protocol-related documents. I fully understand that any changes instituted by the investigator(s) without previous agreement with the Sponsor would constitute a violation of the protocol, including any ancillary studies or procedures performed on study subjects (other than those procedures necessary for the well-being of the subjects).

I agree to conduct this study in accordance with the Declaration of Helsinki principles, International Conference on Harmonisation (ICH) Good Clinical Practice (GCP) guidelines, and applicable regulations and laws. In particular, I will obtain approval by an Institutional Review Board or Independent Ethics Committee (IRB/IEC) prior to study start and signed informed consent from all subjects included in this study. If an amendment to the protocol is necessary, I will obtain approval by an IRB/IEC and ensure approval by regulatory authorities (if applicable) have been obtained before the implementation of changes described in the amendment. In addition, I will allow direct access to source documents and study facilities to Sponsor representative(s), particularly monitor(s) and auditor(s), and agree to inspection by regulatory authorities or IRB/IEC representatives. I will ensure that the study drug(s) supplied by the Sponsor are being used only as described in this protocol. Furthermore, I confirm herewith that the Sponsor is allowed to enter and utilize my professional contact details and function in an electronic database for internal purposes and for submission to Health Authorities worldwide.

	Country	Site number	Town	Date	Signature
Site Principal Investigator				_____	_____

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

LIST OF ABBREVIATIONS AND ACRONYMS

AE	Adverse event
ALT	Alanine aminotransferase
ANCOVA	Analysis of covariance
aPTT	Activated partial thromboplastin time
AST	Aspartate aminotransferase
ATC	Anatomic therapeutic chemical
b.i.d.	*bis in diem*, twice a day
BM	Bowel movement
BMI	Body mass index
BP	Blood pressure
bpm	Beats per minute
CCR	Clinical cure rate
CCRc	Clinical cure rate for cadazolid
CCRv	Clinical cure rate for vancomycin
CDAD	*Clostridium difficile*-associated diarrhea
CDI	*Clostridium difficile* infection
CI	Confidence interval
CLSI	Clinical and Laboratory Standards Institute
C_{max}	Maximum plasma concentration
CRA	Clinical research associate
CRF	Case report form
CRO	Contract research organization
CTCAE	Common Terminology Criteria for Adverse Events
CV	Curriculum vitae
DBP	Diastolic blood pressure
ECCR	Early clinical cure rate
ECG	Electrocardiogram
EIA	Enzyme immunoassay
EMA	European Medicines Agency
EOS	End-of-Study
EOT	End-of-Treatment
[redacted]	[redacted]
ES	Re-treatment extension with cadazolid set
ESCMID	European Society of Clinical Microbiology and Infectious Diseases
ETR	Early treatment response
FAS	Full analysis set
FDA	Food and Drug Administration
FMT	Fecal microbiota transplant

GCP	Good clinical practice
GDH	Glutamate dehydrogenase
h	Hour
HR	Heart rate
HVAS	Hypervirulent analysis set
IB	Investigator's brochure
ICF	Informed consent form
ICH	International Conference on Harmonisation
ICR	Investigator's judgment of clinical response
ICRR	Investigator's judgment of clinical response rate
ICU	Intensive care unit
IDMC	Independent Data Monitoring Committee
IDSA	Infectious Diseases Society of America
IEC	Independent Ethics Committee
INR	International normalized ratio
IRB	Institutional Review Board
ISDAC	Independent Statistical Data Analysis Center
ISF	Investigator site file
ISR	Investigator's judgment of sustained response
ISRR	Investigator's judgment of sustained response rate
IVRS	Interactive voice response system
LC-MS/MS	Liquid chromatography-tandem mass spectrometry
LOQ	Limit of quantification
MAD	Multiple-ascending dose
MedDRA	Medical Dictionary for Regulatory Activities
MIC	Minimal inhibitory concentration
mITT	Modified intent-to-treat
MTF	Metronidazole treatment failure
NBM	Normalization of bowel movements
NBMR	Normalization of bowel movement rate
NDA	New drug application
NED	New episode of diarrhea
NI	Non-inferiority
PCR	Polymerase chain reaction
PI	Package insert
PKS	Pharmacokinetic analysis set
PP	Per-protocol
PPI	Proton pump inhibitor
PPS	Per-protocol analysis set
PRO	Patient reported outcomes

PROAS	PRO analysis set
q.i.d.	Four times a day
QTc	QT interval corrected for heart rate
QTcB	QT corrected with Bazett's formula
QTcF	QT corrected with Fridericia's formula
REA	Restriction endonuclease assay
ROD	Resolution of diarrhea
RR	Recurrence rate
SAE	Serious adverse event
SAP	Statistical analysis plan
SBP	Systolic blood pressure
SI	Standardized International
SCR	Sustained cure rate
SCRAS	Screened analysis set
SCRc	Sustained cure rate cadazolid
SCRv	Sustained cure rate vancomycin
SD	Standard deviation
SE	Standard error
SHEA	Society for Healthcare Epidemiology of America
SOC	System organ class
SOP	Standard operating procedure
SS	Safety set
SUSAR	Suspected unexpected serious adverse reaction
TEAE	Treatment-emergent adverse event
TOC	Test-of-cure
UBM	Unformed bowel movement
ULN	Upper limit of the normal range
US	United States
USPI	United States package insert
VRE	Vancomycin-resistant enterococci
WHO	World Health Organization
WPAI	Work productivity and activity impairment

PROTOCOL SYNOPSIS AC-061A301

TITLE	AC-061A301, A multi-center, randomized, double-blind study to compare the efficacy and safety of cadazolid versus vancomycin in subjects with *Clostridium difficile*-associated diarrhea (CDAD).
MAIN OBJECTIVES	**Primary objective** To determine whether the clinical response after 10-day oral administration of cadazolid is non-inferior to oral vancomycin in subjects with CDAD. **Secondary objectives** To determine whether oral administration of cadazolid for 10 days is superior to oral vancomycin in the sustained clinical response of subjects with CDAD. To determine whether the resolution of diarrhea (ROD) is more rapid with oral administration of cadazolid compared to vancomycin. To determine whether CDAD symptoms, as reported by the subject, show larger improvements from baseline with oral administration of cadazolid compared to vancomycin. **Meta-analysis objective** To determine whether oral administration of cadazolid for 10 days is superior to oral vancomycin in the sustained clinical response of subjects with CDAD due to hypervirulent strains. **Safety objective** To determine safety and tolerability of an oral administration of cadazolid compared to vancomycin. Exploratory objectives are described in the core protocol.
DESIGN See Section 3	Prospective, multi-center, double-blind, double-dummy, randomized, parallel group, active controlled Phase 3 study
PERIODS See Section 3	**Screening Period** starts with the signature of the informed consent form (ICF) and ends with subject randomization (within 48 hours of the signature of the ICF). **Treatment Period** starts after randomization, with the first dose of study drug, and ends on the day of the last dose of study drug (EOT) 10 days later.

	Follow-up Period starts after the last dose of study drug and ends approximately 30 days after the last dose of study drug (Visit 5). **Re-treatment extension with cadazolid**: Subjects who experience a recurrence and provide consent may enter a re-treatment extension with cadazolid consisting of a 10-day treatment of cadazolid followed by an approximately 30-day follow-up period. Subject participation in the study will be up to 44 days; up to 88 days for subjects participating in the re-treatment extension with cadazolid.
PLANNED DURATION	Approximately 40 months.
SITES / COUNTRIES	Approximately 100 sites in approximately 20 countries.
SUBJECTS / GROUPS	Approximately 630 subjects will be randomized to cadazolid or vancomycin by Interactive Voice Response System (IVRS), ratio 1:1, stratified by first occurrence or first recurrence and by site.
KEY INCLUSION CRITERIA See Section 4.2	Signed Informed Consent. Male or female ≥ 18 years of age. Females of childbearing potential must agree to use an adequate and reliable method of contraception. Subject with a diagnosis of mild-moderate or severe CDAD (first occurrence or first recurrence within 3 months) with: Diarrhea: a change in bowel habits with > 3 unformed bowel movements (UBMs) within 24 hours prior to randomization, **AND** Positive *C. difficile* GDH and toxin test (by enzyme immunoassay approved by the sponsor) on the same stool sample produced within 72 hours prior to randomization.
KEY EXCLUSION CRITERIA See Section 4.3	More than one previous episode of CDAD in the 3-month period prior to randomization. Evidence of life-threatening or fulminant CDAD. Likelihood of death within 72 hours from any cause. History of inflammatory colitides, chronic abdominal pain, or chronic diarrhea, or known positive diagnostic test for enteropathogens.

	Inability to take oral medication, or subjects with feeding tubes (i.e., when study drug would have to be given by a feeding tube). Antimicrobial treatment active against CDAD administered for > 24 hours except for metronidazole treatment failures (MTF). Planned treatment with forbidden concomitant medications. Fecal microbiota transplant (FMT), immunoglobulin therapy, and investigational drug to prevent or treat CDAD within 1 month prior to randomization Monoclonal antibodies against *C. difficile* within 6 months prior to randomization. Previous vaccination against *C. difficile*. Previous participation in a clinical trial with cadazolid. Known hypersensitivity or contraindication to study drugs, oxazolidinones, or quinolones. Females who are breastfeeding. Investigational site staff members or relatives, and Actelion employees. Unable or unwilling to comply with all protocol requirements.
STUDY TREATMENTS See Section 5.1	**Investigational drug** Cadazolid 250 mg or matching placebo granules for oral suspension twice daily with or without food. **Comparator** Oral vancomycin 125 mg or matching placebo capsules 4 times daily with or without food.
CONCOMITANT MEDICATIONS See Section 5.2	**Prohibited medications** Antimicrobial treatment active against CDAD and FMT up to Visit 5 unless provided for clinical failure or recurrence. Other medication active against CDAD (e.g., cholestyramine, probiotics) up to Visit 5. Initiation of treatment with opiates or change in dose or regimen resulting in an increased opiate effect up to 2 days after EOT. Anti-peristaltic medications, kaolin, pectin and charcoal containing anti-diarrheals up to Visit 5.

ENDPOINTS See Section 6.1	**Primary endpoint** Clinical Cure defined as: • Resolution of Diarrhea (ROD: ≤ 3 UBMs per day for at least 2 consecutive days) on study treatment, maintained for 2 days after EOT **AND** • No additional antimicrobial treatment active against CDAD or FMT between first dose and 2 days after EOT (inclusive). **Secondary endpoints** Sustained Cure defined as: • Clinical Cure **AND** • No Recurrence Recurrence is defined for subjects with Clinical Cure as: • New episode of diarrhea (NED: > 3 UBMs within 1 day) occurring between 3 days after EOT and Visit 5 **AND** • Positive *C. difficile* GDH and toxin stool test on the same stool sample **AND** • Antimicrobial treatment active against CDAD or FMT started between 3 days after EOT and Visit 5 (or participation in the re-treatment extension with cadazolid). Time to ROD, defined as: • The time (h) elapsed between the first dose of study drug and the time when ROD is considered achieved. Absolute change from baseline in CDAD DaySyms Patient Reported Outcomes (PRO). CDAD DaySyms total daily score change from baseline up to Day 12. **Meta-analysis endpoint** • Sustained Cure (as defined above) in hypervirulent strains (currently defined as Strains 027, 078, and 244). Additional endpoints are described in the core protocol.

ASSESSMENTS	Refer to the schedule of assessments in Table 1.
STATISTICAL METHODOLOGY See Section 11	**Sample Size**: Assuming a Clinical Cure Rate (CCR) of 85% in the Per-protocol analysis set (PPS) for cadazolid and vancomycin treatment groups, a power of 90%, a fixed type I error of 2.5% (one-sided), and a non-inferiority (NI) margin of 10%, a total sample size of 536 (268 in each treatment group) evaluable subjects is required in the PPS. Taking into account approximately 15% of subjects not qualifying for the per-protocol (PP) population, approximately 630 subjects will be randomized into the trial. As approximately 95% of the randomized subjects are assumed to qualify for the modified intent-to-treat (mITT) analysis set (598 subjects), the power for demonstrating non-inferiority in clinical cure on this population is higher than 90%. **Main efficacy analysis.** Clinical Cure Rate (CCR, %) is the variable to be analyzed for the primary endpoint. The aim is to demonstrate that the CCR for cadazolid (CCRc) is not inferior to the CCR for vancomycin (CCRv), accounting for a NI margin of 10%. NI in CCR of cadazolid against vancomycin will be assessed based on the difference between proportions in CCRc and CCRv. The null hypothesis is: $H_0^{(1)}$: CCRc − CCRv ≤ −10% versus the alternative hypothesis: $H_1^{(1)}$: CCRc − CCRv > −10%. The main analysis will be on the mITT analysis set and on the PPS. NI is claimed if it is demonstrated on both analysis sets, by a one-sided 0.025 significance level, using the lower bound of the two-sided 95% confidence interval (CI) of the difference between proportions. **Secondary efficacy analysis:** If NI for CCR is demonstrated in both analysis sets, mITT and PPS, the superiority of cadazolid on the secondary endpoint Sustained Cure Rate (SCR) will be tested on the mITT analysis set at the two-sided alpha 0.05. If the lower limit of the two-sided 95% CI is greater than zero, the statistical superiority of cadazolid versus vancomycin in SCR will be established. After this is

	demonstrated, the superiority in time to ROD (shorter median time) will be tested. If demonstrated, then the superiority in the CDAD DaySyms scores changes from baseline will be tested. This full hierarchical testing strategy is used to control the experimentwise α level at 0.05. **Meta-analysis:** A meta-analysis of this trial and the similar Phase 3 trial AC-061A302 will be utilized for the assessment of cadazolid superiority in the Sustained Cure of subjects with CDAD due to hypervirulent strains. **Safety analysis**: Treatment-emergent adverse events and serious adverse events will be summarized per treatment arm and overall, by frequency, intensity and relationship to study medication. Other safety variables, such as laboratory assessments, vital signs and ECGs will be summarized.
STUDY COMMITTEES	Steering Committee, Independent Data Monitoring Committee
SUB-STUDIES	As part of this protocol, at selected sites and in subjects who consent to participate, the plasma concentrations of cadazolid will be analyzed. In addition, the following sub-studies, independent from the core study protocol and at selected sites in subjects who consent to participate (through an informed consent separate from the main protocol informed consent), will be performed according to stand-alone protocols: • Psychometric validation of CDAD DaySyms PRO • Gut microbiome assessment and fecal cadazolid concentrations.

Table 1 Visit and assessment schedule

See Section 7 for complete information.

PERIODS	**Name**	**SCREENING**	**TREATMENT**			**FOLLOW-UP**			**-**
	Duration	**Up to 48 hours**	**10 to 11 days**			**28 to 32 days**			**-**
VISIT	**Number**	1		2	3	4	4.a, 4.b,…	5	U1, U2, …
	Name	**Screening**	**Randomization**				**NED**		**Unscheduled**
EVALUATION	**Time**	Day –2 to Day 1 (within 48 h)	Day 1	Day 5 or Day 6 *On site or by phone*	Day 8 to Day 10 *incl. Premature Disc.*	*EOT + 2 to 4 days On site or by phone*	*If applicable, in case of NED* [5, 6]	*EOT + 28 to 32 days or Study Withdrawal*	*If applicable*
Informed consent		X							
Fecal sampling for microbiology		X			X		X		
***C. difficile* GDH and toxin test**		within 72 h (MTF: 48h)					X		
Demographics		X							
Medical history		X							X
Concomitant medications		X		X	X	X	X	X	X
Physical examination		X			X		X	X	X
Body weight/height		X[1]			X			X	X
Vital signs / Body temperature		X[1]			X		X	X	X
Hematology/coagulation/chemistry		X [2]			X		X	X	X
12-lead ECG		X			X			X	X
Randomization			X						
Study Drug			X[3]	and Study Drug Journal completion					
Stool Diary			←	———	———	———	———	→	
CDAD DaySyms PRO Diary		←	———	———	———	→			
Interview (face to face or by phone)			Daily[7]				Twice weekly		
Investigator assessment				X		X		X	
Pharmacokinetic sample *Sub-study*					X				
WPAI:CDAD		X			X			X	
AEs/SAEs [4]		X	X	X	X	X	X	X	X

1. Height collected at Visit 1 only. **2.** Includes pregnancy test for women of childbearing potential. **3.** After randomization. **4.** All AEs and SAEs are reported after the first dose of study drug. Between informed consent and first dose of study drug, only SAEs are reported. **5.** Subject may enter re-treatment extension with cadazolid in case of recurrence [Table 3]. **6.** All assessments must be performed in the case of a recurrence. **7.** Interviews start from Day 2.

AE = adverse event, *C-difficile* = *Clostridium difficile*, CDAD = *Clostridium difficile*-associated diarrhea, ECG = electrocardiogram, EOT = End-of-Treatment, GDH = glutamate dehydrogenase, MTF = metronidazole treatment failure, NED = new episode of diarrhea, PRO = patient reported outcomes, SAE = serious adverse event, WPAI = work productivity and activity impairment.

CORE PROTOCOL

1 BACKGROUND AND RATIONALE

1.1 Indication

1.1.1 Introduction

Clostridium difficile-associated diarrhea (CDAD) is an infectious disease of the gastrointestinal tract. CDAD usually occurs in patients with a history of antibiotic use that allows *C. difficile* to grow and elaborate virulent toxins that can cause intestinal inflammation and diarrhea.

C. difficile is a spore-forming Gram-positive anaerobic bacillus present throughout the environment. Acid-resistant spores are ingested and passed through the stomach to germinate in the small bowel and eventually colonize the colon where they can be a harmless component of normal gut flora. Some strains of *C. difficile* elaborate toxins, termed A and B, that cause disease by invading epithelial cells, altering their cytoskeleton and resulting in a disruption of the epithelial barrier. These toxins then invade the intestinal mucosa where they also function as potent immunostimulatory molecules promoting inflammation [Poutanen 2004, Kelly 2008]. The resulting *C. difficile* infection (CDI) usually manifests as diarrhea but can cause a range of clinical symptoms with some patients only experiencing a transient nuisance diarrhea, while others rapidly progress to frequent loose stools, abdominal pain with fever and marked leukocytosis.

The typical patient with CDAD is in his or her 60s, often has comorbid illnesses, has received antibiotics within the past 2 months [Poutanen 2004, Pachecho 2013], and clinically presents with a broad spectrum of severity. CDAD may result in only a mild diarrhea lasting a few days that requires no treatment, to the more protracted severe disease with extensive colonic inflammation, associated characteristic pseudomembranous colitis and signs of systemic toxicity [Dallal 2002, Voelker 2010]. Risk factors for severe disease include age greater than 65, comorbid illnesses, concomitant acid suppression therapy, low antibody response, and the need to prolong inciting antibiotic therapy [Bauer 2009, Louie 2013b]. Although risk factors for severe disease have been reported, there is no consensus on a specific CDAD severity index. Severe disease is often defined based on fever (temperature > 38.5 °C), leukocytosis (> 15,000 cells/mm^3), or increase in serum creatinine (1.5 times above the premorbid level) [Bauer 2009, Cohen 2010]. Pseudomembranes on endoscopy and imaging criteria are sometimes used to define severe disease but information on these criteria are not always available. Subjects not meeting these criteria are classified as having mild–moderate CDI. When CDAD becomes fulminant and unresponsive to medical therapy, a life-saving emergent colectomy is required but can be associated with mortality as high as 40% [Bhangu 2012].

1.1.2 Epidemiology

CDAD incidence has been increasing in recent years and disease severity and mortality has also been increasing, in part due to the emergence of the 027/BI/NAP1 hypervirulent strain. A recent review suggests that on an annual basis in the United States there are 333,000 cases of CDAD, with 15,000 to 20,000 deaths at a cost of $3.2 billion [Dubberke 2010]. In Europe a survey of 34 countries revealed the incidence rate of CDAD to be 4.1 per 10,000 patient days per hospital [Bauer 2011]. The increased frequency and severity of CDAD in Western countries has been attributed to wider antibiotic use in an ever-growing elderly population that is living longer with multiple comorbid illnesses and immunosenescence, such that CDAD has now surpassed methicillin-resistant *Staphylococcus aureus* rates of infection in some community hospitals [Miller 2011]. Furthermore, severe disease with its associated mortality is also on the rise and thought to be related to the increased global spread of a virulent strain (genotyped as 027/BI/NAP1) that is capable of elaborating 20-fold more toxin than other toxigenic strains [Miao He 2012, Loo 2005].

In addition to the morbidity and mortality of the acute episode of CDAD, once cured 25–30% of patients will experience recurrent CDAD within 1 month of treatment, with either the same strain of *C. difficile* (relapse) or a different strain (re-infection). Recurrence rates can range as high as 65%, particularly in older patients and those continuing to receive concomitant antibiotics [Garey 2008, Louie 2013b, Hu 2009].

1.1.3 Current and future therapies

Several factors, including the 027/BI/NAP1 hypervirulent strain and the recurrence rate, have contributed to the overall morbidity and mortality of CDAD and have resulted in the re-emergence of *C. difficile* as a major global health problem. New drug therapy that reduces recurrence rates, or improves outcomes for patients infected with the hypervirulent strain or those with severe disease, remains a significant unmet medical need.

Discontinuation of concurrent antibiotics may allow the normal colonic flora to regenerate and may prevent further *C. difficile* growth. However, discontinuing antibiotics often does not resolve CDAD, and many patients are unable to discontinue antibiotics for an ongoing intercurrent infection.

Approved in 1986, oral vancomycin remains the mainstay of therapy, alongside metronidazole. Vancomycin has been proven efficacious in mild–moderate disease in the setting of clinical trials and is approved in many countries for the treatment of CDAD without any qualifier on the severity of disease. It is generally not used in the initial episode of mild–moderate disease. Current standard of care for mild–moderate disease in patients with an initial episode of CDAD remains metronidazole [Cohen 2010,

Bauer 2009]. Randomized data from the control arms of the recent tolevamer trials also suggested lower clinical response rates in metronidazole subjects compared to vancomycin subjects (72.7% versus 81.1%; odds ratio [95% confidence interval (CI)]: 1.681 [1.114, 2.537], p = 0.0134) [Johnson 2012]. For severe disease in the initial episode or in recurrences, vancomycin at the dose of 125 mg 4 times daily is recommended therapy by the Society for Healthcare Epidemiology of America / Infectious Diseases Society of America (SHEA/IDSA) and European Society of Clinical Microbiology and Infectious Diseases (ESCMID) guidelines. It is also recommended to treat patients with multiple recurrences with an extended course of vancomycin, often with a tapering or pulsed regimen.

Fidaxomicin has proven to be comparable to vancomycin with regard to cure rates, and importantly, has demonstrated higher Sustained Cure Rate (SCR) in 2 randomized double-blind trials [Louie 2011, Cornely 2012]. The decrease in recurrence rate is thought to be due to the reduced attenuation of colonic flora with fidaxomicin treatment compared with vancomycin treatment [Tannock 2010]. However, fidaxomicin's SCR against the virulent 027/BI/NAP1 strain was not better than vancomycin.

1.2 Study drugs

1.2.1 Cadazolid

This section is a brief summary of available data about cadazolid that is relevant to the study. For more detailed information, please see the Investigator's Brochure (IB) [Cadazolid IB].

Cadazolid is a novel antibiotic with bactericidal activity against *C. difficile*, including hypervirulent and moxifloxacin-resistant strains such as 027/BI/NAP1.

Minimal inhibitory concentrations (MICs) of cadazolid against clinical isolates of *C. difficile* range from 0.06 to 0.5 μg/mL. Cadazolid retains its activity against *C. difficile* over the pH range 6.2–7.5, which is found at relevant segments of the affected gut. Cadazolid is a potent inhibitor of bacterial protein synthesis and strongly inhibits toxin synthesis in cultures of toxigenic *C. difficile* strains in a manner superior to vancomycin and metronidazole. In contrast to vancomycin, *in vitro* experiments demonstrate that cadazolid prevents sporulation in vegetative cells at sub-MIC concentrations.

Two Phase 1 studies with cadazolid were conducted in healthy subjects: a single-ascending dose study (AC-061-101) and a multiple-ascending dose (MAD) study (AC-061-102). In these studies, 48 healthy male subjects were exposed to cadazolid at single oral doses of 30–3000 mg or multiple doses of 300–3000 mg twice daily for 10 days. All oral doses of cadazolid were well tolerated. Systemic cadazolid exposures were extremely low (maximum plasma concentration: 6.88 ng/mL), and increased in a less than dose-proportional manner over the dose range tested.

An open-label study (AC-061-103) was completed in subjects with severe CDAD to investigate the pharmacokinetics, safety, and tolerability of cadazolid following a single oral dose of 3000 mg in 6 subjects (4 males, 2 females). This dose was well tolerated in all subjects. Peak cadazolid plasma concentrations (C_{max}) (geometric mean 2.6 ng/mL) were low and were comparable to those observed in healthy subjects.

In healthy subjects, negligible quantities of drug were detected in the urine: on Day 10 approximately 0.01% or less of the oral dose was excreted in urine following the 300–3000 mg b.i.d. dosing regimen (Study AC-061-102). Almost the entire oral dose was recovered as unchanged drug in the feces. Similar results were observed in subjects with severe CDAD (AC-061-103). Thus, the majority of the oral dose is available to exert its maximum clinical effect in the gastrointestinal tract.

A Phase 2 study (AC-061A201) investigating the efficacy and safety of cadazolid in 84 adult subjects with CDAD has been completed. This study had a prospective, multi-center, double-blind, randomized, stratified, parallel-group, double-dummy design, and included a vancomycin reference arm. Three doses of cadazolid (250 mg, 500 mg, and 1000 mg, orally, b.i.d.) and vancomycin (125 mg orally, q.i.d.) administered for 10 days were evaluated. The modified Clinical Cure rates defined as the proportion of subjects having 3 or fewer unformed stools for at least 2 days and no further need for CDAD therapy at test-of-cure (TOC) 24–72 hours after the last dose of study treatment showed that all cadazolid doses were similarly effective, providing similar or numerically higher results versus vancomycin; e.g., 100% for the cadazolid 250 mg dose group versus 89.5% for vancomycin in the Per-protocol analysis set (PPS). Modified recurrence rates, defined as the proportion of subjects clinically cured who subsequently had a new episode of diarrhea and a positive *C. difficile* toxin test up to 4 weeks after the last dose of study treatment, were 18.8% for cadazolid 250 mg dose group versus 36.8% for vancomycin modified intent-to-treat (mITT) analysis set. The resulting modified Sustained Cure (Modified Clinical Cure without modified recurrence) rate was higher for cadazolid 250 mg dose group than for vancomycin (76.5% versus 54.5%, mITT analysis set) [Louie 2013a].

In the Phase 2 study, similar to that observed in both healthy subjects and subjects with severe CDAD, cadazolid showed very low systemic availability (maximum individual plasma concentration: 18.9 ng/mL in the cadazolid 1000 mg dose group). All oral doses of cadazolid were well tolerated.

1.2.2 Vancomycin

See Section 1.1.3 for information on vancomycin in the treatment of CDAD.

1.3 Purpose and rationale of the study

Cadazolid has demonstrated activity against *C. difficile* in nonclinical *in vitro* and animal studies. High concentrations of cadazolid in the intestine and low systemic absorption make it an attractive candidate for the treatment of CDAD in man. Phase 2 study results have suggested clinical response similar to or numerically higher than vancomycin, and higher Sustained Cure rates than vancomycin. Based on the Phase 2 results, a cadazolid dose of 250 mg twice daily for 10 days was chosen for further evaluation in a Phase 3 program. The selected dose is expected to optimize the clinical response and the impact on the gut microbiome, leading to improved Sustained Cure relative to comparator.

This study is 1 of 2 pivotal Phase 3 trials with identical design, each aimed at determining the efficacy and safety of the studied regimen of cadazolid relative to that of vancomycin in subjects with mild–moderate or severe CDAD. The identical study design will allow a pooled analysis of these 2 pivotal Phase 3 trials, including a predefined assessment of Sustained Cure in subjects with CDAD due to hypervirulent strains (defined currently as Polymerase Chain Reaction [PCR] ribotypes 027, 078, and 244). In addition, the 2 identical Phase 3 studies will allow specific analyses to be performed through the enrollment of subjects into shared protocol sub-studies (pharmacokinetic, gut microbiome, and the CDAD DaySyms Patient Reported Outcomes (PRO) psychometric validation).

2 STUDY OBJECTIVES

2.1 Primary objective

To determine whether the clinical response after 10-day oral administration of cadazolid is non-inferior to oral vancomycin, in subjects with CDAD.

2.2 Secondary objectives

- To determine whether oral administration of cadazolid for 10 days is superior to oral vancomycin in the sustained clinical response of subjects with CDAD.
- To determine whether the resolution of diarrhea (ROD) is more rapid with oral administration of cadazolid compared to vancomycin.
- To determine whether CDAD symptoms as reported by the subject show larger improvements from baseline with oral administration of cadazolid compared to vancomycin.

2.3 Objective of the meta-analysis of AC-061A301 and AC-061A302

To determine through a meta-analysis of both studies whether oral administration of cadazolid for 10 days is superior to oral vancomycin in the sustained clinical response of subjects with CDAD due to hypervirulent strains.

2.4 Exploratory objectives

To evaluate the plasma cadazolid concentration at approximately 2 h post-dose.

2.5 Safety objective

To determine safety and tolerability of an oral administration of cadazolid compared to vancomycin.

2.6 Objective of the re-treatment extension with cadazolid

To describe the clinical response, sustained clinical response and safety in subjects with a first occurrence of CDAD at study entry (cadazolid or vancomycin subjects), who experience a recurrence of CDAD and are re-treated with cadazolid for 10 days.

3 OVERALL STUDY DESIGN AND PLAN

3.1 Study design

This is a prospective, multi-center, double-blind, double-dummy, randomized, parallel group, active controlled, Phase 3 study. The study will be conducted in approximately 100 sites in approximately 20 countries worldwide.

Approximately 630 adult subjects will be randomized 1:1, stratified by first occurrence or first recurrence and by site, to the 2 treatment groups (approximately 315 subjects per group): cadazolid and vancomycin to ensure that at least 536 subjects will be included in the PPS.

Randomization will proceed until the required number of subjects has been reached and will be competitive across participating sites.

The study consists of the following study periods:

Screening Period lasts up to 48 hours from signature of the informed consent form (ICF) and ends with subject randomization. Subjects (or the legally authorized representative) must sign the ICF prior to protocol specified screening procedures. Baseline is defined as the last assessment prior to initiation of study treatment.

Treatment Period starts after randomization with the first dose of study drug at the end of Visit 1 (Day 1 of study), and ends with End-of-Treatment (EOT) on the day of the last dose of study drug (which is either Day 10 or Day 11 of the study to account for 10 complete days of treatment).

During this period, the following evaluations are performed:

- Visit 2 (on site or by telephone) on Day 5 of study (or Day 6 if the visit cannot take place on Day 5). This evaluation may be done by telephone if, in the investigator's

opinion, a face-to-face visit is not necessary. At this visit, the investigator's assessment of Early Treatment Response (ETR) occurs.

- Visit 3 (on site), on Day 8–10 of study, is performed while the subject is taking study drug. In case of premature discontinuation of study drug, Visit 3 is performed and the follow-up period takes place as planned.

In addition, unscheduled visits may also take place anytime during the treatment period.

Follow-up period starts immediately after the last dose of study drug, and ends approximately 30 days after the last dose of study drug, at Visit 5.

During this period, the following evaluations are performed:

- Visit 4 (on site or by telephone) occurs 2–4 days after the EOT. This visit may be done by telephone if, in the investigator's opinion, a face-to-face visit is not necessary. At this visit, the investigator will evaluate and record in the case report form (CRF) the Clinical Cure [according to the primary endpoint definition – see Section 6.1.1] on which the investigator will base the management of the subject for the rest of the study. In addition, the investigator will provide his/her judgment of Clinical Response [Section 6.1.2].

- Unscheduled visits (Visit 4.a, 4.b, etc.) to evaluate for recurrence occur when a subject experiences a new episode of diarrhea (NED) at any time between Visit 4 and 5. When a NED happens, the subject must contact the site immediately and an unscheduled visit (on site) should be organized within 48 h.

- Visit 5 (on site) occurs 30 ± 2 days after the EOT. At this visit, the investigator's judgment of sustained response (ISR) occurs. In case of early study withdrawal, Visit 5 should be scheduled within 48 h.

In addition, unscheduled visits (not due to a NED) may also take place during the treatment and follow-up periods, in which case study-related information will be collected.

The study also includes an extension:

Re-treatment extension with cadazolid: Subjects who experience a recurrence during the study, whether they have been treated with cadazolid or vancomycin, may enter a re-treatment extension with cadazolid (after providing a specific informed consent) consisting of a 10-day treatment of cadazolid followed by an approximately 30-day follow-up period (full details of visit and assessment schedule is provided in Section 8.4).

Study diaries

During the study (from Visit 1 to Visit 5), diaries and a journal are completed daily for each subject to collect the following information:

1. Frequency and consistency of stools [Section 7.2.1].

2. Study drug intake [Section 5.1.9].

3. CDAD DaySyms PRO – completed by all subjects who personally signed the ICF (i.e., not signed by a legally authorized representative) [Section 7.2.4].

Subjects should be contacted daily up to Visit 4 and twice weekly thereafter for interviews (either face-to-face or by telephone) to encourage diary compliance and for investigator evaluation of CDAD status based on the stool diary completion (i.e., frequency and consistency of stools).

Sub-studies

As part of this protocol, at selected sites and in subjects who consent to participate, the plasma concentrations of cadazolid will be analyzed [see Section 7.5].

- Pharmacokinetics:

To have at least 50 subjects with a cadazolid sample, a sub-population of at least 120 participating subjects from this study and study AC-061A302 will be recruited. In these subjects, plasma concentration of cadazolid will be determined.

In addition, the following sub-studies independent from the core study protocol and at selected sites in subjects who consent to participate (through an informed consent separate from the main protocol informed consent) will be performed according to stand-alone protocols:

- Psychometric validation of CDAD DaySyms PRO:

In a sub-population of approximately 165 participating subjects from this study and study AC-061A302 who personally signed the ICFs (i.e., not signed by a legally authorized representative) for AC-061A301 and for the sub-study, data required for the assessment of the final content validity of the CDAD DaySyms PRO as well as its psychometric properties will be collected. Subjects will be asked to complete 4 additional questionnaires at Visit 1 and Visit 3.

- Gut microbiome assessment and fecal cadazolid concentrations:

In a sub-population of approximately 100 participating subjects from this study and study AC-061A302, subjects will provide stool samples with the objective of analyzing

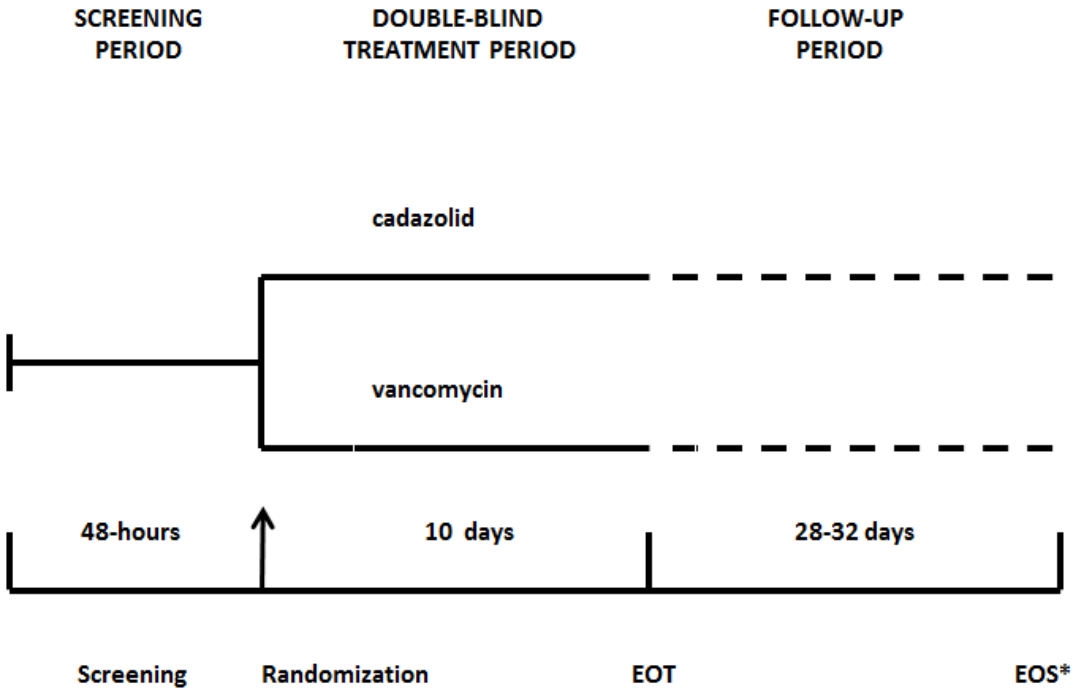
intestinal flora diversity and its variation on treatment and after treatment, the emergence of resistance, and, if needed, determining the fecal cadazolid concentration.

Study duration

Subject participation in the study will be up to 44 days; up to 88 days for subjects participating in the re-treatment extension with cadazolid.

Table 1 shows a schematic representation of the assessments during the study, and the overall study design is depicted in Figure 1.

Figure 1 Study design



*Unless subject enters the Re-treatment extension with cadazolid
EOT=end of treatment; EOS=end of study

3.2 Study design rationale

This is a prospective, multi-center, double-blind, double-dummy, randomized, stratified, parallel group, active controlled, Phase 3 study comparing the efficacy and safety of cadazolid versus vancomycin in subjects with CDAD, including hypervirulent strains.

Subjects with mild–moderate or severe CDAD will be enrolled in the study based on the diarrhea criteria documented within a pre-specified period and a positive result from a *C. difficile* GDH and toxin assay, as suggested in the addendum to the note for guidance on evaluation of medicinal products indicated for treatment of bacterial infections (CPMP/EWP/558/95 REV 2) [EMA 2011]. Only subjects with first occurrence or first recurrence of CDAD will be eligible since treatment for subjects with multi-recurrences is not standardized by regimen, dose, and/or duration.

Given the higher recurrence rate in subjects with a prior episode of CDAD, randomization is stratified on this factor (first occurrence versus first recurrence) to avoid imbalance between the treatment arms within each of these two strata. In addition, randomization will be stratified by site to avoid imbalance between treatment arms. Many factors can affect the Clinical Cure, recurrence and Sustained Cure of subjects with CDAD; in addition to the first occurrence versus first recurrence, age, comorbidity, immunosuppression, severity of illness, concomitant antibiotic use, and the hypervirulent strain may also impact the clinical outcome, alone or in combination. The study does not exclude subjects with immunosuppression, subjects requiring antibiotics, severe disease, subjects currently failing metronidazole, and CDAD due to hypervirulent strains. It is impractical to include all these factors for stratification. However, the site could show specific characteristics about the kind of patients that enroll (in-patients versus out-patients, severe subjects versus mild–moderate, the incidence of hypervirulent strains, etc.). An imbalance of the treatment groups within confounding factors could occur with central 1:1 randomization. Given the importance of these variables on outcome, including the hypervirulent strains, block randomization by site will more uniformly distribute the treatment arms across these factors.

The recruitment of subjects with CDAD due to hypervirulent strains currently defined as ribotypes 027, 078, and 244 will be monitored on an ongoing basis to ensure that the pooled analysis will have sufficient power to achieve its objective. Assuming approximately 25% of subjects enrolled present with hypervirulent strain, the pooled analysis will have 80% power to detect superiority of 15% on SCR. Consequently, to ensure recruitment of at least 315 subjects with hypervirulent strain in both clinical trials, geography or historical evidence of high incidence of these cases will be considered in site selection.

In study AC-061A201, the 250 mg b.i.d. dose was effective on the primary endpoint of Clinical Cure after a 10-day oral administration and no further improvement was observed with higher doses. For the secondary endpoints, recurrence and Sustained Cure, cadazolid 250 mg b.i.d. was also consistently effective with no further improvement at higher doses. In addition, this dose markedly decreased *C. difficile* vegetative forms as well as spores with no-to-minimal impact on the intestinal microflora. In addition,

cadazolid was well tolerated at all doses tested. As a result, the 250 mg dose has been selected for the Phase 3 studies.

Vancomycin has been proven efficacious in mild–moderate disease in the setting of clinical trials and is approved in many countries for the treatment of CDAD without any qualifier on the severity of disease. It is generally not used in the initial episode of mild–moderate disease. Vancomycin at a dose of 125 mg 4 times daily orally administered is approved for the treatment of CDAD in the United States (US) [Vancomycin USPI] and in Europe. The use of oral vancomycin is recommended as the treatment of choice for severe CDAD by both ESCMID and SHEA/IDSA guidelines and for recurrent episodes of CDAD in many countries [Bauer 2009, Cohen 2010], because of relatively high failure rates of metronidazole in recent reports and a slower clinical response to metronidazole compared with oral vancomycin treatment [Wilcox 1995, Musher 2005, O'Brien 2007, Zar 2007]. Metronidazole, though commonly used in the initial treatment of mild–moderate CDAD, is not licensed for this indication. Therefore, oral vancomycin, a licensed antibiotic for the treatment of CDAD, has been chosen as the comparator. The non-inferiority (NI) trial design is justified, as oral vancomycin has clearly demonstrated to be an effective treatment in CDAD. The justification for a 10% NI margin is provided in Appendix 7.

The use of kaolin or related products, charcoal containing anti-diarrheals, or binding agents such as cholestyramine is prohibited during the trial, and should not be administered, if possible, during the screening period. Co-administration of kaolin, activated charcoal, and tolevamer with cadazolid resulted in a 4- to 8-fold increase in cadazolid MIC with kaolin to more than a 64-fold increase in cadazolid MIC with charcoal (Actelion data on file).

The primary endpoint is Clinical Cure, which is defined by the ROD using records of stool frequency and consistency, and provided that no new antimicrobial treatment for CDAD is initiated, using concomitant medications records. The definition of the endpoint is similar to, albeit more stringent than the clinical Phase 3 studies of fidaxomicin. While in the pivotal studies of fidaxomicin (most recent trials having led to registration in CDAD) Clinical Cure was determined by the investigator based on a composite definition of cure at the EOT (including objective components such as unformed bowel movements [UBMs] and treatment but also the subjective component of investigator clinical judgment of significant improvement), in this study Clinical Cure is determined based on ROD, at a fixed time point: EOT + 2 days. This is a more robust definition which avoids different clinical judgment among investigators. This feature of the endpoint was criticized during the fidaxomicin NDA review. However, to allow fair comparison with the fidaxomicin trials an investigator judgment of clinical response is added as exploratory objective.

The secondary endpoint, sustained cure, is consistent with the endpoints suggested in the EMA guidance and consistent with the endpoint used in the evaluation of fidaxomicin to treat CDAD [Louie 2011]. Showing NI clinical cure rate (CCR) and superior SCR versus vancomycin, by hierarchical testing should provide robust evidence of reduction of recurrence of CDAD with cadazolid.

There is an unmet medical need for a treatment with improved SCR in subjects with CDAD due to hypervirulent strains. A central microbiology laboratory is required to determine if CDAD is due to hypervirulent strains. The proportion of subjects at the time of randomization with infection due to hypervirulent strains is expected to be approximately 25% based on the clinical Phase 3 studies of fidaxomicin. A meta-analysis using the data from the two pivotal Phase 3 trials will provide a sufficient number of subjects to evaluate the Sustained Cure of CDAD caused by hypervirulent strains.

Deviation from adherence to protocol tends to reduce the ability of a clinical trial to discriminate between treatments and therefore favors the demonstration of equivalence or NI. Every effort will be made to prevent violation of the protocol by providing clear instructions to the investigators, closely monitoring the data collected, and taking corrective actions in a timely fashion. In this study, with a primary analysis based on NI, both the PPS and the mITT analysis set are considered equally important. Subjects with protocol non-compliance that impacts the assessment of Clinical Cure will be excluded from the PPS. The sample size calculation has accounted for the potential exclusion of up to 15% of subjects from the PPS [see Section 11.2.4 for reasons for exclusion]. Sample size estimations were used to determine the number of subjects necessary to ensure 90% power in statistical analysis on the PPS, which implies a power over 90% for the statistical analysis on the co-primary population mITT.

The pharmacokinetics sub-study is performed to explore the range of systemic cadazolid plasma concentrations in subjects with CDAD administered the granule formulation. The 2 h post-dose has been shown in previous healthy subjects (AC-061-101 and AC-061-102) to represent the peak cadazolid concentrations. This single sample was used in study AC-061A201 in subjects with CDAD, where the powder formulation was administered. In order to get a good estimate of the plasma concentration range, approximately 50 evaluable samples from subjects on cadazolid treatment are required, accounting for the 1:1 randomization scheme.

In clinical practice, recurrences are common following treatment for CDAD. The re-treatment extension with cadazolid has been added to collect additional information on cadazolid treatment of first recurrences. The re-treatment extension with cadazolid will involve only subjects (cadazolid or vancomycin) with a first occurrence of CDAD at initial study entry who experience a first recurrence of CDAD during the study. Given the limited number of potential subjects who experience a recurrence and who are eligible for

the re-treatment extension with cadazolid in this study, and in order to maximize the data treating recurrences with cadazolid, all subjects who agree to participate will be given open-label cadazolid, as opposed to continuing the same treatments used in the first occurrence or to re-randomizing subjects to cadazolid or vancomycin. This re-treatment extension with cadazolid provides several unique opportunities to study:

- First recurrences following vancomycin treatment. Guidelines generally recommend metronidazole for first occurrence; therefore most of the subjects who enter the trial as a first recurrence in the main study will have been previously treated with metronidazole. The re-treatment extension with cadazolid will provide an opportunity to describe cadazolid efficacy and safety in a subset of subjects who have experienced a first recurrence following vancomycin treatment.
- First recurrences following cadazolid treatment. Since many first recurrences are treated with a second course of the same antibiotic, per guidelines, the re-treatment extension with cadazolid will provide an opportunity to describe cadazolid efficacy and safety in subjects who have experienced a first recurrence following cadazolid treatment.

3.3 Study committees

A Steering Committee composed of external experts has been established to participate in the design of the protocol, oversee the conduct of the study, evaluate the results and support publications. The Steering Committee is governed by a charter.

An Independent Data Monitoring Committee (IDMC) will be established prior to first subject randomization. The IDMC will have the overall responsibility for continuously safeguarding the interests of subjects by periodically monitoring study data as well as all relevant product-related information that may emerge during the conduct of the program. The IDMC is governed by a charter.

4 SUBJECT POPULATION

4.1 Subject population description

The study will enroll adult subjects aged 18 and older with CDAD.

CDAD is defined by the presence of diarrhea within 24 h prior to randomization with a stool test within 72 h prior to randomization positive for *C. difficile* GDH and toxin A/B by an enzyme immunoassay (EIA).

Enrollment will proceed until at least 630 subjects are randomized, to ensure that at least 536 subjects will be included in the PPS.

Subjects are allowed to be re-screened if they failed eligibility and were not randomized.

4.2 Inclusion criteria

For inclusion in the study, all of the following inclusion criteria must be fulfilled:

1. Signed informed consent prior to any study-mandated procedure.
2. Male or female ≥ 18 years of age at the screening visit.

   Non-pregnant women of childbearing potential:
   A woman is considered to be of childbearing potential unless she meets at least one of the following criteria:

   - Previous bilateral salpingo-oophorectomy or hysterectomy;
   - Premature ovarian failure confirmed by a health care professional;
   - XY genotype, Turner syndrome, uterine agenesis;
   - Postmenopausal, defined as 12 consecutive months with no menses without an alternative medical cause (ICH M3 definition).

   A non-pregnant woman of childbearing potential is eligible only if:

   - The absence of pregnancy is confirmed by a negative urine (or plasma/serum) pregnancy test at Visit 1;
   - She agrees to use one of the following methods of contraception from Visit 1 until 7 days after study drug discontinuation:
     - Condoms, diaphragm or contraceptive sponge if used in combination with a spermicide.
     - Intra-uterine devices.
     - Injectable contraceptive agents, levonorgestrel implants, or transdermal contraceptive hormone patches. If a hormonal contraceptive is chosen, it must be taken for at least 1 month prior to randomization.
     - Alternatively a sterilization method (tubal ligation or partner's vasectomy) is considered acceptable.

   She is in a situation of abstinence from intercourse with a male partner, when this is in line with the preferred lifestyle of the subject (e.g., homosexual women or women in a religious order – e.g., nuns).

   In case of oral contraception, an additional method must be employed, as diarrhea may affect the effectiveness of the oral contraceptive pill.

   Rhythm methods or the use of a condom by a male partner alone are not considered as acceptable methods of contraception.

3. Subject with a diagnosis of mild–moderate or severe CDAD (first occurrence or first recurrence within 3 months of randomization) with:
    - Diarrhea, defined as a change in bowel habits with > 3 UBMs, in the 24 h prior to randomization,
      AND
    - Positive *C. difficile* GDH and toxin A and/or B stool test, on the same sample collected no more than 72 h prior to randomization using an EIA test approved by the sponsor.

Note 1: If a positive test using an EIA test approved by the sponsor has been performed prior to screening within the time window mentioned above, the test does not need to be repeated and its result will document eligibility. If a non-approved test has been used for the diagnosis of CDAD prior to screening, an EIA test approved by the sponsor must show a positive result on a sample collected no more than 72 h prior to randomization, after the subject has signed the ICF to confirm eligibility.

Note 2: MTFs may be enrolled in the study. An MTF is defined as a subject who has received metronidazole for at least 72 hours for the current CDAD episode and is failing therapy defined as 1) continue to meet the definition of diarrhea (> 3 UBMs) without significant clinical improvement in the judgment of the investigator, and 2) remain GDH and toxin positive (with a positive stool test done on the same sample collected no more than 48 h prior to randomization). Metronidazole therapy must be stopped prior to first dose of study drug.

4.3 Exclusion criteria

For inclusion in the study, none of the following exclusion criteria must be met:

1. More than one previous episode of CDAD in the 3-month period prior to randomization.

2. Fulminant or life-threatening CDAD. If in the judgment of the investigator there is a suspicion of fulminant or life-threatening CDAD, the presence of any of the following criteria during the 72 h period prior to randomization and related to the fulminant or life-threatening CDAD episode excludes the potential subject from the study:

    - Septic shock – a systolic blood pressure (BP) < 90 mmHg or a mean arterial pressure < 70 mmHg in the absence of other causes of hypotension and that persists despite adequate fluid resuscitation
    - Peritonitis
    - Ileus

- Toxic megacolon
- Significant dehydration based on investigator judgment
- White blood cells count > 30.0×10^9/L
- Core body temperature > 40 °C

3. Concurrent immediately life-threatening disease or condition (likelihood of death within 72 h).
4. History of inflammatory colitides (e.g., ulcerative colitis or Crohn's disease, microscopic colitis, collagenous colitis) or chronic abdominal pain or chronic diarrhea of any etiology, or known positive diagnostic test for enteropathogens.
5. Vomiting or other condition that interferes with the ability to take oral medication or subjects with feeding tubes (i.e., when study drug would have to be given by the feeding tube).
6. Antimicrobial treatment active against CDAD administered for > 24 h except for metronidazole treatment failures [see Note 2 to Section 4.2].
   Note 3: Although the screening period is 48 hours, the subject must have ≤ 24 hours of antimicrobial treatment active against CDAD to qualify for the study, unless an MTF (see Note 2).
7. Planned additional treatment with antimicrobial medication active against CDAD, fecal microbiota transplant (FMT), or any forbidden concomitant medications [see Concomitant medications, Section 5.2].
8. FMT, intravenous immunoglobulins, or any investigational drug to prevent or treat CDAD in the 1 month (or 5 half-lives in case of investigational drug, whichever is longer) period prior to randomization.
9. Treatment with monoclonal antibodies against *C. difficile* in the 6-month period prior to randomization.
10. Investigational vaccination against *C. difficile*.
11. Previous participation in a clinical trial with cadazolid.
12. Known hypersensitivity or contraindication to any excipients of the investigational drug formulations, or to oxazolidinones, quinolones, or vancomycin.
13. Women who are breastfeeding.
14. Investigational site staff members or relatives, and Actelion employees.

15. Unable or unwilling to comply with all protocol requirements including study visits, study procedures, diary completion, medication adherence and appropriate study medication storage at home.

16. Any circumstances or conditions, which, in the opinion of the investigator, may affect the subject's full participation in the study, or compliance with the protocol.

5 TREATMENTS

5.1 Study treatments

5.1.1 Study drugs

Study drugs include the investigational drug cadazolid, the active comparator vancomycin and their matching placebos administered during the course of the study.

5.1.2 Investigational drug (cadazolid) and matching placebo

Oral cadazolid and its matching placebo are provided by Actelion as granules for oral suspension to be reconstituted prior to administration. Cadazolid is supplied at the dose of 250 mg in an aluminum sachet, matched for content, taste and appearance (before and after reconstitution) between the active drug and its matching placebo.

The inactive ingredients of the formulation are listed in the IB [Cadazolid IB].

The suspension must be reconstituted prior to administration, in accordance with the following instructions:

1. Open the sachet and pour its content into a glass (or a cup, a beaker or a goblet)
2. Add approximately 25 mL (about 2 tablespoons) of water to the glass.
3. Swirl immediately by hand with the base of the glass resting on a flat horizontal surface until a uniform suspension is obtained.
4. Drink the suspension.

Repeat steps 2 to 4 once to allow appropriate rinsing of the glass.

The oral suspension should be drunk within 5 min of reconstitution. Preparation and administration of at least the first dose of study drug by the subject must be done under the guidance of appropriately delegated site staff and documented in the source notes and CRF. If the suspension is not taken within 5 min of reconstitution, then it can be administered within the next 2 h after having resumed at Step 3.

5.1.3 Comparator (vancomycin) and matching placebo

Oral vancomycin and its matching placebo are provided as capsules. Each capsule of vancomycin contains 125 mg of active drug (Vancocin®, ViroPharma). Vancomycin

capsules have been over-encapsulated to maintain blinding. Over-encapsulation does not alter the dissolution of vancomycin.

The major excipients of the formulation are: FD&C Blue No. 2, gelatin, ferric oxide red, ferric oxide yellow, polyethylene glycol and titanium dioxide.

Vancomycin or its matching placebo does not require preparation; the capsule must be swallowed intact with or without water. Vancomycin or its matching placebo may be taken at the same time as the time-matched cadazolid placebo or cadazolid.

5.1.4 Dosing scheme

Each subject will receive blinded study medication administered orally each day; subjects will take doses approximately 6 hours apart (40 doses; every 6 hours for 10 days). On a daily schedule, this corresponds approximately to study drug doses at breakfast, lunch, late afternoon (dinner) and bedtime. Study drug can be taken with or without food.

Subjects randomized to cadazolid will receive 1 sachet of reconstituted cadazolid suspension twice daily and 1 capsule of placebo matching vancomycin 4 times daily. Subjects randomized to vancomycin will receive 1 capsule of vancomycin 4 times daily and 1 sachet of reconstituted placebo matching cadazolid suspension twice daily. Each time a sachet is taken, a capsule must be taken concomitantly [see Table 2].

Each dose of study drug, including date/time and composition (capsule/sachet) will be recorded in the Study Drug Journal by the subject or the study personnel.

Irrespective of the time of day the subject is randomized into the study, the first dose of study drug will include 1 sachet and 1 capsule under the guidance of site personnel. This will ensure that the first administration will always contain an active study drug. If study drug is interrupted for ≥ 1 dose (approximately 12 h or more since last dose), then it must be restarted with the subject taking 1 sachet and 1 capsule.

Table 2 Dosing scheme

	Cadazolid group	Vancomycin group
T_0	■ ○	□ ●
T_0 + 6h	○	●
T_0 + 12h	■ ○	□ ●
T_0 + 18h	○	●

■ Sachet of cadazolid □ Sachet of placebo matching cadazolid

● Capsule of vancomycin ○ Capsule of placebo matching vancomycin

5.1.5 Treatment assignment

At the beginning of Visit 1 after the subject has signed the ICF:

The investigator/delegate contacts the Interactive Voice Response System (IVRS) and obtains a subject number from the IVRS which will identify the subject throughout the study. Subjects are allowed to be re-screened [Section 5.1.1] if they failed eligibility and were not randomized. The subject number assigned during the first screening procedure will be retained in re-screened subjects.

At the end of Visit 1, after having ensured that the responsible personnel/pharmacist is available to dispense the study drug, all eligible subjects will be assigned a randomization number and a treatment kit:

- The investigator/delegate contacts the IVRS a second time after having confirmed that the subject fulfills all eligibility criteria. The IVRS assigns a randomization number to the subject, which is used to link the subject to 1 of the 2 treatment arms (1:1 ratio). The IVRS also specifies a treatment kit number to be assigned to the subject (the kit matches the treatment arm assigned by the randomization list).

The randomization list is generated by an independent contract research organization (CRO), [redacted]. The randomization code is generated using SAS 9.3. Randomization is stratified by site and by CDAD first occurrence or first

recurrence. The randomization code is kept strictly confidential. It is accessible only to authorized persons who are not involved in the study conduct or final analysis of the study, until the time of unblinding [see Section 5.1.7].

5.1.6 Blinding

This study will be performed in a double-blind fashion. The investigator and study staff, the subjects, the monitors, the sponsor staff (except the Clinical Trial Supply group, Quality Assurance-Global Quality Management, and Global Drug Safety in case of a suspected unexpected serious adverse reaction [SUSAR]), the CRO staff (except the bioanalytical laboratory in charge of analyzing cadazolid plasma concentration and the Independent Statistical Data Analysis Center [ISDAC] in charge of performing the unblinded statistical analysis for the IDMC meetings) will remain blinded to the treatment until study completion; see below for further details on double-blind exclusion. The investigational drug, the active comparator and their matching placebos are indistinguishable and all subject kits will be packaged in the same way.

For the bioanalytical laboratory in charge of analyzing cadazolid plasma concentrations, the randomization code will reside in a secure area of the bioanalytical laboratory server, where only designated people will have access. As per the signed contract, the bioanalytical laboratory will not send any subject or drug concentration data to Actelion, until they are authorized by Actelion in writing at the end of the study. However, they can convey to the study team, any issues relating to sample collection and/or handling/processing (e.g., sample arrived unfrozen, sample hemolyzed) so that corrective actions can be taken across all sites involved in the pharmacokinetic sub-study.

5.1.7 Unblinding

5.1.7.1 Unblinding for final study analyses and reporting

The randomization code will be broken and made available for the final statistical analysis only after study database closure in accordance with the sponsor's standard operating procedures (SOPs).

5.1.7.2 Unblinding before final analyses

An independent statistical data analysis center (ISDAC) [redacted], not otherwise involved in the design, conduct and analysis of the study, will have access to the randomization code in order to prepare unblinded reports for review by the IDMC (for IDMC review meetings during the course of the trial). The randomization code will be made available to the ISDAC in accordance with the sponsor's SOPs.

5.1.7.3 Unblinding of a Suspected Unexpected Serious Adverse Reaction (SUSAR)

When a SUSAR is identified for a subject participating in the study, Actelion Global Drug Safety will request the unblinding of the treatment code for that particular subject. This code will not be communicated to the site staff or to the Actelion study team. Unblinded SUSAR information is only provided to Actelion Global Drug Safety, respective health authorities and Institutional Review Boards/Independent Ethics Committees (IRBs/IECs). SUSARs will be anonymized.

5.1.7.4 Emergency procedure for unblinding

The investigator and the study staff must remain blinded to the subject's treatment assignment. The identity of the study drug may be revealed only if the subject experiences a medical event, the management of which would require knowledge of the blinded treatment assignment. In this case, the investigator can receive the unblinded randomization code for study drug dispensed through the IVRS. In these situations, the decision to unblind a subject's treatment assignment resides solely with the investigator. Whenever possible, and if it does not interfere with (or does not delay) any decision in the best interest of the subject, the investigator is invited to discuss the intended code break with the sponsor.

The site may contact the sponsor for any urgent question, including intended unblinding of treatment assignment, via the medical hotline numbers. These numbers can be found in the Investigator Site File (ISF), and also appear on the study drug and kit labels.

The occurrence of any unblinding during the study must be clearly justified in the subject's site file and in the CRF. The investigator must not disclose the unblinded treatment in the CRF or to the sponsor or its delegates. In all cases, the sponsor must be informed as soon as possible before or after the unblinding.

Refer to the IVRS guidelines for complete information regarding the IVRS procedures for randomization, study drug assignment, and unblinding.

5.1.8 Study drug supply

The sponsor will supply all study drugs to the site according to local regulations.

All drug supplies are to be used only in accordance with this protocol, and not for any other purpose.

5.1.8.1 Study drug packaging and labeling

5.1.8.1.1 Study drug packaging

Study drugs are provided as granules in sachets and capsules in childproof blister packs within a treatment kit.

Each kit contains sachets and capsules for 10 days of treatment plus a reserve for 2 days of treatment:

20 + 4 sachets of cadazolid 250 mg, granules for suspension or matching placebo.
40 + 8 over-encapsulated capsules of vancomycin 125 mg or matching placebo.

5.1.8.1.2 Study drug labeling

Each sachet and each blister pack of 8 capsules are simply labeled, whereas the treatment kit has a label with a detachable label for dispensing purposes. The labeling complies with the applicable laws and regulations of each participating country.

5.1.8.2 Study drug distribution and storage

The study drug kit must be stored in a secure location at a temperature between 2 °C and 25 °C / 35.6 °F and 77 °F; do not refrigerate. A temperature log is to be kept at each location where study drug is stored prior to subject dispensation. The adequacy of storage conditions should be documented. In the case of a temperature excursion, the site monitor must be immediately contacted. Subjects should store the medication at room temperature, less than 25 °C / 77 °F.

An order to dispense study drug must be written by the physician investigator (listed on FDA form 1572 for US sites only and/or the delegation of authority log for all sites). The study drug label must be detached from the kit and placed on an Study Drug Label Dispensing Log. The time of study drug administration and, in the case of the suspension, the time of preparation if the subject is an in-patient must be recorded in the source documents and CRF.

5.1.8.3 Study drug return and destruction

On an ongoing basis and upon study completion or termination of the study, the monitor will prepare used and unused subject kits for return to the local depot where the sponsor or deputy will check drug reconciliation before destruction.

5.1.9 Compliance with study treatment

Study drug accountability must be performed by the study staff on the day of the visit and recorded on the Study Drug Dispensing and Accountability log, in order to ensure that the subject is compliant with study requirements. Study drug accountability is also recorded in the case report form (CRF) and checked by the monitor during site visits.

Subjects are asked to return the treatment kit, including empty sachets and blister packs, and all unused study drug at each site visit. An accurate record of the date and amount of study drug dispensed to each subject must be available for inspection at any time.

For each subject, a Study Drug Journal will be completed daily to record timing of each dose of study drug (sachets and capsules). This information will be utilized to reconcile

with study drug kits and the study drug dispensing and accountability log and for the study staff to enter information on study drug intake and compliance with study treatment in the CRF.

5.1.10 Study drug interruption and premature discontinuation of study drug

Temporary interruption of study treatment, including missed doses must be recorded in the CRF with the reason for interruption. If study drug is interrupted (≥ 1 dose or approximately 12 h or more since last dose), then it must be restarted with the subject taking 1 sachet and 1 capsule. Premature discontinuation of study drug (withdrawal from treatment) may happen either independently from study withdrawal or in conjunction with study withdrawal.

If the subject discontinues from study treatment but remains in the study, all subsequent scheduled visits (telephone or on site) and collection of information (e.g., diary cards) will be performed according to protocol up to Visit 5.

If the subject discontinues not only from study treatment but also does not agree to continue in the study, then the subject withdraws from the study [see Section 9.1].

The reason for premature discontinuation from study drug (withdrawal from treatment), the decision owner (as applicable), and whether subject decision to discontinue study treatment does or does not include study withdrawal, must be documented in the CRF. Reasons for premature discontinuation are mutually exclusive and only the primary reason for premature discontinuation should be recorded in the CRF.

Premature discontinuation can occur at any time during the treatment period. A subject who requires insertion of a feeding tube during the treatment period and who requires administration of study drug through the feeding tube, must discontinue study drug. If the subject is a treatment failure or has received an insufficient duration of therapy prior to premature discontinuation of the study drug (e.g., adverse event [AE]), then the study drug is discontinued and an alternative appropriate therapy for CDAD is initiated [Section 5.2.2].

In case of premature discontinuation, Visit 3 is performed and the subject enters the follow-up period. Every effort must be made to perform the Visit 3 assessments before initiation of alternative therapy for CDAD. Subjects prematurely discontinued from study drug for any reason will not be replaced.

5.2 Previous and concomitant medications

A previous medication is defined as a medication that was previously taken and was stopped prior to the first dose of study medication. A concomitant medication is defined as a medication that started, stopped, or was ongoing between first dose of study medication and Visit 5.

All previous (within 7 days of randomization or in the past 3 months for antibiotics and acid suppressing medications, see Section 7.1) and concomitant medications (including mandatory methods of contraception, see Section 7.1) are to be recorded in CRF (and subject's file) along with the appropriate indication.

5.2.1 Prohibited and allowed concomitant medications

The following concomitant medications are prohibited during the study:

- Antimicrobial treatments active against CDAD or FMT up to Visit 5 unless provided for clinical failure or recurrence [Section 5.2.2]:
  - Antimicrobial treatments active against CDAD include: oral vancomycin, metronidazole, bacitracin, fusidic acid, nitazoxanide, teicoplanin, tigecycline, fidaxomicin, rifampicin/rifampin or rifaximin.
- Other medication active against CDAD including probiotics, intravenous immunoglobulins, and binding agents (e.g., cholestyramine) up to Visit 5.
- Initiation of treatment with opiates after randomization up to and including 2 days after the EOT.
- A change in dose/regimen resulting in an increased opiate effect up to and including 2 days after the last dose of study drug is forbidden.
  Note: In the case of ongoing treatment with opiates at randomization, a stable treatment with opiates or a decrease in the dose/regimen or discontinuation of treatment with opiates is allowed. For equivalence between opiates see Appendix 1.
- Anti-peristaltic medications (e.g., loperamide), kaolin or related products, pectin or charcoal-containing anti-diarrheals up to Visit 5.
- Any investigational drug, antibody or vaccine to prevent or treat CDAD up to Visit 5.

Notes:

1. Proton pump inhibitors (PPIs) and H_2 blockers are allowed during the study.
2. The use of kaolin or related products, charcoal containing anti-diarrheals, or binding agents (e.g., cholestyramine) should be discontinued as soon as possible during the screening period for subjects likely to be randomized.

5.2.2 Alternative therapy for CDAD

In case of Clinical Failure or recurrence, an antimicrobial treatment active against CDAD (or FMT) should be initiated as recommended per local guidelines and/or physician judgment.

Subjects who have a first occurrence of CDAD at baseline, and who experience a recurrence are eligible for participation for re-treatment with cadazolid [Section 8.4].

6 STUDY ENDPOINTS

6.1 Efficacy endpoints

6.1.1 Primary efficacy endpoint

Primary efficacy endpoint determination

Daily, from randomization to Visit 5, the subject or the study personnel (e.g., when the subject is hospitalized) records each bowel movement and the time it is produced, and evaluates whether the bowel movement meets the definition of a UBM (i.e., taking the shape of the container in which it is produced) in a stool diary [see Section 7.2.1 for more information on the stool diary].

Subject interviews are performed face-to-face or by telephone every day starting on Day 2 up to Visit 4 and then twice weekly no more than 4 days apart until Visit 5 by site personnel to document the CDAD status of the subject and to ensure that the subject is collecting the information appropriately in the stool diary since the last subject interview. During these subject interviews, the study personnel will collect and document information regarding the number of UBMs (or presence or absence of > 3 UBMs if exact number unknown) and total number of stools for each day. Based on this information, ROD (ROD: ≤ 3 UBM per day for at least 2 consecutive days) or any NED (NED: > 3 UBMs after ROD) can be assessed [see Section 7.2.2 for more information on subject interviews].

During on-site study visits, the study personnel review the stool diary with the subject to ensure consistency with other source documentation, including the subject interviews. The study personnel ensure that the subject is collecting the information appropriately and reconciles information as necessary.

On each bowel movement entry of the stool diary, every effort should be made to indicate whether this is a UBM or not, even if the timing of the bowel movement is unknown (in which case Not Known will be entered for the time). In the stool diary, reconciled stool diary entries are distinguished from initial subject entries. The stool diary is collected on an ongoing basis and stored at the site.

After reconciliation, the investigator enters the reconciled number of UBMs and the total number of stools in the Daily Stool Information section of the CRF. In the event that the exact number of UBMs is not known but sufficient information is available to document the presence or absence of > 3 UBMs, then this information must be recorded. This section of the CRF must be filled out based on all information available to the investigator, i.e., recorded on the reconciled stool diary and daily subject interview.

The full information from the reconciled stool diary (timing and consistency of each bowel movement) is entered in the Stool Log section of the CRF. The investigator is

responsible for the accuracy of stool data entered into the CRF, based on the review and reconciliation of source documentation. Based on the reconciled information gathered during daily subject interviews up to EOT + 2 days, the investigator can evaluate if the subject meets the 'per-protocol' definition of Clinical Cure and can document it at Visit 4 in the CRF. A worksheet to assist in the determination of the PP definition of clinical cure can be found in the CRF.

At the end of the trial, the information collected in the Daily Stool Information section of the CRF will be used by the sponsor to determine the primary endpoint Clinical Cure (see definition below).

The monitors performing source data verification ensure that the CRF entries match the reconciled stool diary and the documentation of the daily subject interviews.

Definition of the primary endpoint

Clinical Cure

Clinical Cure is defined as:

- ROD (≤ 3 UBMs per day for at least 2 consecutive days) on study treatment and maintained for 2 days after EOT

  AND

- No additional antimicrobial treatment active against CDAD or FMT between first dose of study drug and 2 days after EOT (inclusive).

ROD is determined from the number of UBMs (or if the exact number is missing from presence or absence of > 3 UBMs), per day recorded in the Daily Stool Information section of the CRF. Days are measured according to calendar days.

Antimicrobial treatment active against CDAD or FMT is determined from concomitant medications/procedures CRF pages.

Subjects who do not fulfill the requirements for Clinical Cure are considered Clinical Failures.

6.1.2 Secondary efficacy endpoints

The information collected in the Daily Stool Information section will be used to determine Sustained Cure. The daily timing and consistency of the stools recorded in the Stool Log section will be used to derive Time to ROD.

Sustained Cure

Sustained Cure is defined as:

- Clinical Cure

  AND

- No recurrence

Subjects who do not fulfill the requirements for a Sustained Cure are considered not to be a Sustained Cure.

Recurrence is defined for subjects with Clinical Cure as:

- NED: > 3 UBMs within 1 day between 3 days after EOT and Visit 5

  AND

- Stool test showing positive *C. difficile* GDH and toxin test on the same stool sample (according to EIA tests approved by the sponsor)

  AND

- Antimicrobial treatment active against CDAD (including participation in the re-treatment extension with cadazolid) or FMT started between 3 days after EOT and Visit 5.

NED is determined from the number of UBMs, or if the exact number is missing from presence or absence of > 3 UBMs, per day collected in the Daily Stool Information section of the CRF. Days are measured according to calendar days.

A positive stool test is determined from the corresponding unscheduled visit for NED, where the result of the test is recorded.

Antimicrobial treatment active against CDAD or FMT is determined from concomitant medications/procedures CRF pages.

Subjects with Clinical Failure, or with Clinical Cure and recurrence, are considered to be without Sustained Cure.

Please note that recurrence, according to the same definition, is per se an exploratory efficacy endpoint [Section 6.1.4].

Time to Resolution of Diarrhea

Time to ROD is defined as:

- The time (h) elapsed between the first dose of study drug

  AND

- The time when ROD is considered achieved

Time when ROD is considered achieved is the time of the last UBM prior to the first 2 consecutive 24 h periods with ≤ 3 UBMs and subsequently ≤ 3 UBMs per day maintained up to 2 days after EOT. The 2 consecutive 24 h periods and last UBM date/time prior to that period is determined from the Stool Log section of the CRF. The condition of maintaining ≤ 3 UBMs per calendar day is determined from the Daily Stool Information section of the CRF.

Absolute change from baseline in CDAD DaySyms PRO total daily score

Absolute change from baseline in CDAD DaySyms PRO total daily score up to Day 12 derived from the daily CDAD DaySyms PRO.

6.1.3 Meta-analysis endpoint

Sustained Cure in subjects with hypervirulent strains (currently defined as Strains 027, 078, and 244) is the same as Sustained Cure defined in Section 6.1.2.

6.1.4 Other efficacy endpoints – supportive of primary and secondary efficacy objectives

Investigator's assessment of ETR at Visit 2

This is assessed by the investigator as improved, unchanged or worsened compared to baseline.

Investigator Judgment of Clinical Response (ICR) Visit 4

Investigator responses for ICR consist of cure, and failure. The investigator will qualify on which basis he/she makes the assessment:

- **Cure** will be qualified by any of the following criteria (more than one criterion may apply):
  - Subject required no additional CDAD therapy or FMT between first dose of study drug up to and including 2 days after EOT.

- Subject had ≤ 3 UBMs for 2 consecutive days on therapy and maintained for 2 days after EOT.
- Subject had a marked reduction in the number of UBMs (in the opinion of the investigator).
- Subject had stabilization and improvement in CDAD signs and symptoms (other than diarrhea – e.g., abdominal pain, fever), or other criteria (e.g., WBC elevation [not due to a clear alternative etiology]).

- **Failure** will be qualified by any of the following criteria (more than one criterion may apply):
  - Subject required additional CDAD therapy or FMT between first dose of study drug up to and including 2 days after EOT.
  - Subject did not have ≤ 3 UBMs for 2 consecutive days on therapy and maintained for 2 days after EOT.
  - Subject did not have a marked reduction in the number of unformed stools (in the opinion of the investigator).
  - Subject had worsening of CDAD signs and symptoms (other than diarrhea – e.g., abdominal pain, fever), or other criteria (e.g., WBC elevation [not due to a clear alternative etiology]).

The investigator will have the opportunity to specify any additional criterion which applies in the CRF.

Investigator's judgment of Sustained Response (ISR) at Visit 5

Investigator responses for ISR consist of Sustained Cure and not a Sustained Cure. The investigator will qualify on which basis he/she makes the assessment.

Sustained Cure will be qualified by:

- Subject was considered a cure at the ICR

  AND

- Subject met one or more of the following criteria:
  - Subject required no additional CDAD therapy or FMT between EOT + 3 days up to and including Visit 5.
  - Subject maintained ≤ 3 UBMs per day between EOT + 3 days up to and including Visit 5.
  - Subject maintained a marked reduction in the number of UBMs (in the opinion of the investigator) between EOT + 3 days up to and including Visit 5.

- Subject maintained the stabilization and improvement in CDAD signs and symptoms (other than diarrhea – e.g., abdominal pain, fever), or other criteria (e.g., WBC elevation [not due to a clear alternative etiology]) between EOT + 3 days up to and including Visit 5.
- Subject maintained other criteria (specified in ICR response) between EOT + 3 days up to and including Visit 5; please specify criteria.

Not a Sustained Cure will be qualified by:

- Subject was considered a failure at the ICR.

  OR

- Subject met one or more of the following criteria:
  - Subject required additional CDAD therapy or FMT between EOT + 3 days up to and including Visit 5.
  - Subject had a repeat stool test between EOT + 3 days up to and including Visit 5 showing positive *C. difficile* GDH and toxin test on the same sample (according to EIA tests approved by the sponsor).
  - Subject did not maintain $\leq$ 3 UBMs per day between EOT + 3 days up to and including Visit 5.
  - Subject did not maintain a marked reduction in the number of UBMs (in the opinion of the investigator) between EOT +3 days up to and including Visit 5.
  - Subject did not maintain the stabilization and improvement in CDAD signs and symptoms (other than diarrhea – e.g., abdominal pain, fever), or other criteria (e.g., WBC elevation [not due to a clear alternative etiology]) between EOT + 3 days up to and including Visit 5.
  - Subject did not maintain other criteria (specified in ICR response) between EOT + 3 days up to and including Visit 5; please specify criteria.

The investigator will have the opportunity to specify any additional criterion which applies in the CRF.

Early Clinical Cure by Day 5

Early Clinical Cure by Day 5 is defined as:

- ROD by Day 5: $\leq$ 3 UBMs for at least 2 consecutive days prior to or including Day 5 and maintained up to EOT + 2 days,

  AND

- No additional antimicrobial treatment active against CDAD or FMT between first dose of study drug and EOT + 2 days as defined in Section 6.1.1.

Normalization of Bowel Movements (NBM) defined as:

- ≤ 2 bowel movements per day and no UBM on at least 2 consecutive days up to EOT and maintained up to 2 days after the last dose of study drug,

  AND

- No additional antimicrobial treatment active against CDAD or FMT between randomization and 2 days after EOT.

Time to return to usual stools

- Time to return to usual stools is defined as the time (days) elapsed between the first dose of study drug and the first day where the stools have returned to usual for a subject i.e., the number of bowel movements (including UBM) per day ≤ number of bowel movements reported as usual by the subject and maintained for at least 3 calendar days.

Recurrence [Section 6.1.2]

Recurrence can be further classified as relapse or re-infection based on microbiology results [see Section 7.2.5.2]

Time to recurrence

Defined as the time (h) elapsed between the last dose of study drug and the onset time of NED determined to be a recurrence.

The information collected in the Stool Log section of the CRF are used to determine time to recurrence, and the information collected in the Daily Stool Information section of the CRF to determine all the other endpoints.

Change from baseline in CDAD DaySyms PRO scores – supportive of the CDAD DaySyms PRO secondary objective:

- Change (absolute and/or percent) from baseline in CDAD DaySyms PRO total daily score to each day.
- Change from baseline (absolute and/or percent) in CDAD DaySyms PRO domain scores to each day.

Susceptibility of *C. difficile* to cadazolid, vancomycin, linezolid, moxifloxacin, metronidazole, and fidaxomicin.

Change from baseline in susceptibility of *C. difficile* to cadazolid, vancomycin, linezolid and moxifloxacin in case of Clinical Failure or recurrence. Post-baseline MIC increases ≥ 4-fold are considered microbiologically relevant.

Change from baseline to Visit 3 in vancomycin-resistant enterococci (VRE) count for vancomycin-resistant *E. faecium* and vancomycin-resistant *E. faecalis*.

Susceptibility of VRE to cadazolid, vancomycin, linezolid, moxifloxacin, fidaxomicin, daptomycin, tigecycline, ampicillin, gentamicin, and quinupristin-dalfopristin for vancomycin-resistant *E. faecium* and vancomycin-resistant *E. faecalis*.

6.2 Safety and tolerability endpoints

- Deaths up to Visit 5.
- AEs and serious adverse events (SAEs) up to Visit 5.
- Treatment-emergent AEs (TEAEs) and SAEs up to 7 days after the EOT.
- AEs leading to premature discontinuation of study treatment.
- Change from baseline to Visit 3 and Visit 5 in vital signs (SBP, DBP, and HR), body temperature and body weight.
- Marked abnormalities in vital signs (SBP and DBP) up to EOT + 7 days (treatment emergent) and up to Visit 5.
- Change from baseline to Visit 3 and Visit 5 in ECG parameters.
- Marked abnormalities in ECG up to EOT + 7 days (treatment-emergent) and up to Visit 5.
- Change from baseline to Visit 3 and Visit 5 in hematology, coagulation, and blood chemistry parameters.
- Marked abnormalities in hematology, coagulation, and blood chemistry parameters up to EOT + 7 days (treatment-emergent) and up to Visit 5.

6.3 Pharmacoeconomic endpoints

- Length of stay (days) in hospital.
- Length of stay (days) by ward unit (i.e., general ward, step-down/intermediate unit, Intensive Care Unit [ICU]).
- Re-admission/admission to the hospital after start of treatment.
- Visit to emergency department after the start of treatment.

- Change from baseline in work productivity and activity impairment: *C. difficile*-associated diarrhea (WPAI:CDAD) scores.
- WPAI:CDAD scores on absenteeism, presenteeism, work productivity loss, and activity impairment are assessed.

6.4 Pharmacokinetic endpoints

Plasma cadazolid concentrations at 2 h post-dose at Visit 3.

7 STUDY ASSESSMENTS

7.1 Baseline parameters

Baseline demographics include sex, age, ethnicity, and race and are to be recorded in the CRF at Visit 1, including for subjects screened but not randomized.

Medical history is to be documented at Visit 1 and recorded in the CRF.

The following must be systematically recorded:

- Chronic medical conditions and new acute medical conditions in the past 6 months
- Exposure to healthcare settings in the past 3 months
- Antibiotic exposure in the past 3 months
- Use of acid-suppressing medications in the past 3 months
- History of chemotherapy and immunosuppression
- History of gastrointestinal surgery including appendectomy
- Number of daily bowel movements (including UBM) reported as usual by the subject, prior to the episode of CDAD
- Serum creatinine, prior to the episode of CDAD, if available
- Concomitant baseline medications
- For female subjects, presence or absence of child-bearing potential, as well as a situation of true abstinence from intercourse with a male partner (in this case, the reason should be collected in source document), or mandatory methods of contraception (see below) must be recorded in the CRF.
  - Condoms, diaphragm or contraceptive sponge if used in combination with a spermicide.
  - Intra-uterine devices.
  - Injectable contraceptive agents, levonorgestrel implants, or transdermal contraceptive hormone patches. If a hormonal contraceptive is chosen, it must be taken for at least 1 month prior to randomization. In case of oral contraception, an additional method must be employed, as diarrhea may affect the effectiveness of the oral contraceptive pill.

- Alternatively a sterilization method (tubal ligation or partner's vasectomy) is considered acceptable.

Baseline CDAD disease characteristics must be documented in CRF, and include:

- First CDAD occurrence or first recurrence; documentation of the first episode, including treatment, dose, and dates should be collected in subjects with a first recurrence
- Metronidazole Treatment Failure (MTF)
- In-patient or out-patient status at randomization
- Number of bowel movements, including UBMs within 24 h prior to randomization
- Abdominal physical examination (normal/abnormal and details)
- Severe CDAD is defined as any **one** of the following:
  1. Maximum core temperature recorded at baseline > 38.5 °C
  2. WBC > 15.0×10^9/L (based on Central Laboratory)
  3. Rise in serum creatinine > 50% compared to pre-CDAD diagnosis

  Note: Mild–moderate disease is considered if none of these criteria that define severe CDAD are satisfied.
- Presence of pseudomembranes or histopathology classified as pseudomembranous colitis (for subjects having had endoscopy)
- Presence of distension of large intestine, colonic wall thickening, including low attenuation mural thickening, pericolonic fat stranding, or ascites not explained by other causes (for subjects having had imaging)
- Date and time of fecal sampling for *C. difficile* strain based on Restriction Endonuclease Assay (REA) type and PCR ribotype [Section 7.2.5.2].

7.2 Efficacy assessments

7.2.1 Stool diary

Stool frequency and consistency is recorded daily in a diary from randomization to Visit 5. The subject or the study personnel (e.g., when the subject is hospitalized) record each bowel movement and the time it is produced, and evaluate whether the bowel movement meets the definition of a UBM (i.e., taking the shape of the container in which it is produced). Entries made by the study personnel in the stool diary are distinguished from entries made by the subject. The stool diary is shown in Appendix 2.

During on-site study visits, the study personnel review the stool diary with the subject to ensure consistency with source documentation including the daily subject interviews [Section 7.2.2]. The study personnel ensure that the subject is collecting the information appropriately and reconciles information as necessary in the appropriate column in the stool diary or an alternative document to record reconciliation (reconciled stool diary entries). For each bowel movement entry, every effort should be made to indicate

whether this is a UBM or not, even if the timing of the bowel movement is unknown (in which case Not Known will be entered for the time). The reconciled stool diary entry is collected on an ongoing basis and stored at the site following the end of subject participation in the study.

- Based on this diary, several endpoints including Clinical Cure and Sustained Cure (Recurrence), are determined.
- The investigator may consider the subject as treatment failure (in case of no change or worsening diarrhea and other signs and symptoms of CDAD) at any time during the treatment period.
- The investigator evaluates ETR at Visit 2, investigator judgment of Clinical Response (ICR) at Visit 4, and investigator judgment of sustained Response (ISR) at Visit 5.

The subject and the study staff can determine when a NED happens and triggers an unscheduled visit for recurrence; the subject must contact the site immediately and a visit should be scheduled within 48 hours.

7.2.2 Subject interviews

Subject interviews (face-to-face or by telephone every day starting from Day 2 up to Visit 4 and then twice weekly no more than 4 days apart until Visit 5) are performed by site personnel to document the status of the subject. An interview log will be provided to investigators/study personnel to document site communication with the subject.

During the subject interview the following information is discussed with the subject:

1. Date, time, and consistency of each bowel movement (BM) since last subject interview;
2. Change in symptoms of CDAD since last subject interview;
3. New concomitant medications or treatments since last subject interview;
4. Compliance with study drug and completion of the Study Drug Journal and reminder to record doses taken, missed doses, and to retain the study drug containers for collection at the on-site visits;
5. New AEs since last subject interview.

During these subject interviews, the study personnel will collect and document information regarding each BM and can determine the number of UBMs per day and total number of stools per day. In the event that the exact number of UBMs is not known but sufficient information is available to document the presence or absence of > 3 UBMs then this information will be recorded in the source documentation and reconciled with the stool diary. Based on this information, ROD or any NED can be assessed by the investigator.

An interview log will document the dates of the interviews in the CRF and the information collected or documentation that each element of the subject interview was discussed with the subject will be recorded in the subject interview source documentation. This documentation will be used by the study personnel to reconcile information collected by subject in the stool diary [see Section 7.2.1 above]. The remainder of the information collected (signs and symptoms, concomitant treatment, AEs or compliance) must be recorded in the corresponding CRF sections. The source documentation of the subject interviews is stored at the site during and following the end of subject participation in the study.

7.2.3 Investigator assessments

Investigator assessments are performed at Visit 2, Visit 4 and Visit 5.

At Visit 2 for ETR, the investigator determines the subject response based on the:

1. Frequency and consistency of stool
2. Signs and symptoms of CDAD
3. Additional antimicrobial treatment active against CDAD or FMT

Investigator responses consist of improved, unchanged, or worsened.

At Visit 4, the investigator evaluates and records in the CRF if the subject fulfills the PP definition of Clinical Cure [as defined in Section 6.1.1] on which the investigator will base the management of the subject for the rest of the study. In addition, the investigator provides his/her own judgment of Clinical Response [ICR – see Section 6.1.4]: Clinical Cure and ICR consist of cure or failure.

At Visit 5, the investigator evaluates and records in the CRF if the subject fulfills the PP definition of Sustained Cure [as defined in Section 6.1.2]. In addition, the investigator provides his/her own judgment of Sustained Response [ISR – see Section 6.1.4]: Sustained Cure and ISR consist of Sustained Cure and not Sustained Cure.

These assessments must be recorded in the CRF.

7.2.4 CDAD DaySyms PRO

The CDAD DaySyms PRO is a new questionnaire that has been developed by the sponsor in accordance with the FDA Guidance for Industry on Patient-Reported Outcome Measures: Use in Medical Product Development to Support Label Claims [FDA 2009] to assess the symptoms of CDAD from a subject perspective. The items forming the CDAD DaySyms PRO were developed through a literature review, discussions with expert clinicians, an initial phase of interviews with US subjects suffering from CDAD to assess symptoms frequent and relevant to them, and a second phase of interviews of US and Canadian subjects to confirm the items in the draft instrument. Results suggest that the

CDAD DaySyms PRO is a comprehensive measure assessing symptoms which are relevant to subjects with CDAD, and that the CDAD DaySyms PRO is easy to complete for subjects.

The CDAD DaySyms PRO comprises 13 items with a recall period of 24 h. The 13 items are:

1. Diarrhea
2. Feeling that you need to empty your bowels right away
3. Needing to go to or use the bathroom more than usual
4. Passing gas (flatulence)
5. Abdominal cramping
6. Abdominal pain
7. Feeling bloated (feeling like you need to loosen your clothes)
8. Feeling tired
9. Lack of energy
10. Lightheadedness
11. Dizziness
12. Lack of appetite
13. Nausea.

The questionnaire will be administered in a paper-and-pencil format daily. The items are scored on a five-scale response scale (None, Mild, Moderate, Severe, and Very Severe). The scoring of the scale will be established during the psychometric validation of the instrument [Appendix 3].

All subjects who personally signed the ICF (i.e., the ICF was not signed by a legally authorized representative) will complete the questionnaire daily from Visit 1 to Visit 4. These subjects must complete the CDAD DaySyms PRO by themselves without help from anybody else. The first completion of the CDAD DaySyms PRO will take place prior to randomization. From then onwards until Visit 4, the subject will complete the questionnaire in the evening of each day. The data from the paper forms will be transferred into the study database via double data entry.

The sponsor has the right to use the CDAD DaySyms PRO in this study, as it was developed by Actelion.

All translations have been carried out according to international guidelines on translations and linguistic validations.

7.2.5 Stool microbiology

Fecal sampling for microbiology is performed at baseline for subject eligibility and to document the *C. difficile* baseline characteristics.

Samples are also taken at Visit 3 and in case of NED (Visit 4.a, 4.b, etc.) If no stool is available on the day of Visit 3 or Visit 4, then a stool sample may be collected within 24 h after the visit and brought to the study site as described above; this is not valid for the Visit 1 sample that must be collected prior to randomization.

The subject will be instructed to collect the stool sample ideally within 24 h of the scheduled appointment or agreed upon drop-off time, and to store the sample in a refrigerator until departing for the study site. The subject should record the date and time at which the sample was produced. The samples should be delivered to the site using cooler bag and gel packs.

At the study site the investigator will store two tubes containing stools from each sample at around −70 °C/−94 °F (in exceptional cases and after written confirmation by the sponsor, samples may be stored below −20 °C/−4 °F in a non-frost-free freezer), one tube for the microbiology laboratory and one back-up tube kept until it is later sent to the microbiology laboratory. Date and time at which the stool sample was produced are recorded on the tube labels and in the CRF.

In case of NED, only UBMs must be collected. If the stool sample is collected at the subject's place of residence, the subject should call the study site immediately to make an appointment or arrangements to drop the sample at the study site as soon as possible.

The subjects will receive guidelines for fecal sampling and delivery to the study site.

The study sites will be instructed to ship the tubes at regular intervals, on dry ice, to the Central Laboratory. The tubes containing stools will be stored frozen at the Central Laboratory [redacted]. Investigators will not receive results from the central microbiology laboratories but from the sponsor after the end of the trial.

7.2.5.1 C. difficile GDH and toxin A/B tests

At Visit 1, a positive GDH and toxin A/B test, performed locally, is needed to document CDAD diagnosis and confirm subject eligibility.

The sponsor will provide an EIA test to all sites wherever and whenever possible, following local regulations. This test detects glutamate dehydrogenase (GDH) antigen in addition to detecting toxin A and B.

If a GDH and toxin A/B test (using an EIA test approved by the sponsor) has been performed prior to screening and returned a positive result, the test should not be repeated as this result will document eligibility.

If a non-approved test has been used for the diagnosis of CDAD prior to screening, then after the subject has signed the ICF, a GDH and toxin A/B test (using an EIA test approved by the sponsor) must show a positive result to confirm eligibility.

The GDH and toxin A/B test for eligibility must be performed on the same sample of stool (UBM only) collected within the 72 h (48 h in case of MTF) prior to randomization.

In case of suspected recurrence during a NED, a GDH and toxin A/B test (using an EIA test approved by the sponsor) must be performed on the same sample of stool (UBM only) to confirm recurrence.

If the sponsor-provided test is used, then the result of the test will be recorded in the CRF while the lot number and expiration date of the test will be recorded in the site source documentation. If the sponsor-provided test is not used, then the name of the sponsor-approved GDH and toxin A/B test, with the results of the test, will be recorded in the CRF. A negative GDH or toxin A/B test result for the screening sample will exclude the subject from the mITT analysis set.

7.2.5.2 C. difficile strain identification and susceptibility

The Central Laboratory analyzes the feces received in the frozen tubes to isolate *C. difficile* at baseline and in cases of Clinical Failure or recurrence. The *C. difficile* isolates are then tested for susceptibility against cadazolid, vancomycin, linezolid, moxifloxacin, metronidazole, and fidaxomicin in accordance with the Clinical and Laboratory Standards Institute (CLSI) testing standards.

The *C. difficile* isolates are shipped to specialty laboratories for identification of *C. difficile* strains (REA and PCR ribotyping) and susceptibility testing will be performed for all subjects, at baseline and in cases of treatment failure or recurrence. This will lead to identification of hypervirulent strains currently defined as PCR ribotype 027, 078, or 244 [Freeman 2010, Lim 2014, De Almeida 2013]. New hypervirulent strains emerging during the course of the study may be added. Strain identification will also be used to differentiate relapse (*C. difficile* strain identical to the baseline strain) from re-infection (*C. difficile* strain different from the baseline strain) in subjects with recurrence.

7.2.5.3 Vancomycin-resistant enterococci (VRE) quantitative culture and susceptibility

The Central Laboratory analyzes the feces received in the frozen tubes to culture and enumerate VRE isolates (*Enterococcus faecium* and *Enterococcus faecalis*) from Visit 1 and Visit 3 fecal specimens for all subjects. The VRE isolates are then tested for

susceptibility against cadazolid, vancomycin, linezolid, moxifloxacin, fidaxomicin, daptomycin, tigecycline, ampicillin, gentamicin, and quinupristin-dalfopristin in accordance with the CLSI testing standards.

7.3 Safety and tolerability assessments

The definitions, reporting, and follow-up of AEs, SAEs and pregnancies are described in Section 10.1.1.

7.3.1 Vital signs

BP and HR are to be measured, and recorded in the CRF, at Visit 1 (prior to randomization), Visit 3, unscheduled visits (if applicable), and Visit 5.

Systolic BP (SBP) and diastolic BP (DBP) are to be measured using the same type of device throughout the study, the dominant arm being preferred. Measurements are to be recorded preferably in the supine position and after the subject has rested for 5 min or more. If possible, HR should be measured at the same time as BP.

HR and BP are recorded in the CRF. Clinically relevant vital signs abnormalities, which meet the definition of an AE [Section 10.1.1], must be recorded by the investigator on the AE page of the CRF.

Body temperature is measured or (if available in the subject medical chart) collected at Visit 1 (prior to randomization), Visit 3, unscheduled visits (if applicable), and Visit 5, and must be recorded in the CRF (in case of multiple measurements available in a day the maximum temperature of the day must be entered in the CRF).

Recommended body temperature measurements are rectal, oral, ear, or axilla. Body temperature is recorded in the CRF. Conversion to core body temperature will be performed programmatically.

Marked abnormalities in BP are defined in Appendix 4.

7.3.2 ECG

A standard 12-lead ECG is to be performed, at rest with the subject in the supine position for a 5-min period, at Visit 1 (prior to randomization) Visit 3, unscheduled visits (if applicable), and Visit 5.

ECGs are read by a local cardiologist after each subject has completed the study and then the information is entered in the CRF. If the site is unable to identify a local cardiologist, ECGs will be evaluated by [redacted] and the information will be directly transferred electronically to Actelion.

The following quantitative variables will be measured and recorded in the CRF: PR (ms), QRS (ms), QT (ms), HR (beats per minute [bpm]).

QT_C (ms) will be calculated according to Bazett's and Fridericia's formula (QTcB = QT/(RR) ½ and QTcF = QT/(RR) ⅓, respectively).

ECG findings based on the local cardiologist's interpretation will be recorded in the ECG CRF page. Clinically relevant ECG findings, as evaluated by the investigator, made after study drug initiation, which meet the definition of an AE [Section 10.1.1], must be recorded on the AE page of the CRF.

7.3.3 Weight and height

Body weight is to be measured and recorded in the CRF at Visit 1 (prior to randomization), Visit 3, unscheduled visits (if applicable), and Visit 5. Height is to be measured and recorded in the CRF at Visit 1. Body mass index (BMI) will be derived from height and weight.

7.3.4 Physical examination

A physical examination is to be performed and recorded in the CRF at Visit 1 (prior to randomization), Visit 3, unscheduled visits, and Visit 5.

Physical examination should be recorded by body system in the CRF as normal or abnormal. If an abnormality is found then it should be specified in the CRF page, describing the signs related to the abnormality (e.g., systolic murmur) not the diagnosis (e.g., mitral valve insufficiency). Clinically relevant findings (other than those related to CDAD) that are present at Visit 1 must be recorded on the Medical History CRF page. Clinically relevant physical exam findings made after study drug initiation, which meet the definition of an AE [Section 10.1.1], must be recorded by the investigator on the AE page of the CRF.

7.3.5 Laboratory assessments

7.3.5.1 Type of laboratory

The [redacted] Global Central Laboratory will be used for all hematology, coagulation, and blood chemistry. All Central Laboratory reports will be communicated to the investigator.

At the time of randomization, local laboratory results are used for checking subject eligibility since Central Laboratory results would not be available within the 48 h screening period. The local laboratory results (with the corresponding normal ranges) and pregnancy test will be recorded in the CRF. Laboratory parameters required at Visit 1 are repeated at the Central Laboratory to be used as the baseline values for safety laboratory endpoints specified in the protocol. Laboratory certification / reference ranges /

laboratory director's Curriculum Vitae (CV) will be collected by the sponsor for all local laboratories providing data that are entered into the clinical and/or safety databases.

Analysis of laboratory parameters for safety monitoring, including the routine monitoring of laboratory parameters and protocol mandated follow-up in case of abnormal values, is performed by the Central Laboratory. In specific circumstances (such as an emergency) the initial detection of an abnormality and/or its follow-up may be done by local laboratory testing. In such instances, the local laboratory results (with the corresponding normal ranges) will be entered in the clinical database via dedicated CRF pages; this includes all laboratory results associated with an unscheduled visit, as well as all abnormal laboratory values associated with an AE, including all follow-up results to document resolution. In addition, laboratory certification / laboratory director's CV will be collected if not already collected as part of the baseline local laboratory certification.

Clinically relevant abnormalities from Central Laboratory results (or local laboratory when applicable) made after study drug initiation, which meet the definition of an AE [Section 10.1.1], must be recorded by the investigator on the AE page of the CRF. In the case of a clinically significant abnormality, repeated assessments are mandatory until return to baseline, stabilization or until the change is no longer clinically relevant.

Results from the Central Laboratory and local laboratories will be used for safety laboratory endpoints determination. With Sponsor approval, if a laboratory test is unable to be analyzed by the Central Laboratory for technical reason and if values have been obtained on the same day from a local laboratory for this laboratory test, then the local value will be entered in the CRF.

7.3.5.2 Laboratory parameters

Hematology

- Hemoglobin, hematocrit
- Platelet count
- Erythrocyte count
- Leukocyte count with differential

Coagulation

- Prothrombin time / international normalized ratio (INR)
- Activated partial thromboplastin time (aPTT)

Chemistry

- Aminotransferases (AST/ALT), alkaline phosphatase, total and direct bilirubin
- Creatinine, blood urea / blood urea nitrogen

- Sodium, potassium, chloride, calcium
- Protein, albumin

Hematology, coagulation and chemistry laboratory assessments are performed at Visit 1 (prior to randomization), Visit 3, unscheduled visits, and Visit 5. Subjects do not need to fast.

Pregnancy test

A urine or plasma/serum pregnancy test will be performed locally at screening for women of childbearing potential to document eligibility prior to randomization.

Approximately 13.2 mL of blood (i.e., 2 mL of blood for the hematology tests; 8.5 mL for chemistry tests, and 2.7 mL for coagulation tests) will be collected for the Central Laboratory tests at each visit where laboratory parameters are assessed. The date and time of collection of samples for laboratory tests will be recorded in the CRF. Further details regarding blood sampling procedures, collection and shipment of samples and reporting of results are described in the [redacted] Global Central Laboratory Manual. Definitions for marked laboratory abnormalities are listed in Appendix 4. Normal ranges are presented in Appendix 5.

7.4 Pharmacoeconomic assessments

7.4.1 Work Productivity and Activity Impairment Questionnaire: *Clostridium difficile*-associated diarrhea V2.0 (WPAI:CDAD)

The WPAI:CDAD is a 6-question, patient-reported quantitative assessment of the amount of absenteeism, presenteeism, work productivity loss, and activity impairment attributable to CDAD during the previous 7 days.

The sponsor has adapted for CDAD the specific health version of the WPAI Version 2 which was developed by Reilly Associates [Reilly 1993; see Appendix 6].

All subjects who sign the ICF themselves will complete the WPAI:CDAD. The WPAI:CDAD will be completed by the subject at Visit 1, Visit 3 and Visit 5. These subjects must complete the WPAI:CDAD by themselves without help from anybody else.

It is recommended that at Visit 3 and Visit 5 the WPAI:CDAD questionnaire is completed prior to any clinical assessments. Preferably, subjects would complete the WPAI:CDAD before any interaction with healthcare providers to avoid any potential bias or impact of interventions in their responses.

Subjects will complete the questionnaire on a paper form. The data from the paper forms will be transferred into the study database via double data entry.

The sponsor has notified the developer about the use of the instrument in this study. No license is required for the administration of the instrument.

7.4.2 Other pharmacoeconomic assessments

Information on the subject's total length of stay in the hospital and the length of stay in specific types of hospital units including general ward, step-down/intermediate care unit, and an ICU (in days) will be collected.

ICU (or critical care unit) is defined as a hospital unit in which concentrated special equipment and specially trained personnel take care of seriously ill subjects requiring immediate and continuous attention.

Step-down/intermediate care unit is defined as a transitional unit for subjects from ICU that provides close monitoring and provision of non-critical care.

Information on the reason of the subject's (re-)admission to hospital will be captured as a variable, differentiating between admission for CDAD and admission for other reasons.

Information on the subject's location (e.g., home, nursing home, specific hospital ward) prior to admission to hospital as well as the subject's location upon discharge will be collected.

Information on the type and number of CDAD-related procedures (FMT, hemi-colectomy, colectomy) will be collected.

This information collected as described above will be used for the pharmacoeconomic evaluation.

7.5 Pharmacokinetic assessments

In a sub-population of at least 120 subjects pooled from AC-061A301 and AC-061A302, on Visit 3 following a sachet and capsule dose, a 3 mL blood sample will be drawn approximately 2 h post-dose for determination of cadazolid plasma concentrations. The actual time post-dose will be used in the assessment (Note: All PK blood samples taken up to 3 h post-dose will not be considered as protocol deviations). The time of the 3 previous sachet/capsule doses prior to this blood sample, whether the last dose prior to this blood sample was taken within 1 h of consuming a meal (not a snack e.g., bag of crisps or a biscuit), and the time of blood sample collection should be recorded in the CRF.

Approximately 3 mL of blood will be collected by direct venipuncture in an antecubital vein in the arm in a Monovette tube (or equivalent) containing ethylenediaminetetraacetic acid. Immediately following collection of the required blood volume, the tubes will be slowly tilted backwards and forwards (no shaking) to bring the anti-coagulant into

solution, and immediately cooled on ice. Within 30 min of collection, the tubes will be centrifuged at approximately 2000 g for 10 minutes at 2–8 °C. When a centrifuge that can be cooled is not available, the blood samples and the bucket of the centrifuge must be cooled on ice prior to centrifugation. The plasma will then be transferred into one labeled polypropylene tube, avoiding carry-over of erythrocytes. All samples will be stored in an upright position at −70 °C or lower. The actual clock time of collection of the blood sample will be entered in the CRF.

Labeling
The tubes for both collecting blood for cadazolid determination and storage of plasma samples will be provided along with pre-printed labels to each of the participating sites by the central laboratory before starting the study. They will provide fields to collect the following information:

Actelion Pharmaceuticals Ltd
AC-061A301
Plasma
Centre number
Subject number
Study day
Date / Time collected

Bioanalysis
All samples from subjects on cadazolid will be analyzed by the bioanalytical laboratory. Twenty percent of the samples from subjects on vancomycin will be analyzed by the bioanalytical laboratory as negative controls. No samples should be destroyed until the study report is signed off.

Cadazolid plasma concentrations will be determined using a validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) assay. The foreseen limit of quantification (LOQ) is 0.25 ng/mL. Concentrations will be calculated by interpolation from a calibration curve. Quality control samples will be analyzed throughout the study, and their measured concentrations will be determined between-run and overall precision and accuracy of the analysis.

Shipping procedures
The plasma samples for cadazolid determination must be shipped from the sites to the Central Laboratory, then to the bioanalytical laboratory on a regular basis, as agreed by the sponsor. The samples must be packed securely with completed shipment forms, (detailing what the tubes contain, subject numbers, site number and date) in polystyrene insulated shipping containers, together with enough dry ice to keep the package frozen for at least 48 h. Arrangements must be made with the regional central laboratory before

the site sends the sample to the Central Laboratory, then with the bioanalytical laboratory before the regional central laboratory sends the samples to the bioanalytical laboratory.

8 SCHEDULE OF VISITS

Table 1 provides an overview of the chronological sequence of the assessments.

At each study visit, study personnel must remind women of child-bearing potential to use the methods of contraception defined for this study.

Unless otherwise specified, study visits must be performed on site; however, visits at the subject's place of residence may be acceptable after having received documented agreement from the sponsor.

The following tasks must be carried out by a physician investigator:

- Informed consent process
- Specific protocol procedures including the decision on subject eligibility, laboratory test result interpretation (including GDH and toxin A/B test), study drug prescription (decision on start and discontinuation), assessment of AEs/SAEs, and ECG reading for the ongoing safety assessment of the subject (Note: Only ECG interpretations made by the local cardiologist are entered in the CRF [see Section 7.3.2]).
- CRF/Query sign off.

The physical exam and some tasks related to the informed consent process can be delegated to an advanced practice role (e.g., nurse practitioner) provided that this is in accordance with local regulations, site-specific SOPs and the person's education and certification.

8.1 Screening and Randomization / Visit 1

The Screening Period will last for up to 48 h prior to randomization and includes:

- After discussing the study with the investigator and after agreeing to study participation by signing the ICF, subjects will be assigned a subject number by the IVRS provider. For any subject signing an ICF, it is the responsibility of the investigator to obtain written informed consent prior to any screening assessment. If a study-specific procedure or assessment has been performed as part of routine assessments and the results are available prior to the signing of the ICF, such procedure or assessment may be used to assess eligibility and does not have to be repeated. In such cases, it must be clear from the source document when and for which reason the assessment was done prior to the signing of the informed consent. The subject number will identify the subject throughout the study. Subjects are allowed to be re-screened if they failed eligibility and were not randomized. The

subject number assigned during the first screening procedure will be retained in rescreened subjects.

- *C. difficile* GDH and toxin test (approved by the sponsor). At investigational sites where the sponsor-approved *C. difficile* GDH and toxin test is not standard of care, a two-part ICF may be used: Section A covering the *C. difficile* GDH and toxin test only, and Section B covering the enrollment into the study.
- Fecal sampling for microbiology
- Recording of demographics, medical history, and disease characteristics
- Recording of previous (within 7 days of randomization or in the past 3 months for antibiotics and acid suppressing medications) and concomitant medications (including mandatory methods of contraception)
- Recording of body temperature (maximum value if multiple assessments), vital signs (BP, HR), body weight, and height
- Physical examination
- Hematology, coagulation, blood chemistry, and urine (or plasma/serum) pregnancy test for women of childbearing potential
- 12-lead ECG
- Completion of the CDAD DaySyms PRO diary
- WPAI:CDAD
- Recording of SAEs.

The investigator will check all inclusion/exclusion criteria to ascertain final eligibility. The reasons for screening failure are documented in the IVRS system and in the CRF (screening information is collected for all screen failure subjects).

After all screening assessments are done and eligibility determined, and after having ensured that the pharmacist is available to dispense study drug, the subject is randomized in the IVRS. As a result, a randomization number and a treatment kit number are assigned. Randomization is followed by the administration of the first dose of study drug (1 sachet **AND** 1 capsule). Preparation and administration of, at least, the first dose of study drug by the subject must be done by or under the guidance of the investigator or the study coordinator.

Subjects are instructed on study drug administration and Study Drug Journal, prohibited medications and therapy, stool diary, and CDAD DaySyms PRO. They are also provided with the necessary kit and guidelines for feces collection, sampling, and delivery.

Subjects are allowed to leave the study site only after the investigator has confirmed that their clinical status is compatible with being discharged. Subjects are reminded to contact the site immediately in case of an AE.

8.2 Treatment period

The treatment period starts after randomization with the administration of the first dose of study drug (1 sachet **AND** 1 capsule). Preparation and administration of, at least, the first dose of study drug prepared by the subject must be done under the guidance of the site personnel.

The treatment period ends 10–11 days later, on the day of the last dose of study drug, depending on the time of the day the subject takes the first dose. If the subject is an out-patient or at the time of discharge from the hospital between Visit 1 and Visit 3, the subject receives the remainder of the treatment kit. During the entire treatment period, a Study Drug Journal recording study drug intake is completed daily by the subject or the study personnel (if hospitalized).

Women of childbearing potential are reminded to use acceptable methods of contraception from Visit 1 until 7 days after study drug discontinuation; methods of contraception are recorded in the CRF. All subjects are interviewed face-to-face or by telephone every day up to Visit 4 and then twice weekly up to Visit 5.

8.2.1 Visit 2

Visit 2 takes place on Day 5 (or Day 6 if the visit cannot take place on Day 5) – this evaluation may be done at the site or by telephone, if in the investigator's opinion, a face-to-face visit is not necessary. Visit 2 includes:

- Subject interview
- Review of stool diary and reconciliation of source documents if necessary (if on-site visit),
- Verify compliance with CDAD DaySyms PRO
- Review of Study Drug Journal (if on-site visit)
- Investigator assessment of Early Treatment Response
- Recording of concomitant medications, including repeated instructions on prohibited medications
- Recording of AEs and SAEs

Subjects are also provided with the necessary kit and guidelines for feces collection, sampling, and delivery (if on-site visit and this has not been done at Visit 1).

Premature discontinuation of study drug including treatment failure (if there is no change or worsening of the subject then the investigator may consider the subject a treatment failure) may occur at any time during the treatment period, in particular during subject interviews (i.e., telephone calls) and planned visits/evaluations (i.e., Visit 2 and Visit 3). In case of premature discontinuation of study drug, Visit 3 is performed and the

follow-up period takes place as planned. Every effort must be made to perform the Visit 3 assessments before initiation of alternative therapy for CDAD.

8.2.2 Visit 3

Visit 3 occurs on site between Day 8 and Day 10 while the subject is taking study drug. Visit 3 includes:

- Administer WPAI:CDAD first
- Fecal sampling for microbiology
- Subject interview
- Review of stool diary and reconciliation of source documents if necessary, verify compliance with CDAD DaySyms PRO
- Review of Study Drug Journal
- Physical examination and recording of body weight
- Recording of vital signs (BP, HR) and temperature
- Hematology, coagulation, and chemistry tests
- 12-lead ECG
- Recording of concomitant medications
- Recording of AEs and SAEs
- Provide subjects with the necessary kits for microbiology sampling during the Follow-up Period
- Schedule an appointment for Visit 4 (if a face-to-face visit is necessary) and Visit 5.

8.3 Follow-up period

The follow-up period starts immediately after the last dose of study drug and ends approximately 30 days after the last dose of study drug.

8.3.1 Visit 4

Visit 4 occurs 2–4 days after the EOT (at site or by telephone if it is the investigator's opinion that a face-to-face visit is not necessary).

Visit 4 includes:

- Subject interview
- Review of stool diary and reconciliation of source documents if necessary (if on-site visit)
- Verify compliance with CDAD DaySyms PRO
- Review of Study Drug Journal (if on-site visit)
- Recording of concomitant medications
- Recording of AEs and SAEs

- Investigator's evaluation of the subject by the per-protocol definition of Clinical Cure (please note a subject can only be declared a Clinical Cure when there is ROD and no additional CDAD therapy through EOT + 2 days). The follow-up is performed similarly, with the same scheduled assessments, whether the subject is assessed as clinically cured or not.
- Investigator's judgment of clinical response (ICR)

8.3.2 Unscheduled visits for recurrence

Unscheduled visits (Visit 4.a, 4.b, etc.) to evaluate for recurrence may occur when a subject experiences a NED at any time between Visit 4 and 5. Under some conditions [see Section 8.4], the subject may enter the re-treatment extension. Subjects cannot be enrolled into the re-treatment extension if Visit 5 occurs after end of treatment + 32 days.

See below in Section 8.3.4. for information on unscheduled visits not related to NED that can occur at any time during the study. Unscheduled visits for recurrence may include:

- Subject interview
- Review of stool diary and reconciliation of source documents if necessary.
- Review of Study Drug Journal (if Visit 4 not an on-site visit)
- Physical examination
- Recording of vital signs (BP, HR) and temperature
- Hematology, coagulation, and chemistry tests
- Recording of concomitant medications
- GDH and toxin A/B test on the same stool sample, in case of suspected recurrence
- Fecal sampling for microbiology (only in the subjects having recurrence)
- Recording of AEs and SAEs.

Note: All assessments must be performed in the case of a recurrence.

Subject may enter the re-treatment extension with cadazolid in case of recurrence [see Section 8.4].

8.3.3 Visit 5

Visit 5 takes place 30 ± 2 days after last dose of study drug (in case of study withdrawal, Visit 5 should be scheduled within 48 h [see Section 9.1] and includes:

- Administer WPAI:CDAD first
- Subject interview
- Review of stool diary and reconciliation of source documents if necessary
- Review of Study Drug Journal (if Visit 4 not an on-site visit and no episode of NED)
- Physical examination, and recording of body weight

- Recording of vital signs (BP, HR) and temperature
- Hematology, coagulation, and chemistry tests
- 12-lead ECG
- Recording of concomitant medications
- Recording of CDAD related procedures (FMT, colectomy, hemi-colectomy)
- Recording of AEs and SAEs
- Investigator's evaluation of the subject by the per-protocol definition of Sustained Cure
- Investigator's judgment of sustained response (ISR)

8.3.4 Unscheduled visits

Unscheduled visits (Visit U1, U2, etc.) unrelated to NED may occur between scheduled visits as described above. The date of the visit and the reason for the visit, as well as data related to study assessments performed at unscheduled visits will be collected in the CRF.

8.4 Re-treatment extension with cadazolid (optional)

Subjects who experience a NED [see Section 6.1.2] and fit with eligibility criteria, including a positive GDH and toxin test (performed on the same stool sample), etc., are invited to enter, after providing informed consent, a re-treatment extension consisting of a 10-day treatment with cadazolid followed by an approximately 30-day follow-up period. The subjects will follow the same visit schedule (visits are labeled by adding 'Re-' in front of the visit number) as described in the main study protocol. The description of the procedures to be followed during the visits, re-treatment Visit 1 (Re-1) to re-treatment Visit 5 (Re-5), are identical to the corresponding visits in the main study protocol (Visit 1–5) that are described in Section 8.1, Section 8.2, Section 8.3, and Section 8.4, with the exceptions listed later in this section. The schedule of assessment for the re-treatment extension with cadazolid is described in Table 3.

If the subject does not qualify for the re-treatment extension with cadazolid, an alternative therapy is prescribed for the recurrence and the subject remains on the original visit and assessment schedule.

When a subject experiences a recurrence and participation in the re-treatment extension is confirmed (after the subject has signed the ICF for re-treatment and eligibility is confirmed), Visit 5 in the main study is not performed. Assessments that would have been performed as part of Visit 5 in the main study are performed as part of re-treatment Visit 1 (Re-1). Subjects cannot be enrolled into the re-treatment extension if Visit 5 occurs after end of treatment + 32 days.

Assessments performed after signing the Informed Consent for the re-treatment extension are recorded in the Visit Re-1 section of the CRF. Assessments performed at the NED

visit, prior to signing the Informed Consent for re-treatment extension and as long as they have been performed within 24 h of enrollment (72 h of enrollment for the fecal sampling), do not need to be repeated for Visit Re-1. In these cases it will be recorded on the applicable eCRF at visit Re-1 that the assessment was not performed (i.e., assessment performed and recorded at the NED visit).

The re-treatment extension with cadazolid will involve only subjects (cadazolid or vancomycin) with a first occurrence of CDAD at initial study entry who experience a first recurrence of CDAD during the study. The subject has to fulfill the following eligibility criteria:

Inclusion criteria

1. Signed informed consent prior to any re-treatment extension-mandated procedure.
2. Male or female ≥ 18 years of age at the screening visit.

   A non-pregnant woman of childbearing potential [as defined in Section 4.2] is eligible only if:

   - The absence of pregnancy is confirmed by a negative urine (or plasma/serum) pregnancy test at Visit Re-1
   - She agrees to use one of the methods of contraception [defined in Section 4.2] from Visit Re-1 until 7 days after study drug discontinuation.
3. Subject with a diagnosis of mild–moderate or severe CDAD at Visit Re-1 with:
   - Diarrhea, defined as a change in bowel habits with > 3 UBMs, in the 24 h prior to enrollment into the re-treatment extension,
     AND
   - Positive *C. difficile* GDH and toxin A and/or B stool test, on the same sample collected no more than 72 h prior to enrollment using an EIA test approved by the sponsor.

Exclusion criteria

1. Any previous episode of CDAD in the 3-month period prior to initial randomization into the study.
2. Fulminant or life-threatening CDAD. If in the judgment of the investigator there is a suspicion of fulminant or life-threatening CDAD, the presence of any of the following criteria during the 72 h period prior to enrollment and related to the fulminant or life-threatening CDAD episode excludes the potential subject from the re-treatment extension with cadazolid:

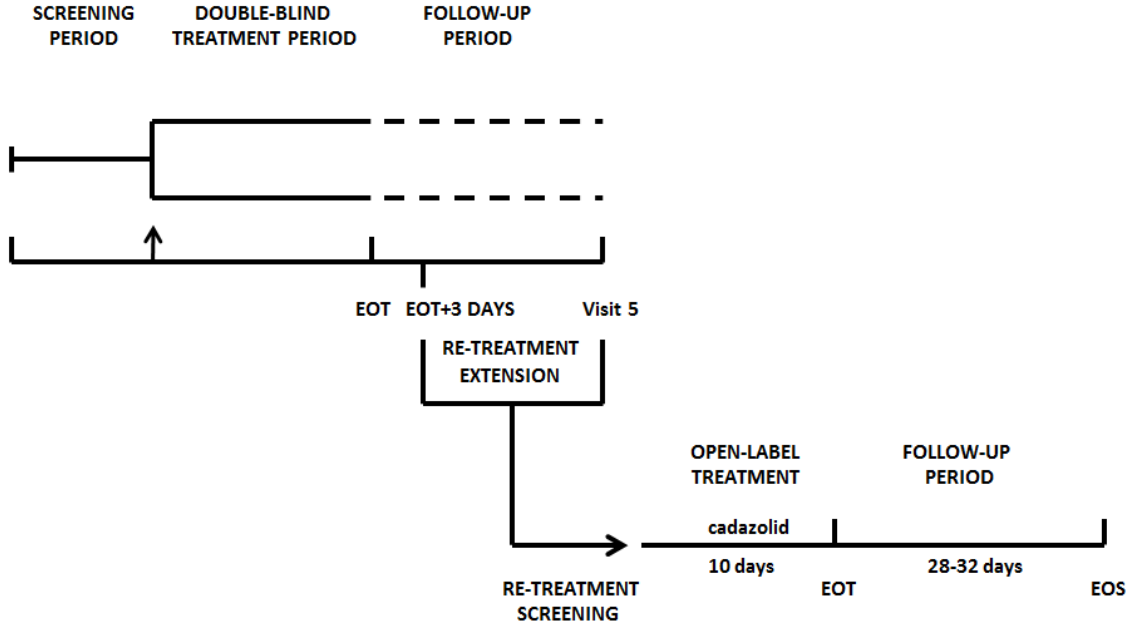
- Septic shock – Systolic BP < 90 mmHg or a mean arterial pressure < 70 mmHg in the absence of other causes of hypotension and that persists despite adequate fluid resuscitation
- Peritonitis
- Ileus
- Toxic megacolon
- Significant dehydration based on investigator judgment
- White blood cells count > 30.0×10^9/L
- Core body temperature > 40 °C

3. Concurrent immediately life-threatening disease or condition (likelihood of death within 72 h).
4. History of inflammatory colitides (e.g., ulcerative colitis or Crohn's disease, microscopic colitis, collagenous colitis) or chronic abdominal pain or chronic diarrhea of any etiology, or known positive diagnostic test for enteropathogens.
5. Vomiting or other condition that interferes with the ability to take oral medication or subjects with feeding tubes (i.e., when study drug would have to be given by the feeding tube).
6. Antimicrobial treatment active against CDAD administered for > 24 h for the current episode of CDAD.
7. Planned additional treatment with antimicrobial medication active against CDAD, FMT, or any forbidden concomitant medications [see Concomitant medications, Section 5.2].
8. FMT, intravenous immunoglobulins, or any investigational drug to prevent or treat CDAD in the 1 month (or 5 half-lives in case of investigational drug, whichever is longer) period prior to enrollment.
9. Treatment with monoclonal antibodies against *C. difficile* in the 6-month period prior to enrollment.
10. Investigational vaccination against *C. difficile*.
11. Previous participation in a clinical trial with cadazolid prior to initial randomization into the study.
12. Known hypersensitivity or contraindication to any excipients of the investigational drug formulations, or to oxazolidinones or quinolones.
13. Women who are breastfeeding.

14. Investigational site staff members or relatives, and Actelion employees.
15. Unable or unwilling to comply with all protocol requirements including study visits, study procedures, diary completion, medication adherence, and appropriate study medication storage at home.
16. Any circumstances or conditions, which, in the opinion of the investigator, may affect the subject's full participation in the study, or compliance with the protocol.

The study design including the re-treatment extension is depicted below.

Figure 2 Study design including re-treatment extension



EOT=end of treatment; EOS=end of study

A subject is considered to have completed the extension and the study if re-treatment Visit 5 (Re-5) is performed at least 28 days after the last dose of open-label cadazolid.

Upon eligibility, the subject number is kept. A re-treatment kit number is assigned and an additional 10-day treatment with cadazolid is provided.

The study treatment is an oral administration of cadazolid 250 mg twice daily (with or without food): 1 sachet of reconstituted cadazolid suspension is to be taken approximately 12 h apart, i.e., at breakfast and late afternoon (dinner), or at lunch and bedtime. Randomization and blinding (or unblinding) procedures do not apply. The treatment duration is also 10 days until EOT when the last dose is taken.

Efficacy endpoints in the re-treatment extension with cadazolid are captured and determined in the same manner as the efficacy endpoints in the randomized blinded part of the study.

AEs in the re-treatment extension with cadazolid are captured and analyzed in the same manner as AEs in the randomized blinded part of the study. A TEAE is any AE from study drug initiation until 7 days after the last dose of study drug [see Section 10.1.1]. AEs that started during the re-treatment extension with cadazolid screening period are recorded as AEs in the main study. All AEs are reported as part of the re-treatment extension with cadazolid after the first dose of open-label study drug. AEs ongoing at the time of first dose of open-label cadazolid and worsening thereafter must be reported as new AEs.

All re-treatment endpoints are exploratory.

Table 3 Visit and assessment schedule for the re-treatment extension with cadazolid

PERIODS	Name	SCREENING	TREATMENT			FOLLOW-UP			-
	Duration	Up to 24 hours	10 to 11 days			28 to 32 days			-
VISIT	Number	Re-1		Re-2	Re-3	Re-4	Re-4.a, Re-4.b,…	Re-5	Re-U1, Re-U2, …
	Name	Eligibility	Re-treatment kit delivery				NED		Unscheduled
EVALUATION	Time	Day –1 to 1 (within 24 h)	Day 1	Day 5 or 6 *On site or by phone*	Day 8 to 10 *incl. Premature Disc.*	*EOT + 2 to 4 days On site or by phone*	*If applicable, in case of NED*[4]	*EOT + 28 to 32 days or Study Withdrawal*	*If applicable*
Informed consent		X							
Fecal sampling for microbiology		X			X		X		
***C. difficile* GDH and toxin test**		within 72 h					X		
Concomitant medications		X		X	X	X	X	X	X
Physical examination		X			X		X	X	X
Body weight		X			X			X	X
Vital signs / Body temperature		X			X		X	X	X
Hematology/coagulation/ chemistry		X[1]			X		X	X	X
12-lead ECG		X			X			X	X
Re-treatment kit delivery			X						
Study Drug intake			X[2]	and Study Drug Journal completion					
Stool Diary			←					→	
CDAD DaySyms PRO Diary		←				→			
Interview (face to face or by phone)			Daily[5]				Twice weekly		
Investigator assessment				X		X		X	
WPAI:CDAD		X			X			X	
AEs/SAEs[3]			X	X	X	X	X	X	X

1. Includes pregnancy test for women of childbearing potential. **2.**After re-treatment kit delivery. **3.** All AEs and SAEs are reported after the first dose of study drug. Between informed consent and first dose of study drug, AEs and SAEs are reported as part of the main study. **4.** All assessments must be performed in the case of a recurrence. **5.** Interviews start on Day 2.
AE = adverse event, *C-difficile* = *Clostridium difficile*, CDAD = *Clostridium difficile*-associated diarrhea, ECG = electrocardiogram, EOT = end-of-treatment, GDH = glutamate dehydrogenase, NED = new episode of diarrhea, PRO = patient reported outcomes, SAE = serious adverse event, WPAI = work productivity and activity impairment.

9 STUDY COMPLETION

All randomized subjects who received study drug must be followed up to Visit 5, whether or not they are prematurely discontinued from study treatment. Randomized subjects who do not receive a dose of study drug will complete Visit 5 as soon as possible.

Subjects are considered to have completed the study if Visit 5 or Re-5, in case of re-treatment extension with cadazolid, is performed at least 28 days after last dose of study drug.

9.1 Withdrawal from study

A subject will be considered as withdrawn from the study if he/she (or the physician on behalf of the subject) withdraws consent to be treated **and** to be followed up. A subject who prematurely discontinues study drug is **not** considered as withdrawn from the study, and will be followed up as outlined in Section 8.3, unless the subject withdraws from the study.

A subject will also be considered as withdrawn from the study if he/she dies, or is lost to follow-up.

The site must take preventive measures to avoid a subject being lost to follow-up (i.e., document in subject's notes and CRF different ways to contact subjects, including telephone number, home address, e-mail address, person to be contacted in case the subject cannot be reached). If the subject cannot be reached then the site must make a reasonable effort to contact the subject and document in subject's notes and CRF all attempts. The following methods must be used: at least 3 telephone calls must be placed to the last available telephone number and 1 registered letter must be sent by mail to the last available home address. Additional methods may be acceptable if they are compliant with local rules/regulations (e.g., visit to the subject's home) and respect the subject's right to privacy.

If the subject is still unreachable after all contact attempts listed above, then he/she will be considered to be lost to follow-up.

9.2 End of the trial

The end of the trial occurs when all the subjects have completed Visit 5 or Re-5 for those participating in the re-treatment extension with cadazolid.

9.3 Premature termination or suspension of the study

Actelion reserves the right to terminate the study at any time globally or locally. Investigators can terminate the participation of their site in the study at any time.

If the study is prematurely suspended or terminated, then Actelion will promptly inform the investigators, the IRBs/IECs and health authorities as appropriate, and provide the reasons for the suspension or termination.

If the study is prematurely suspended or terminated for any reason, then the investigator, in agreement with Actelion, must promptly inform all enrolled subjects, and ensure their appropriate treatment and follow-up.

In addition, if the investigator suspends or terminates the study without prior agreement from Actelion, then the investigator must promptly inform Actelion and the IRB/IEC, and must provide Actelion and the IRB/IEC with a detailed written explanation of the termination or suspension.

If the IRB/IEC suspends or terminates its approval / favorable opinion of the study, then the investigator must promptly notify Actelion and provide Actelion with a detailed written explanation of the termination or suspension.

Any premature suspension or termination of the study must be discussed with the IDMC and the Steering Committee.

9.4 Medical care of subjects after the End-of-Treatment

When a subject is declared a Clinical Failure, or at the time of recurrence if the subject does not participate in the re-treatment extension with cadazolid, the investigator/delegate will explain to subjects what treatment / medical care would be necessary and available according to local practice and regulation.

10 SAFETY DEFINITIONS AND REPORTING REQUIREMENTS

10.1 Adverse events

10.1.1 Definitions of adverse events

An AE is any adverse change from the subject's baseline condition, i.e., any unfavorable and unintended sign, including an abnormal laboratory finding, symptom or disease, that occurs during the course of the study, whether or not considered related to the study drug.

A TEAE is any AE from study drug initiation until 7 days after the last dose of study drug whether or not considered related to the study drug. See also Section 8.4 for AEs during the re-treatment extension.

AEs include:

- Exacerbation of a pre-existing disease.
- Increase in frequency or intensity of a pre-existing episodic disease or medical condition.

- Disease or medical condition detected or diagnosed after study drug administration even though it may have been present prior to the start of the study.
- Continuous persistent disease or symptoms present at baseline that worsen following the start of the study.
- Abnormal assessments e.g., change on physical examination, ECG findings, if they represent a clinically significant finding that was not present at baseline or worsened during the course of the study.
- Laboratory test abnormalities if they represent a clinically significant finding, symptomatic or not, which was not present at baseline or worsened during the course of the study or led to dose reduction, interruption or permanent discontinuation of study drug.
- Overdose, misuse and abuse of the study drug should be reported as AEs and errors with the study medication should be documented in the study drug log of the CRF.

10.1.2 Intensity of adverse events

The intensity of clinical AEs is graded on a three-point scale – mild, moderate, severe – and is reported on specific AE pages of the CRF.

If the intensity of an AE worsens during study drug administration, only the worst intensity should be reported on the AE page. If the AE lessens in intensity, no change in the severity is required.

The 3 categories of intensity are defined as follows:

Mild
The event may be noticeable to the subject. It does not influence daily activities, and usually does not require intervention.

Moderate
The event may make the subject uncomfortable. Performance of daily activities may be influenced, and intervention may be needed.

Severe
The event may cause noticeable discomfort, and usually interferes with daily activities. The subject may not be able to continue in the study, and treatment or intervention is usually needed.

A mild, moderate, or severe AE may or may not be serious [see Section 10.3.1]. These terms are used to describe the intensity of a specific event. Medical judgment should be made on a case per case basis.

Seriousness, rather than severity assessment, determines the regulatory reporting obligations.

10.1.3 Relationship to study drug

Each AE must be assessed by the investigator as to whether or not there is a reasonable possibility of causal relationship to the study drug, and reported as either 'suspect' (i.e., related) or 'not suspect' (i.e., not related).

10.1.4 Reporting of adverse events

All AEs occurring after study drug initiation and up to Visit 5 (30 ± 2 days after last dose of study drug) must be recorded on specific AE pages of the CRF.

10.1.5 Follow-up of adverse events

If an AE is still ongoing at the End-of-Study (EOS), then the AE must be followed until it is stable or no longer clinically significant.

10.2 Serious adverse events

10.2.1 Definitions of serious adverse events

10.2.1.1 Serious adverse events

An SAE is defined by the ICH guidelines as any AE fulfilling at least one of the following criteria:

- Fatal
- Life-threatening: refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event that hypothetically might have caused death had it been more severe
- Requiring inpatient hospitalization, or prolongation of existing hospitalization
- Resulting in persistent or significant disability or incapacity
- Congenital anomaly or birth defect
- Medically significant: refers to important medical events that may not immediately result in death, be life-threatening, or require hospitalization but may be considered to be SAEs when, based upon appropriate medical judgment, they may jeopardize the subject, and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions above.

The following reasons for hospitalizations are exempt from being reported:

- Hospitalizations for cosmetic elective surgery, or social and/or convenience reasons
- Hospitalizations for pre-planned standard monitoring of a pre-existing disease or medical condition that did not worsen (e.g., hospitalization for coronary angiography in a subject with stable angina pectoris)
- Hospitalizations for elective treatment of a pre-existing disease (present at baseline) or medical condition that did not worsen (e.g., elective hip replacement for arthritis)

However, complications that occur during hospitalization are AEs or SAEs (for example, if a complication prolongs hospitalization).

10.2.1.2 Serious adverse events associated to study design or protocol mandated procedures

An SAE is defined as related to study design or protocol-mandated procedures if it appears to have a reasonable possibility of a causal relationship to either the study design or to protocol-mandated invasive procedures. For example, a complication of an invasive procedure specifically required by the protocol.

10.2.2 Reporting of serious adverse events

10.2.2.1 During screening period

All SAEs, occurring after signing the ICF must be reported (whether considered associated or not associated to study design or study-mandated procedures).

These SAEs must be reported on an SAE form and in the CRF (AE section).

10.2.2.2 During treatment period

All SAEs, regardless of causal relationship, must be reported.

These SAEs must be reported on an SAE form and in the CRF (AE section).

10.2.2.3 During follow-up period

All SAEs, regardless of causal relationship, which occur within 30 days after study drug discontinuation must be reported.

These SAEs must be reported on an SAE form and in the CRF (AE section).

All SAEs must be reported to the IRB/IEC according to the site-specific IRB/EC reporting requirements. Copies of all communication regarding SAEs/pregnancy to the IRB/IEC and to the sponsor must be appropriately filed in the ISF.

10.2.3 Follow-up of serious adverse events

SAEs still ongoing at EOS must be followed up until resolution or stabilization, or until the event outcome is provided (e.g., fatal outcome).

10.2.4 After the follow-up period

New SAEs occurring after the follow-up period must be reported to the Actelion Drug Safety Department within 24 h of the investigator's knowledge of the event, **only** if considered causally related to previous exposure to the study medication by the investigator.

10.2.5 Reporting procedures

All SAEs must be reported by the investigator to the Actelion Drug Safety Department within 24 h of the investigator's first knowledge of the event.

All SAEs must be recorded on an SAE form, irrespective of the study drug received by the subject and irrespective whether or not this event is considered by the investigator to be related to study drug.

The SAE forms must be faxed to the Actelion Drug Safety Department (contact details are provided on the SAE form). The investigator must complete the SAE form in English, and must assess the causal relationship of the event to study drug.

Follow-up information about a previously reported SAE must also be reported within 24 h of receiving it. The Actelion Drug Safety Department may contact the investigator via email and/or fax to obtain further information.

If the subject is hospitalized in a hospital other than the study site, it is the investigator's responsibility to contact this hospital to obtain all SAE-relevant information and documentation.

10.3 Pregnancy

10.3.1 Reporting of pregnancy

Irrespective of the treatment received by the subject, any pregnancy detected during study drug administration and up to 30 days following the last dose of study drug, must be reported within 24 hours of the investigator's knowledge of the event.

Pregnancies must be reported on the Actelion Pregnancy form and faxed to the Actelion Drug Safety Department (see contact details provided on the pregnancy form). Pregnancies must be reported on an AE page of the CRF.

If a pregnancy is detected while on study drug, then permanent discontinuation of study drug must be considered. The investigator must counsel the subject and discuss the risks of continuing with the pregnancy and the possible effects on the fetus.

10.3.2 Follow-up of pregnancy

Any pregnancy must be followed to its conclusion and its outcome must be reported to the Actelion Drug Safety Department.

For any ongoing pregnancy, investigator should provide follow-up information at the beginning of each trimester.

10.4 Study safety monitoring

Blinded clinical study safety information (e.g., AEs, SAEs, laboratory values, ECGs, vital signs, and project-specific labs/examinations as required) is monitored and reviewed on a continuous basis by Actelion to ensure subjects' safety as well as data quality. These activities are complementary to the IDMC's activities, which include monitoring safety and efficacy data periodically in an unblinded fashion [see Section 3.3].

11 STATISTICAL METHODS

All statistical analyses will be conducted by Actelion or by designated CROs supervised by Actelion.

A Statistical Analysis Plan (SAP) will provide full details of the analyses, data displays, and algorithms to be used for data derivations. Statistical analysis of pharmacokinetic endpoints will be described in a separate statistical analysis plan.

For subjects entering into the re-treatment extension, only data collected prior to starting re-treatment with cadazolid (first re-treatment dose) will be considered for all variables specified below, unless specifically mentioned as in Section 8.4. For subjects entering the re-treatment extension with cadazolid, a similar definition will be applied to estimate the exploratory endpoints Clinical Cure, recurrence and Sustained Cure assessed during the re-treatment extension with cadazolid.

11.1 Variables

11.1.1 Primary variable

CCR (%) is the variable to be analyzed for the primary endpoint Clinical Cure, as defined in Section 6.1.1 and will be derived by the sponsor. CCR is the proportion of subjects assessed as meeting the criteria for Clinical Cure, where Clinical Cure is defined for a subject meeting both the following:

- ROD (≤ 3 UBMs per day for at least 2 consecutive days) on study treatment and maintained for 2 days after EOT

  AND

- No additional antimicrobial treatment active against CDAD or FMT between first dose of study drug and 2 days after EOT (inclusive).

ROD is determined from the number of UBMs, or if the exact number is missing, from presence or absence of > 3 UBMs per day recorded in the Daily Stool Information section of the CRF. Days are measured according to calendar days.

Antimicrobial treatment active against CDAD or FMT is determined from concomitant medication/procedure CRF pages.

Subjects who do not fulfill the requirements for Clinical Cure are considered Clinical Failure.

11.1.2 Secondary variables

11.1.2.1 Sustained cure rate (SCR)

SCR (%) is the variable to be analyzed for the secondary endpoint Sustained Cure [as defined in Section 6.1.2], and will be derived by the sponsor. SCR is the proportion of subjects assessed as meeting the criteria for Sustained Cure, where Sustained Cure is defined for each subject having Clinical Cure and no recurrence.

Recurrence is defined for subjects with Clinical Cure as:

- NED (> 3 UBMs on any day between 3 days after EOT and Visit 5)

  AND

- Stool test showing positive *C. difficile* GDH and toxin test on the same sample (according to EIA tests approved by the sponsor)

  AND

- Antimicrobial treatment active against CDAD (including participation in the re-treatment extension with cadazolid) or FMT started between 3 days after EOT and Visit 5).

NED is determined from the number of UBMs, or if the exact number is missing from presence or absence of > 3 UBMs, per day collected in the Daily Stool Information section of the CRF. Days are measured according to calendar days.

A positive stool test is determined from the corresponding unscheduled visit for NED, where the result of the test is recorded.

Antimicrobial treatment active against CDAD or FMT is determined from concomitant medication/procedure CRF pages. Participation in the re-treatment extension is derived from enrollment into the re-treatment extension as recorded in the CRF.

NED should precede both the positive *C. difficile* GDH and toxin test and antimicrobial treatment active against CDAD (including participation in the re-treatment extension with cadazolid) or FMT. To avoid temporal dissociation between NED and the positive *C. difficile* GDH and toxin test and treatment of recurrence, at least one day with > 3 UBMs must be present within a window of 4 days prior to (and including) either the date of the fecal sampling for the GDH and toxin test or the start date of antimicrobial

treatment active against CDAD or the date of FMT. If the date of fecal sampling or start date of antimicrobial treatment active against CDAD are not the same, it is enough that at least one of these time windows contain a day with > 3 UBMs to confirm a recurrence.

If a subject has missing information on UBMs within the window 4 days prior to and including the date of fecal sample–or antimicrobial treatment, the rules governing the handling of missing UBMs described in the SAP will be applied.

If a fecal sample for the GDH and toxin test is taken or antimicrobial treatment active against CDAD is started before a NED, it will not be considered a recurrence. Fecal sampling for the GDH and toxin test should be prior to or on the start date of antimicrobial treatment or FMT. Fecal sampling for the GDH and toxin test taken 1 or more days after the start date of a antimicrobial treatment active against CDAD or FMT are not considered in determining recurrence.

Where multiple GDH and toxin tests are performed during a period with daily number of UBMs > 3, the latest fecal sample for the GDH and toxin test prior to (and including) antimicrobial treatment is selected. Where > 3 UBMs occur on 1 or more days within the 4 day window prior to (and including) start date of antimicrobial treatment and the most recent GDH or toxin test is negative and performed on a fecal sample collected prior to this 4 day window, the negative GDH or toxin test is not considered and the subject will be considered to have a recurrence.

Subjects with Clinical Failure or with Clinical Cure and recurrence are considered to be without Sustained Cure.

11.1.2.2 Time to Resolution of Diarrhea (ROD)

Time to ROD is defined as the time (h) elapsed between the first dose of study drug and the time when ROD is considered achieved.

Time when ROD is considered achieved is the time of the last UBM prior to the first 2 consecutive 24 h periods with ≤ 3 UBMs and subsequently ≤ 3 UBMs per day maintained up to 2 days after EOT. The two consecutive 24 h periods and last UBM date/time prior that period is determined from the Stool Log section of the CRF. The condition of maintaining ≤ 3 UBMs per calendar day is determined based on the number of UBMs per day as described for Clinical Cure [Section11.1.1].

Subjects without Clinical Cure are censored at EOT day. Similarly, subjects who have withdrawn from the study, died, or are lost to follow-up before EOT + 2 days are censored at the expected EOT day, i.e., 10 days after starting medication. This will constitute a right-censored observation.

11.1.2.3 Absolute change from baseline in CDAD DaySyms PRO total daily score

The absolute change in CDAD DaySyms PRO total daily score will be derived as the difference between the post-baseline and the baseline CDAD DaySyms PRO total daily score for each day. Data from all available days will be used for the analysis, fitting a general mixed model for repeated measurements [see Section 11.3.4.3]. Comparison between cadazolid and vancomycin will be made up to Day 12.

The exact definition of the CDAD DaySyms PRO total score will be determined in the psychometric validation of the CDAD DaySyms PRO conducted in parallel, based on a blinded analysis of the sub-study for psychometric validation of AC-061A301 and study AC-061A302.

11.1.2.4 Other efficacy variables

11.1.2.4.1 Investigator's assessment of Early Treatment Response at Visit 2

The variable is defined as the ETR as assessed by the investigator at Visit 2 as improved, unchanged or worsened compared to baseline.

11.1.2.4.2 Investigator's judgment of Clinical Response Rate (ICRR) at Visit 4

ICRR (%) is the proportion of subjects cured at Visit 4 based on the investigator's judgment of Clinical Response.

11.1.2.4.3 Investigator's judgment of Sustained Response Rate (ISRR)

ISRR (%) is the proportion of subjects with a Sustained Cure at Visit 5 based on the investigator's judgment of Sustained Response.

11.1.2.4.4 Early Clinical Cure Rate (ECCR) by Day 5

ECCR (%) is the proportion of subjects assessed as meeting the criteria for ECC by Day 5, where ECC is defined for a subject meeting both the following:

- ROD by Day 5: ≤ 3 UBMs for at least 2 consecutive days prior to or including Day 5 and maintained up to EOT + 2 days
- No additional antimicrobial treatment active against CDAD or FMT between first dose of study drug and EOT + 2 days as defined in Section 6.1.1

ROD by Day 5 is determined from the number of UBMs in the same way as for Clinical Cure [Section 11.1.1]. To be considered with Early Clinical Cure the subject must have ≤ 3 UBMs per day from Day 4 up to EOT + 2 days. If the number of UBMs on one or more days is missing or 'unknown' then the subject is considered a Failure.

11.1.2.4.5 Normalization of Bowel Movements rate (NBMR)

It is the proportion of subjects with NBM, where NBM is defined as:

- ≤ 2 bowel movements per day and no UBM on at least 2 consecutive days up to EOT and maintained up to 2 days after last dose of study drug

  AND

- No additional antimicrobial treatment active against CDAD or FMT between first dose and EOT + 2 days after EOT.

The number of bowel movements and UBMs are determined from the Daily Stool Information of the CRF.

Additional antimicrobial treatment active against CDAD or FMT is derived as defined in Section 11.1.1.

11.1.2.4.6 Time to return to usual stools

Time to return to usual stools is defined as the time (days) elapsed between the first dose of study drug and the first day where the stools have returned to usual for a subject, i.e., the number of bowel movements (including UBMs) per day ≤ number of bowel movements reported as usual by the subject and maintained for at least 3 calendar days. Number of usual bowel movements is reported by the subject at baseline and recorded in the CRF. Number of bowel movements per day is determined from the Daily Stool Information section of the CRF (where days are measured according to calendar days) between date of first dose up to Visit 5.

Subjects without return to usual stools as defined above are censored at Visit 5. Subjects without a Visit 5 assessment who died, withdrew from the study or are lost to follow-up, are censored at the date of death, study withdrawal or last contact, respectively.

11.1.2.4.7 Recurrence Rate

Recurrence Rate (RR, %) is the proportion of subjects assessed as having a recurrence [see Section 11.1.2] out of subjects meeting the criteria for Clinical Cure.

Recurrence is classified either as:

- Relapse: Identical strains identified either by REA typing or ribotyping at new episode of diarrhea (Visit 4.a, 4.b, etc.) and baseline .
- Re-infection: Different strains identified either by REA typing or ribotyping at new episode of diarrhea (Visit 4. a, 4.b, etc.) and baseline

11.1.2.4.8 Time to recurrence

Defined for subjects meeting the criteria of recurrence as the period (h) between the time of the last dose of study drug and the onset time of NED determined to be recurrence. Onset time of NED is defined as the time of the first UBM recorded in the Stool Log of the CRF in the day of NED (first day with > 3 UBMs between EOT + 3 days and Visit 5 date as recorded in the Daily Stool Information section of the CRF) determined to be recurrence. Subjects considered Sustained Cure, i.e., clinically cured but without establishment of recurrence or death, are censored at the Visit 5. Subjects who died after EOT + 3 days are censored at the time of death. Subjects who are lost to follow-up without a Visit 5, are censored at the last contact date.

Subjects not clinically cured and not at risk for a recurrence are not considered for this endpoint.

11.1.2.4.9 Change from baseline in CDAD DaySyms PRO scores

- Absolute (and/or percent) change from Baseline in CDAD DaySyms PRO total score per day.
- Absolute (and/or percent) change from baseline in each of the domain score(s) per day.

11.1.2.4.10 Susceptibility of C. difficile (MIC50 and MIC90, in μg/mL)

Susceptibility is defined as the minimum inhibitory concentration (MIC in μg/mL) of the test agent (cadazolid, vancomycin, linezolid, moxifloxacin, metronidazole and fidaxomicin) which inhibits bacterial growth:

- At baseline
- In case of Clinical Failure, as the MIC of *C. difficile* from a fecal sample collected at Visit 3
- In case of Recurrence as the maximum MIC of *C. difficile* in fecal samples collected at any Visit 4.a, 4.b, etc.

11.1.2.4.11 Change from baseline in susceptibility of C. difficile

This variable is measured only in subjects with Clinical Failure or recurrence. It is defined as the relative change in MIC of cadazolid, vancomycin, linezolid and moxifloxacin at post-baseline compared to baseline: MIC at post-baseline / MIC at baseline, categorized as ≥ 4-fold decrease, 2-fold decrease, no change, 2-fold increase or ≥ 4-fold increase. The MIC at post-baseline is defined for Clinical Failures and recurrence as specified in Section 11.1.2.4.10.

11.1.2.4.12 Change from baseline to Visit 3 in VRE count

It is defined as the difference between baseline and Visit 3 VRE counts.

The Central Laboratory provides density of VRE counts (log10 cfu/mL) at Visit 1 and Visit 3 for all subjects. VRE counts are defined as the sum of counts for isolates identified as vancomycin-resistant *Enterococcus faecium* and vancomycin-resistant *Enterococcus faecalis*.

11.1.2.4.13 Susceptibility of VRE to cadazolid, vancomycin, linezolid, moxifloxacin, fidaxomicin, daptomycin, tigecycline, ampicillin, gentamicin, and quinupristin-dalfopristin

Susceptibility for vancomycin-resistant *E. faecium* and vancomycin-resistant *E. faecalis* isolates is defined as the minimum inhibitory concentration (MIC in μg/mL) of the test agent (cadazolid, vancomycin, linezolid, moxifloxacin, fidaxomicin, daptomycin, tigecycline, ampicillin, gentamicin, and quinupristin-dalfopristin) which inhibits bacterial growth.

11.1.3 Safety variables

- Deaths up to Visit 5
- AEs and SAEs up to Visit 5
- TEAEs and treatment-emergent SAEs up to 7 days after EOT, day inclusive
- AEs leading to premature discontinuation of study treatment
- Change from baseline up to Visit 3 and Visit 5 in vital signs (SBP, DBP, and HR), body temperature and body weight
- Marked abnormalities in vital signs (SBP and DBP) up to EOT + 7 days (treatment emergent) and up to Visit 5
- Change from baseline in ECG parameters up to Visit 3 and Visit 5
- Marked abnormalities in ECG up to EOT + 7 days (treatment-emergent) and up to Visit 5
- Change from baseline in hematology, coagulation, and chemistry parameters up to Visit 3 and Visit 5
- Marked abnormalities in hematology, coagulation and blood chemistry parameters up to EOT + 7 days (treatment-emergent) and up to Visit 5.

11.1.4 Other variables

11.1.4.1 Exposure to study drug

- The duration of exposure is defined as the time elapsing between study drug initiation and discontinuation, inclusive. This will be determined for cadazolid and vancomycin for double-blind treatment and for re-treatment with cadazolid (open-label treatment).
- The mean daily dose per subject is defined as the ratio between the total study drug dose taken during the treatment period and the total exposure time.

- Compliance (%) is defined as percent of doses taken out of planned doses (20 sachets for cadazolid, and 40 capsules for vancomycin) and determined separately for sachets and capsules.

11.1.4.2 Demographic variables

- Country.
- Sex.
- Age categories.
- BMI.
- Race and Ethnicity.
- Severe CDAD is defined as any **one** of the following:
  a. Maximum core temperature recorded at baseline > 38.5 °C.
  b. WBC > 15.0×10^9/L (based on Central Laboratory).
  c. Rise in serum creatinine > 50% compared to pre-CDAD diagnosis.

11.1.4.3 Baseline disease characteristics variables

- Prior antimicrobial treatment active against CDAD.
- First occurrence / first recurrence.
- Initial strain of CDAD (hypervirulent or not).
- In-patient or out-patient at randomization.
- Baseline frequency of UBMs.

11.1.4.4 Pharmacoeconomic variables

11.1.4.4.1 Hospitalization

- Length of stay in hospital (days)
- Length of stay by ward unit (general ward, step-down/intermediate unit, ICU [days])
- Frequency of re-admissions/admissions to hospital after start of treatment
- Frequency of emergency department visits after start of treatment

11.1.4.4.2 Change from baseline in WPAI:CDAD scores

Absolute WPAI:CDAD scores and absolute change from baseline for each WPAI:CDAD score at/to each visit where the WPAI:CDAD questionnaire was administered will be summarized.

WPAI:CDAD scores are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity. The 4 WPAI:CDAD scores are defined as follows, based on the scores Q1–Q6 of Questions 1–6:

- Absenteeism: Percent work time missed due to CDAD = Q2 / (Q2+Q4) * 100
- Presenteesism: Percent impairment while working due to CDAD = Q5/10 *100
- Work productivity loss: Percent overall work impairment due to CDAD = Q2/(Q2+Q4) + [(1-Q2/(Q2+Q4)) * (Q5/10)] * 100
- Activity Impairment: Percent activity impairment due to CDAD Q6/10 * 100

11.1.4.5 Pharmacokinetic variables

Cadazolid plasma concentration 2 h post-dose at Visit 3.

11.1.4.6 Re-treatment extension with cadazolid variables

Variables for the re-treatment extension with cadazolid will be defined in the same way as for the corresponding variables defined for the treatment and follow-up periods of the randomized blinded part of the study and will be defined and fully described in the SAP.

11.2 Analysis sets

The following analysis sets are defined.

11.2.1 Screened analysis set (SCRAS)

This analysis set includes all subjects screened even if they were not eligible.

11.2.2 Full analysis set (FAS)

The Full analysis set (FAS) includes all subjects allocated to a study treatment based on the randomization-assigned treatment.

11.2.3 Modified intent-to-treat (mITT) analysis set

The mITT analysis set includes subjects in the FAS who received at least one dose of study drug and had a confirmed diagnosis of CDAD; i.e.,

- > 3 UBMs in the 24 h prior to randomization.
- Positive GDH and toxin test (EIA approved by the sponsor) on the same stool sample produced in the 72 h (48 h for MTFs) period prior to randomization.

It is based on the randomization-assigned treatment.

11.2.4 Per-protocol analysis set (PPS)

The PPS comprises all subjects from the mITT analysis set without major protocol violations or conditions that might affect the evaluation of the effect of the study drug on

the primary endpoint. The following conditions will lead to the exclusion of a subject from the PP analysis:

At study entry:

- More than one previous episode of CDAD in the 3-month period prior to randomization
- Fulminant or life threatening CDAD
- Concurrent immediately life-threatening disease or condition
- History of inflammatory colitides, chronic abdominal pain, or chronic diarrhea, or known positive diagnostic test for enteropathogens
- Antimicrobial treatment active against CDAD > 24 h (except for MTFs)
- Any investigational drug to prevent or treat CDAD

During study:

- Any prohibited concomitant medication, except treatment with antimicrobial medication active against CDAD or FMT or any investigational therapy to treat CDAD after randomization and up to and including 2 days after EOT [see Section 5.2]
- Insufficient course of therapy: Subjects classified as a Clinical Failure with less than 6 doses of cadazolid or 12 doses of vancomycin. Subjects classified as a Clinical Cure with less than 16 doses of cadazolid or 32 doses of vancomycin
- Received study treatment differed from randomized study treatment
- Subject was unblinded before Database Lock
- Insufficient information to determine Clinical Cure
- Positive diagnostic test for enteropathogens during treatment period.

It is estimated that approximately 15% of the randomized subjects will not be part of the PPS.

11.2.5 Hypervirulent analysis set (HVAS)

The Hypervirulent analysis set (HVAS) includes subjects in the mITT analysis set who are confirmed with CDAD due to a hypervirulent strain at baseline.

11.2.6 PRO analysis set (PROAS)

The PRO analysis set includes subjects in the mITT analysis set who have signed the ICF themselves. It is based on the assigned treatment rather than the actual received treatment.

11.2.7 Safety set (SS)

The Safety Set (SS) includes all randomized subjects who received at least one dose of study drug, based on the actual treatment received.

11.2.8 Re-treatment extension with cadazolid Set (ES)

The Re-treatment extension with cadazolid set (ES) includes all subjects enrolled in the re-treatment extension and received at least one dose of open-label cadazolid.

11.2.9 Pharmacokinetic analysis set (PKS)

This analysis set comprises all subjects who provide an evaluable plasma cadazolid concentration (quantifiable or BLQ) at approximately 2 h post-dose on Day 8–10 (Visit 3).

11.2.10 Role of the different analysis sets

Subject disposition and study completion/discontinuation, including reason for screening failure, will be summarized for the SCRAS. Protocol violations, baseline demographic and disease characteristics, and previous/concomitant medications, will be summarized in the FAS.

The main analysis of NI for the primary variable will be performed on the PPS and on the mITT. All other efficacy analyses will be performed on the mITT analysis set. The analysis of relevant secondary endpoints may be repeated on the PPS for sensitivity.

The HVAS will be used in the meta-analysis, described in Section 11.4. The SS will be used for the analyses of the safety variables and treatment exposure.

Analyses of variables based on the CDAD symptoms diary will be performed on the PROAS (excluding subjects who participate in the PRO sub-study). Analyses of WPAI:CDAD questionnaire will be performed on the PROAS. Analyses of endpoints for the extension treatment will be performed on the ES. Analyses of pharmacokinetic endpoints will be analyzed on the PKS.

Listings will be prepared on the FAS, unless specified otherwise.

11.2.11 Randomization and stratification

Approximately 630 subjects will be randomized 1:1 to the 2 treatment groups (315 cadazolid and 315 vancomycin), to ensure that at least 536 subjects will be available in the PPS and 598 in the mITT analysis set. Randomization will be stratified on CDAD first occurrence or first recurrence and on study site, using the IVRS system.

11.3 Description of statistical analyses

11.3.1 General considerations

- Unblinding of study drug code for the data analysis will occur after database closure in accordance with Actelion SOPs.
- The experimentwise significance level will be controlled at the overall two-sided $\alpha = 0.05$
- Covariates will include stratification factor first occurrence and first recurrence and geographical region. It is anticipated that the number of subjects enrolled at some sites may be small. Therefore the sites will be pooled considering geographical region, pooling first according to country of the site. Countries may be further combined according to geographical region to ensure sufficient patients in each strata for the analyses. Regions may include North America, South America, Western Europe, Eastern Europe, Asia. Details for the pooling of sites will be defined in the SAP. Other covariates/subgroups to be explored include the following anthropometric and baseline characteristics: sex, race, age category, baseline frequency of UBMs, in-patient status, initial severity of the disease, prior antimicrobial treatment active against CDAD, and initial strain of CDAD (hypervirulent or not).
- AEs and medical history will be coded according to MedDRA dictionary (Version 16 or more recent version if available). Concomitant medications will be coded according to the WHO drug code and the anatomic therapeutic chemical (ATC) class code (Version 01Mar2010 or later).
- SAS version 9.2 or higher will be used for all the statistical analysis.
- All analysis variables will be listed and presented in tables; variables to be presented in figures will be specified in the SAP.
- Data will be listed and summarized by appropriate descriptive statistics (tables or figures), typically including:
  - Number of non-missing observations, number of missing observations, mean, standard deviation (SD), minimum, Q1, median, Q3 and maximum for continuous variables
  - Number of events, number of censored observations and Kaplan-Meier estimates of the survival function will be provided for time-to-event variables.
  - Number of non-missing observations, number of missing observations and frequency with percentage per category (percentages based on the sum of number of non-missing observations and total number of observations) for categorical variables.
  - For susceptibility data: Number of non-missing observations, number of missing observations, geometric mean, median (MIC50), 90% quantile (MIC90), minimum, maximum.

Absolute changes from baseline are defined as: post-baseline value minus baseline value, i.e., a positive sign indicates an increase as compared to baseline.

A percentage change from baseline is defined as the absolute change from baseline divided by the baseline value (if the baseline value > 0) and then multiplied by 100.

For susceptibility (MIC) endpoints change from baseline is defined as: post-baseline value divided by baseline value.

Handling of dropouts and missing data
This section describes the concepts to be used. Further details for handling of missing data and missing dates and times will be described in the SAP. Additional imputations methods may be specified in the SAP to assess sensitivity of analyses to the imputation methods used.

In general, summaries and analyses will be based upon observed data i.e., without imputation of missing values; exceptions to this rule for the most important variables are described below.

- **Clinical Cure**

In case of death for any cause, or lost to follow-up, or study withdrawal prior to EOT + 2 days, the subject is considered a Clinical Failure.

- **Sustained Cure**

In case of death for any cause, study withdrawal or lost to follow-up from EOT + 3 days and prior to EOT + 28 days, the subject is considered not a Sustained Cure.

11.3.2 Analysis of subject disposition and study completion / withdrawal

The number and percentage of subjects in the different analysis sets will be summarized and listed together with reasons for exclusion from the relevant set. The number and percentage of subjects screened, entered and who completed the study or withdrew prematurely (by reason of withdrawal) will be presented in tables and listings. Summaries will be presented by treatment group and overall for the SCRAS. Similarly, number and percentage of subjects receiving extension treatment will be presented.

11.3.3 Analysis of the primary variable

11.3.3.1 Hypotheses and statistical inference

The objective of the study is met if it is demonstrated that the CCR for cadazolid (CCRc) is not inferior to the CCR for vancomycin (CCRv), accounting for a NI margin of 10%. The justification of the choice of 10% a NI margin is detailed in Appendix 7.

The main analysis will be done on the mITT analysis set and on the PPS: NI must be shown in the mITT analysis set and in the PPS to claim NI of cadazolid versus vancomycin. A one-sided 0.025 significance level is applied in both analysis sets.

The null hypothesis is: $H_0^{(1)}$: CCRc − CCRv ≤ −10%,
and the alternative: $H_1^{(1)}$: CCRc − CCRv > −10%.

If the lower bound of a two-sided 95% CI of the difference between proportions is greater than −10%, the NI of cadazolid versus vancomycin for Clinical Cure will be demonstrated.

The NI assessment on the mITT analysis set and the PPS will compose the first part of the hierarchical testing procedure [Figure 3].

11.3.3.2 Statistical model

No statistical model is defined – the analysis is based on the CI of the crude difference in proportions.

11.3.3.3 Main analysis

The main analysis is carried out as described in Section 11.3.3.1. The CCR will be summarized per treatment group. Two-sided 95% confidence intervals (CIs) for the difference of cure rates will be estimated using the Wilson's Score method for the difference of proportions [Newcombe 1998].

The main analysis will be conducted on the mITT analysis set and the PPS.

Imputation methods for the substitution of incomplete, missing, or potentially biased data are defined in Section 11.3.1, but will be more detailed in the SAP.

11.3.3.4 Supportive/sensitivity analyses

A stratified Cochran-Mantel-Haenszel analysis will be applied as supportive analysis stratified by first occurrence and first recurrence (IVRS) and geographical region based on the PPS and mITT analysis set. For the average risk difference the Cochran-Mantel-Haenszel estimate and corresponding CI will be derived using normal approximation.

Consistency of results on CCR will be explored over different subgroups defined by anthropometric and baseline characteristics: first occurrence and first recurrence (IVRS), geographical region, sex, race, age category, baseline frequency of UBMs, in-patient status, initial severity of the disease, presence of prior antimicrobial treatment active against CDAD and initial strain of CDAD (hypervirulent or not).

The CCR will be summarized by treatment group per subgroup with 95% CIs for the difference of cure rates estimated using the Wilson's Score method [Newcombe 1998].

For each anthropometric and baseline characteristic a logistic regression will be applied on the CCR, including the treatment group, the baseline characteristic and the interaction of both of them. Treatment effects in each subgroup will be reported with odds ratios (cadazolid/vancomycin) and 95% CI for each baseline characteristic together with the overall treatment effect and 95% CI, and will be presented in a Forest plot [Cuzick 2005]. An exploratory test for the interaction will be performed at two-sided $\alpha = 0.1$.

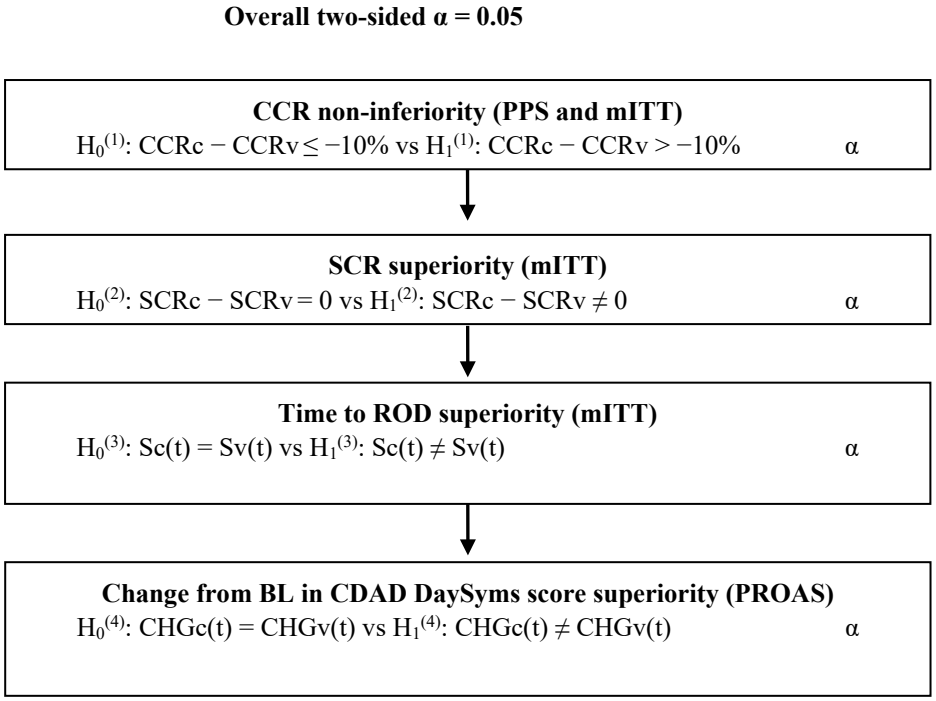
Any additional sensitivity analyses of the CCR will be specified in the SAP.

11.3.4 Analysis of the secondary variables

11.3.4.1 Sequential test strategy for secondary variables

The analyses of the secondary analyses will be performed by implementing a hierarchical strategy to control the experimentwise α level [Dmitrienko 2010] as displayed in Figure 3 with overall two-sided $\alpha = 0.05$. The secondary efficacy endpoints will be tested in a sequential conditional manner, starting with SCR.

Figure 3 Hierarchical testing strategy



CCR = Clinical Cure Rate; SCR = Sustained Cure Rate; ROD = Resolution of Diarrhea; S(t) = ROD survival function; CHG(t) = Change from BL in CDAD DaySyms Score; PPS = Per-protocol analysis set; mITT = modified intent-to-treat; PROAS = PRO analysis set.

11.3.4.2 Hypotheses and statistical inference

If NI for CCR is demonstrated in both PPS and mITT analysis set, the SCR will be hierarchically tested on the mITT analysis set at the two-sided alpha 0.05 (equivalent to one-sided test with alpha of 0.025).

The null hypothesis is: $H_0^{(2)}$: SCRc – SCRv = 0%,
and the alternative: $H_1^{(2)}$: SCRc – SCRv ≠ 0%

If the lower limit of the two-sided 95% CI is greater than zero, the statistical superiority of cadazolid versus vancomycin in SCR will be claimed.

If superiority of SCR is demonstrated in the mITT analysis set, the superiority of the secondary endpoints Time to ROD and change from baseline in CDAD DaySyms PRO total daily score up to Day 12 will be hierarchically tested respectively on the mITT analysis set and the PROAS, at the same two-sided alpha 0.05.

For testing time to ROD the null hypothesis is: H_0: Sc(t) = Sv(t) and the alternative is H_1: Sc(t) ≠ Sv(t), where S(t) denotes the survival function.

For testing change from baseline in CDAD DaySyms PRO score up to Day 12 the null hypothesis is: H_0: CHGc(t) = CHGv(t) and the alternative is H_1: CHGc(t) ≠ CHGv(t).

11.3.4.3 Analysis for secondary variables

The analyses for the secondary variables will be conducted for the mITT analysis set.

The CI for the difference in SCR [as defined in Section 11.1.2.1] will be estimated using the Wilson's score method for difference in proportions. The SCR will be summarized by treatment group per subgroup with 95% CIs for the difference of cure rates estimated using same methods and subgroups as for the main analysis.

Additional exploratory subgroup analysis will be conducted using logistic regression as specified for the primary variable.

In the hypervirulent subgroup SCR will be assessed in a meta-analysis involving studies AC-061A301 and AC-061A302 [see Section 11.4].

Analysis of Time to ROD will be conducted using a two-sided stratified log-rank test, including the strata first occurrence and first recurrence and geographical region. Kaplan-Meier estimates for the survival functions will be provided. Sensitivity analysis will be conducted using a stratified Cox proportional hazards model adjusting for covariates, Covariates as specified above will be used with first occurrence and first recurrence and geographical region following strata assignation from the IVRS.

Analysis on absolute (and percent) change from baseline in CDAD DaySyms PRO total daily score up to Day 12 will be conducted using the PROAS (excluding subjects who participate in the PRO sub-study) based on a general mixed ANCOVA model for repeated measurements modeling the absolute change from baseline in CDAD DaySyms PRO total daily scores using all available data. The model will include fixed effects terms for treatment and time and the interaction between treatment and time, the baseline CDAD DaySyms PRO total score as a covariate, and random effects of between subjects (within treatment) and within subjects (residual). Comparison between cadazolid and vancomycin will be made up to Day 12.

As the psychometric validation of the CDAD DaySyms PRO is ongoing further refinements of the analysis will be required and other analyses may be specified after completion of the validation process. Appropriate methods will be further detailed in the statistical analysis plan.

Frequency of subjects that needed antimicrobial treatment active against CDAD or FMT between first dose of study drug and EOT + 2 days, will be summarized by treatment arm. Similar summaries will be prepared for the frequency of subjects that required these kind of medications between EOT + 3 days and Visit 5 or participated in the cadazolid re-treatment extension.

Imputation methods for the substitution of incomplete, missing, or potentially biased data are defined in Section 11.3.1.

11.3.4.4 Supportive/sensitivity analyses of Sustained Cure

A sensitivity analysis of the SCR variable as defined in Section 11.1.2.1 will be performed on the PPS.

In addition, an exploratory analysis of Sustained Cure using a modified definition of recurrence will be performed, where modified recurrence is defined independently of a *C. difficile* stool GDH and toxin test for subjects with Clinical Cure as:

- NED (> 3 UBMs on any day between 3 days after EOT and Visit 5)

  AND

- Antimicrobial treatment active against CDAD (including participation in the re-treatment extension with cadazolid) or FMT started between 3 days after EOT and Visit 5).

Subjects with Clinical Cure but without modified recurrence are considered a Modified Sustained Cure. All other rules, including the handling of dropouts as specified for the variable SCR [Section 11.1.2.1 and 11.3.1] will apply to this modified SCR. The

statistical analysis for this modified SCR will be performed on the mITT analyses set using the same methods as specified for SCR.

Any additional sensitivity analyses of the SCR will be specified in the SAP.

11.3.4.5 Analysis of the other efficacy variables

Analysis for the exploratory variables will be conducted for the mITT analysis set unless otherwise specified.

ICRR (%) will be analyzed in the PPS and mITT analysis set, using the same null hypothesis and statistical methods as for the statistical analysis of primary efficacy endpoint.

ISRR (%) will be analyzed, in the mITT analysis set, using the same null hypothesis and statistical methods as for the statistical analysis of SCR.

RR (%) will be analyzed in an exploratory manner, to support the reduction in recurrence indirectly demonstrated after showing NI on CCR and superiority on SCR.

All the different cure rates (ICRR, ISRR, RR, NBMR, and ECCR) will be summarized per treatment arm. 95% CIs for the difference of cure rates will be estimated using the Wilson's Score method [Newcombe 1998].

Time to return to usual stools will be estimated by Kaplan-Meier. Time to recurrence will be estimated by Kaplan-Meier based on subjects clinically cured only.

Susceptibility test results for *C. difficile* isolates at baseline and post-baseline visits will be summarized with descriptive statistics including MIC range, MIC50, MIC90, and the frequency of isolates at each MIC test concentration for each test agent.

VRE counts at baseline and Visit 3, as well as changes from baseline to Visit 3 VRE counts, will be summarized with descriptive statistics.

Susceptibility test results for vancomycin-resistant *E. faecium* and vancomycin-resistant *E. faecalis* isolates at Visit 1 and Visit 3 will be summarized including MIC range, MIC50, MIC90, and the frequency of isolates at each MIC test concentration for each test agent.

Efficacy endpoints for the extension treatment period will be summarized on the ES, using descriptive statistics and frequencies, overall, and by the randomized treatment group.

All other exploratory efficacy endpoints will be summarized using descriptive statistics using the mITT analysis set.

11.3.5 Analysis of the safety variables

The Safety analysis set will be used to perform all safety analyses. All safety data will be included in listings, with flags for quantitative abnormalities.

11.3.5.1 Adverse events

All AEs will be coded using MedDRA Version 16 or higher. TEAEs and treatment emergent SAEs up to 7 days after last dose of study drug will be tabulated by study treatment, System Organ Class (SOC) and preferred terms within each SOC: number and percentage of subjects who experienced (S)AEs coded with the same preferred term will be displayed. (S)AEs will also be tabulated by maximum intensity and relationship to investigational drug or comparator.

AEs and SAEs from first dose up to Visit 5 will be summarized by treatment group, SOC and preferred terms within SOC. AEs leading to premature discontinuation of study treatment will be summarized in similar manner.

Re-treatment extension-emergent AEs and SAEs (occurring during the re-treatment extension with cadazolid) will be summarized with similar tables, and based on the ES.

Listings will be provided for all reported AEs, including SAEs. In addition, separate listings will be provided for SAEs, for AEs leading to premature discontinuation of study drug, and for AEs leading to death.

11.3.5.2 Deaths

AEs with outcome death will be summarized in a similar manner as described for SAEs, for TEAEs up to 7 days and separately for AEs up to Visit 5.

All deaths will be summarized separated into deaths from first dose to EOT + 7 days, and deaths from first dose of study drug until Visit 5. Listings of all deaths will be provided.

11.3.5.3 Vital signs

Descriptive summary statistics by visit and study treatment will be provided for observed values and absolute changes from baseline in SBP, DBP, HR, body temperature and body weight. The number of subjects with at least one marked abnormality up to EOT + 7 days (treatment-emergent) and up to Visit 5 will be summarized. Treatment-emergent abnormality is defined as an abnormality post first dose study drug not present at baseline. Marked abnormalities are defined in Appendix 4.

11.3.5.4 ECGs

Descriptive summary statistics by visit and study treatment will be provided for observed values and absolute changes from baseline in numeric 12-lead ECG values (HR, PR, QRS, QT, QT corrected for HR on the basis of Bazett's formula [QTcB], QT corrected

for HR on the basis of Fridericia's formula [QTcF]). The number of subjects with at least one marked QT or QTc abnormality up to EOT + 7 days (treatment-emergent) and up to Visit 5 will be summarized. Treatment-emergent abnormality is defined as an abnormality post first dose study drug not present at baseline. Marked abnormalities are defined in Appendix 4.

The number of subjects with abnormal ECG findings not present prior to start of treatment will be summarized.

11.3.5.5 Hematology, coagulation and chemistry

Descriptive summary statistics by visit and study treatment will be provided for observed values and absolute changes from baseline for laboratory tests (hematology, chemistry). Data will be displayed in Standardized International (SI) units as provided by the Central Laboratory. If any local laboratories are used with different normal ranges, then the values and their normal ranges will be normalized using the Central Laboratory values as a reference, and this will be detailed in the SAP.

The number of subjects with at least one treatment-emergent abnormality and the number of subjects with at least one marked abnormality up to EOT + 7 days (treatment-emergent) and up to Visit 5 will be summarized. Treatment-emergent abnormality is defined as an abnormality post-first dose of study drug not present at baseline.

Treatment-emergent laboratory test abnormalities will be assessed according to the normal ranges. Marked abnormalities are defined in Appendix 4.

11.3.6 Analysis of other variables

Pharmacoeconomic hospitalization variables will be summarized with descriptive statistics in the mITT analysis set. Variables related to the WPAI:CDAD questionnaire will be summarized using descriptive statistics on the PROAS.

Cadazolid 2 h post-dose plasma concentrations at Visit 3 will be summarized using descriptive statistics (arithmetic mean, 95% CI of the arithmetic mean, minimum, maximum, median, SD, standard error (SE), and if the data allows geometric mean and 95% CI of the geometric mean).

11.3.7 Exposure to study drug

The duration of exposure and mean daily dose will be tabulated using the usual location and scale statistics by treatment group. Compliance will be tabulated using the usual location and scale statistics by treatment group and separately for sachets and capsules. The cumulative distribution of exposure time by different class intervals (e.g., at least

1 day, etc.) will be tabulated to show the number and percentage of subjects in each class interval.

11.3.8 Demographic and baseline characteristics

Summaries for demographic and baseline characteristics will be performed on the FAS, PPS and the mITT analysis set. The variables country, sex, age categories, BMI, ethnicity, race, disease severity, prior antimicrobial treatment active against CDAD, first occurrence / first recurrence, and initial strain of CDAD (hypervirulent or not), in-patient at randomization, baseline frequency of UBMs, concomitant treatment with opiates at baseline, concomitant antibiotics for infections other than CDAD, concomitant treatment with PPIs will be summarized per treatment group, overall, and also only for the subjects presenting Clinical Cure.

Previous and concomitant medications will be coded according to the WHO drug code and the ATC class code, and summarized by tabulating the number and percentages of subjects having received each treatment.

Summaries of concomitant medications will also be presented separately for concomitant medications according to antimicrobial treatment active against CDAD, concomitant antibiotics for infections other than CDAD and concomitant treatment with PPIs.

Medical history will be coded using MedDRA and summarized in a similar manner to AEs.

Protocol deviations will be listed and summarized by treatment group and type of deviation.

11.4 Meta-analysis

A meta-analysis of trials AC-061A301 and AC-061A302 will be performed to assess the superiority of cadazolid for the SCR in the hypervirulent subgroup. The analysis will be performed if NI in Clinical Cure and superiority in Sustained Cure was demonstrated in the whole mITT analysis set within both individual trials.

The null hypothesis is: $H_0^{(3)}$: SCRc – SCRv = 0%,
and the alternative: $H_1^{(3)}$: SCRc – SCRv ≠ 0%

Superiority of Sustained Cure in the hypervirulent subgroup is assessed using a fixed-effect meta-analysis combining the risk difference estimates from both individual trials using the HVAS. The analysis will be stratified by trial. A CI is derived and superiority test performed based on normal approximation. A separate SAP will be prepared for this analysis.

11.5 Interim analyses

No interim analysis is planned.

11.6 Sample size

Approximately 630 subjects will be randomized 1:1 to the 2 treatment groups (315 cadazolid and 315 vancomycin).

11.6.1 Sample size justification

Assuming a CCR of 85% for cadazolid and vancomycin treatment groups, a power of 90%, a type I error of 2.5% (one-sided) and an NI margin of 10%, a total sample size of 536 (268 in each treatment group) evaluable subjects is required. Taking into account approximately 15% subjects not qualifying for the PPS and approximately 5% not qualifying for the mITT analysis set, a total of approximately 630 subjects will be randomized into the trial.

Assuming 598 subjects in the mITT analysis set (approximately 95% of the randomized subjects) and the same assumptions on CCR as for the PPS, the power for showing NI in clinical cure on the mITT analysis set is 93%. The probability to be successful in both analysis sets is approximately 89% under the assumption of 90% of the subjects in the mITT analysis set qualifying for the PPS. The power for showing superiority of cadazolid versus vancomycin in SCR on the mITT analysis set will be 98%, with a difference in SCR of at least 15%, and 88% with a difference of 12%, assuming the SCRv is 60%, and a two-sided type I error of 0.05. The above results are derived independently of the primary variable and are conditional on reaching this step in the hierarchical testing strategy.

It is estimated that 25% of the subjects would present a hypervirulent strain. Thus, the pooled analysis would be based on approximately 300 evaluable subjects (150 from each trial) in the mITT analysis set.

The power for showing a superiority of at least 15% in SCR, cadazolid versus vancomycin, in the hypervirulent subgroup, with two-sided type I error of 0.05, is approximately 80%, assuming a SCR of 60% in the vancomycin group. The above results are derived independently of the primary and secondary variable and are conditional on reaching this step in the hierarchical testing strategy.

11.6.2 Sample size sensitivity

With a fixed sample size of 536 (268 per treatment group) the power for showing NI depends on the assumed CCR in the vancomycin group. Table 4 displays power calculations for different assumptions for the CCRs for vancomycin with equally large CCR in the cadazolid group, one-sided type I error of 0.025, NI margin of 10% and sample size of 536 as estimated for the PPS. With CCRs ranging from 80–90% the

resulting power ranges from 82–97%. A table like this one for the mITT analysis set would show, using the same assumptions, a range of estimated power slightly higher than for the PPS due to the higher number of subjects. Similarly, the probability to be successful in both the mITT analysis set and the PPS will range from approximately 81–97% depending on the actual CCR rate in both populations.

Table 4 Sample size sensitivity for main analysis (PPS)

Scenario	CCRv (%)	CCRc (%)	Power (%)
1	90.0	90.0	97
2	87.5	87.5	94
3	82.5	82.5	86
4	80.0	80.0	82

CCRc = clinical cure rate for cadazolid, CCRv = clinical cure rate for vancomycin, PPS = Per-protocol analysis set.

Similarly, with fixed sample size, the power for showing superiority of cadazolid versus vancomycin in SCR depends on the SCR in the vancomycin group (SCRv) and the difference between the SCR in the treatment groups. Table 5 displays the power for showing superiority in SCR of cadazolid versus vancomycin with a fixed sample size of 598 in the mITT analysis set, and a two-sided type I error of 5%.

Table 5 Sample size sensitivity for secondary variable Sustained Cure Rate

Scenario	SCRv (%)	SCRc–SCRv (%)	Power (%)
1	65.0	15.0	99
2	55.0	15.0	97
3	65.0	12.0	90
4	55.0	12.0	86

SCRc = sustained cure rate cadazolid, SCRv = sustained cure rate vancomycin.

11.6.3 Sample size re-estimation

No sample size re-estimation is planned.

12 DATA HANDLING

12.1 Data collection

The investigator/delegate is responsible for ensuring the accuracy, completeness, legibility and timeliness of the data reported. All source documents should be completed in a neat, legible manner to ensure accurate interpretation of the data. Data reported in the electronic CRF derived from source documents must be consistent with the source documents.

CRF data will be captured via electronic data capture (using the Rave system provided by Medidata Solutions, Inc., a web-based tool). The investigator and site staff will be trained to enter and edit the data via a secure network, with secure access features (username, password and identification – an electronic password system). A complete electronic audit trail will be maintained. The investigator/delegate will approve the data (i.e., confirm the accuracy of the data recorded) using an electronic signature (ref. to 21 CFR Part 11).

Entries recorded on paper (e.g., stool diary) by a subject are considered source data. The site staff will review and ensure completeness and readability of a subject's entries recorded on paper. The site will also ensure that if any information allowing subject's identification is written on the paper(s) collected, that it will not be visible when copies of the source data are provided to the sponsor for entry into the clinical trial database.

Subject screening and enrollment data will be completed for all subjects (i.e., eligible and non-eligible) through the IVRS system and CRF.

For each subject screened, regardless of randomization or study drug initiation, a CRF must be completed and signed by the investigator/delegate. This also applies to those subjects who fail to complete the study. If a subject withdraws from the study or discontinues treatment prematurely, then the reason must be noted on the CRF.

12.2 Maintenance of data confidentiality

The investigator/delegate must ensure that data confidentiality is maintained. On CRFs or other documents submitted to Actelion, subjects must be identified only by number, and never by name. The investigator/delegate must keep a subject identification code list, at the site, showing the randomization number, the subject's name, date of birth and address or any other locally accepted identifiers. Documents identifying the subjects (e.g., signed ICFs) must not be sent to Actelion, and must be kept in strict confidence by the investigator/delegate.

12.3 Database management and quality control

Electronic CRFs will be used for all subjects. The investigator will have access to the site CRF data throughout the study. The CRF must be kept current to reflect subject status at any time point during the course of the study (i.e., the data should be entered in the system within 10 days of the subject's visit/assessment completion).

While entering the data, the investigator/delegate will be prompted by logical checks/error messages built into the web-based data entry screening performed on the data. Other protocol-specific validation programs will run routinely to perform more extensive data checks for accuracy and completeness. Additional data review will be processed in parallel by the sponsor to look for unexpected patterns in data and study monitoring. In the event that problematic data are detected, a query specifying the problem and requesting clarification will be issued and visible to the investigator/delegate via the CRF, so that the investigator/delegate can respond and clarify directly onto the CRF.

The investigator/delegate must, on request, supply Actelion with any required background data from the study documentation or clinical records. This is particularly important when errors in data transcription are suspected. In the case of health authority queries, it is also necessary to have access to the complete study records, provided that subject confidentiality is protected.

This process will continue until database closure.

Laboratory samples will be processed through a central laboratory and specialist microbiology labs and the results will be sent electronically to Actelion.

Data from local ECGs and local laboratories will be entered into the database by site staff (except in the case of ECG read by [redacted], where data will be transferred electronically to Actelion), and **subject** PRO data will be entered into the database by data management using double data entry. Data from the PK sub-study will be transferred electronically to Actelion following the database closure of both the AC-061A301 and AC-061A302 studies.

After the database has been declared complete and accurate, the database will be closed. Any changes to the database after that time may only be made as described in the appropriate Actelion SOP. At the end of the trial, the investigator will receive the CRF of the subjects of her/his site (including all data changes made) on a CD-ROM or as a paper copy.

13 PROCEDURES AND GOOD CLINICAL PRACTICE

13.1 Ethics and Good Clinical Practice

The sponsor and investigators will ensure that the study is conducted in full compliance with ICH-GCP Guidelines, the principles of the 'Declaration of Helsinki' and with the laws and regulations of the country in which the research is conducted.

13.2 Independent Ethics Committee / Institutional Review Board

The investigator will submit this protocol and any related document provided to the subject (such as Subject Information Leaflet used to obtain informed consent) to an IRB or IEC. Approval from the committee must be obtained before starting the study, and must be documented in a dated letter to the investigator, clearly identifying the study, the documents reviewed, and the date of approval.

Modifications made to the protocol after receipt of the approval must also be submitted as amendments by the investigator to the IRB/IEC in accordance with local procedures and regulations [see Section 13.6].

A list of members participating in the meeting must be provided, including the names, qualifications and functions of these members. If that is not possible, the attempts made to obtain this information along with an explanation as to why it cannot be obtained or disclosed must be documented in the study documentation. If a study staff member was present during a meeting, it must be clear that this person did not vote.

13.3 Informed consent

It is the responsibility of the investigator/delegate to obtain informed consent according to ICH-GCP guidelines and local regulations from each individual participating in this study and/or legally authorized representative. The investigator/delegate must explain to subjects that they are completely free to refuse to enter the study, or to withdraw from it at any time for any reason.

The ICF will be provided in the country local language(s).

Site staff authorized to participate in the consent process and/or to obtain consent from the subject and/or legal representative will be listed on Actelion Delegation of Authority form. A study physician must always be involved in and have oversight of the informed consent process.

In sites participating in the cadazolid plasma concentration sub-study, the subject will have to consent to the study and will have the opportunity to consent to participating in the sub-study. A separate independent consent will also be needed for participation in the re-treatment extension study with cadazolid at the time of a NED.

At sites where the sponsor-approved EIA is not standard of care for the diagnosis of CDAD, a two-part ICF may be used: Section A covering the use of the subjects' stool sample to perform the sponsor-approved EIA, and Section B covering enrollment into the study. The subject and/or legally authorized representative must sign, personally date and time (if appropriate) the ICF before any study-related procedures (i.e., any procedures required by the protocol) begin. If a study-specific procedure or assessment has been performed as part of routine assessments, meets the protocol requirements, and the results are available prior to the signing of the ICF, such procedure or assessment may be used to assess eligibility and does not have to be repeated. In such cases, it must be clear from the source document when and for which reason the assessment was done prior to the signing of the informed consent. The ICF must also be signed, personally dated and timed (if appropriate) by the authorized site staff listed on Actelion Delegation of Authority form.

A copy of the signed and dated ICF is given to the subject and/or legally authorized representative; the original is filed in the site documentation.

The informed consent process must be fully documented in the subject's medical records, including study reference, subject number, date/time (if applicable) when the subject was first introduced to the Actelion clinical study, date/time (if applicable) of consent, who participated in the consent discussion, who consented the subject and any additional person present during the consent process (e.g., subject family member), copy of the signed ICF given to the subject/legally authorized representative.

The subject is also given a clinical study participant card to provide information to medical personnel and/or health care professionals that the subject is participating in a clinical trial – who to contact to receive more information about the trial, and how this person can be reached.

In the event that the site would like to recruit a subject who cannot read or does not speak or understand the ICF language, additional measures must be implemented in order to ensure subject rights are respected and the consent obtained is legally valid. The sponsor, the regulatory authorities (if applicable) and the IRB/IEC must be informed prior to the recruitment. The consent process (e.g., involvement of an impartial witness) must be fully described, submitted to, and approved by the IRB/IEC, according to procedures.

The informed consent process must proceed as follows:

- A physician investigator participates in and has oversight of the informed consent process (i.e., introduction of the study and consent process)
- A physician answers medical questions, questions regarding alternatives to therapy or any other questions from the subject.

- A physician or delegated advance practice role discusses trial procedures and risks.
- A physician or delegated advance practice role answers non-medical questions.
- A physician or delegated advance practice role performs source documentation of the consent.
- Before the subject signs the ICF, a physician verifies with the subject that all questions have been answered.

13.4 Compensation to subjects and investigators

The sponsor provides insurance in order to indemnify (with both legal and financial coverage) the investigator/site against claims arising from the study, except for claims that arise from malpractice and/or negligence.

The compensation of the subject in the event of study-related injuries will comply with applicable regulations.

13.5 Protocol adherence/compliance

The investigator must conduct the study in compliance with the approved version of the protocol and must not implement any violation/deviation/change from the protocol, except when the change involves only logistical or administrative aspects (e.g., change in telephone number), or in case it would be necessary to eliminate an immediate hazard to the subject.

If a protocol violation occurs, the investigator/delegate will inform Actelion or its representative, in a timely manner. The investigator/delegate must document in the CRF and in the subject's file and explain any violation/deviation/change from the approved protocol. Protocol violation/deviation/change must also be submitted to IRB/IEC and to the regulatory authority(ies), according to their requirements.

13.6 Protocol amendment

Any change to the protocol can only be made through a written protocol amendment approved by Actelion. A protocol amendment must be submitted in a timely manner (no more than 30 calendar days after receipt) to IRB/IEC, and regulatory authority(ies), according to their requirements.

Approval must be obtained before any change can be implemented, except when the change involves only logistical or administrative aspects (e.g., change in telephone number), or in case it would be necessary to eliminate an immediate hazard to the subject; in this case. IRB/IEC and regulatory authority(ies) must be notified as soon as possible, according to their requirements, but not later than 15 calendar days.

13.7 Essential documents and retention of documents

The investigator/delegate must maintain adequate records necessary for the reconstruction and evaluation of the study. A number of attributes are considered of universal importance to source data and the records that hold those data. These include that the data and records are accurate, legible, contemporaneous, original (or certified copy), attributable, complete, consistent, enduring and available when needed.

These records are to be classified into two different categories of documents: investigator's file, and subject clinical source documents.

These records must be kept by the investigator for as long as is necessary to comply with Actelion's requirements (e.g., as specified in the clinical study agreement), and national and/or international regulations, whichever would be the longest period. If the investigator cannot guarantee this archiving requirement at the investigational site for any or all of the documents, special arrangements, respecting the data confidentiality, must be made between the investigator and Actelion to store these documents outside the site, so that they can be retrieved in case of a regulatory inspection. No study document should be destroyed without prior written approval from Actelion. Should the investigator wish to assign the study records to another party, or move them to another location, Actelion must be notified in advance.

Documentation of study assessments must be available in the subject medical records (e.g., vital signs, physical examination), including the screening procedures/assessment prior to enrollment.

If the site is using an electronic/computerized system to store subject medical records, it can be used for the purpose of the clinical study if it is validated (as per 21 CFR Part 11 or equivalent standard) and if the monitor has been provided personal access to verify consistency between electronic source data and the CRF during monitoring visits.

If the site is using an electronic/computerized system to store subject medical records but it could not be confirmed that the system is validated or if the monitor could not be provided access to the system, the site is requested to print the complete set of source data needed for verification by the monitor. The print-outs must be signed and dated by the investigator/delegate to confirm that these certified copies are exact copies with the same information as the original subject's data. The print-outs will be considered as the official clinical study records. In order to verify that the process the site uses to prepare certified copies is reliable, the monitor must be able to observe this process and confirm that the comparison of the source documents and the certified copy did not reveal inconsistencies. The monitor does not need to verify this process for all data of all subjects, but at least for some of them (e.g., first subject; regular check during the study of critical data like inclusion/exclusion criteria, endpoints for some subjects) as per

Actelion's instructions. If it is not possible for the monitor to observe this process, then it is not possible to rely on the site's certified copies, and therefore the site cannot be used for the clinical study. The printouts should be filed either with the subject medical records or with the subject's CRF.

13.8 Equipment considerations and quality requirements

Documentation should be made available to the monitor and any potential auditor to verify the qualification of equipment or physical devices used to perform study tasks. This includes thermometers in areas where study drug is stored, freezers that store lab samples, and/or equipment such as ECG machines, scales, etc. used for study assessments. This documentation could include validation or calibration certificates and freezer logs, and should be filed in the ISF. It is the responsibility of the investigational site to maintain these documents. Only certified/calibrated equipment or validated systems should be used.

13.9 Monitoring

The monitor will contact and visit the investigational site regularly, and on request must be permitted to have access to trial facilities and all source documents needed to verify adherence to the protocol and the completeness, consistency and accuracy of the data being entered on the CRFs and other protocol-related documents. Actelion monitoring standards require full verification that informed consent has been provided, and verification of adherence to the inclusion/exclusion criteria, documentation of SAEs, and the recording of the main efficacy, safety and tolerability endpoints; this includes verification that source documentation relevant to the study (e.g., safety events) is reported in the CRFs. Additional checks of the consistency of the source data with the CRFs will be performed according to the study-specific monitoring plan.

The required site personnel must be available during monitoring visits and allow adequate time to meet with the monitor to discuss study related issues. An initiation visit will be performed after the required essential documents are received at Actelion and before the first subject is included in the study. Monitoring visits will be conducted regularly; frequency will be based on subject recruitment rate and critical data collection times. A close-out visit will be performed for any initiated site.

The investigator agrees to cooperate with the monitor(s) to ensure that any issues detected in the course of these monitoring visits are resolved. If the subject is hospitalized or dies in a hospital other than the study site, the investigator is responsible for contacting that hospital in order to document the SAE.

13.10 Audit

Actelion's Global Quality Management representative may audit the investigator site (during the study or after its completion). The purpose of this visit will be to determine the investigator's adherence to ICH-GCP, the protocol, applicable regulations, and the sponsor's requirements (e.g., SOPs). Prior to initiating this audit, the investigator will be contacted by Actelion to arrange a convenient time for the audit.

The investigator and staff must cooperate with the auditor(s) and allow access to all study documentation (e.g., subject records) and facilities.

13.11 Inspections

Health authorities and/or IRB/IEC may also wish to conduct an inspection of Actelion's clinical study (during the study or after its completion).

Should an inspection be requested by a health authority and/or IRB/IEC, the investigator must inform Actelion immediately (usually via the monitor) that such a request has been made.

The investigator and staff must cooperate with inspector(s) and allow access to all study documentation (e.g., subject records) and study facilities.

13.12 Reporting of study results and publication

Study results will be documented in a clinical study report that will be signed by Actelion representatives and the Coordinating Investigator.

The coordinating investigator and the Steering Committee, will have the opportunity to review the analysis of the data and to discuss the interpretation of the study results with the sponsor prior to publication.

Actelion will post results from Phase 2–4 clinical studies on Actelion's Clinical Trial Register, and on external/national registries, as required by law.

In accordance with the Good Publication Practices and ethical practice, the results of the study will be submitted for publication in a peer-reviewed journal. Investigators shall not publish data from an individual study center until after the results of the multicenter center are published. Study results can be submitted for presentation at a congress before publication in a peer-reviewed journal.

Authorship will be determined in accordance with the International Committee of Journal Editors criteria, and be based on:

- Substantial contributions to the conception and design, or acquisition of data, or analysis and interpretation of data

- Drafting of the publication or critical review for important intellectual content
- Providing final approval of the version to be published.

The list of authors of any publication of study results may include representatives of Actelion, and will be determined by mutual agreement.

Any study-related publication written independently by investigators must be submitted to Actelion for review at least 30 days prior to submission for publication or presentation. Upon review, Actelion may provide comments, and may also request alterations and/or deletions for the sole purpose of protecting its confidential information and/or patent rights. Neither the institution nor the investigator should permit publication during such a review period.

Actelion's Policy on Scientific Publications can be found at:

14 REFERENCES

[Agresti 2000] Agresti A, Caffo B. Simple and effective confidence intervals for proportions and difference of propor- tions result from adding two successes and two failures. The American Statistician 2000;54:280-288.

[Bauer 2009] Bauer MP, Kuijper EJ, van Dissel JT. European Society of Clinical Microbiology and Infectious Diseases (ESCMID): treatment guidance document for *Clostridium difficile* infection (CDI). Clin Microbiol Infect. 2009 Dec;15(12):1067–79.

[Bauer 2011] Bauer MP, Notermans DW, van Benthem B, Brazier JS, Wilcox MH, Rupnik M, et al. *Clostridium difficile* infection in Europe: a hospital-based survey. Lancet 2011;377:63–73.

[Bhangu 2012] Bhangu A, Nepogodiev D, Gupta A, Torrance A, Singh P. Systematic review and meta-analysis of outcomes following emergency surgery for *Clostridium difficile* colitis. British Journal of Surgery 2012;99:1501–13.

[Bouza 2008] Bouza E, Dryden M, Mohammed R, Peppe J, Chasan-Taber S, Donovan J, et al. Results of a phase III trial comparing tolevamer, vancomycin and metronidazole in patients with Clostridium difficile-associated diarrhea. Poster Abstract Number: O464. 18th European Congress of Clinical Microbiology and Infectious Diseases Barcelona, Spain, 19–22 April 2008.

[Cadazolid IB] Investigator's Brochure for cadazolid, version 6. Actelion Pharmaceuticals Ltd, June 2013.

[Cohen 2010] Cohen, SH; Gerding DN; Johnson, S et al. Clinical Practice Guidelines for *Clostridium difficile* Infection in Adults: 2010 Update by the Society for Healthcare Epidemiology of America (SHEA) and the Infectious Diseases Society of America (IDSA): SHEA-IDSA Guideline: Infection Control and Hospital Epidemiology 2010 May;31(5).

[Cornely 2012] Cornely OA, Crook DW, Esposito R, Poirier A, Somero MS, Weiss K, et al. Fidaxomicin versus vancomycin for infection with *Clostridium difficile* in Europe, Canada, and the USA: a double-blind, non-inferiority, randomised controlled trial. Lancet Infect Dis. 2012;12: 281–89.

[Cuzick 2005] Cuzick, J. Forest plots and the interpretation of subgroups. The Lancet, 2005;365(9467),1308.

[Dallal 2002] Dallal RM, Harbrecht BG, Boujoukas AJ, Sirio CA, Farkas LM, Lee KL, et al. Fulminant *Clostridium difficile*: An Underappreciated and Increasing Cause of Death and Complications. Annals Of Surgery 2002; 235(3):363–372.

[De Almeida 2013] De Almeida MN, Heffernan H, Dervan A, Bakker S, Freeman JT, Bhally H, et al. Severe *Clostridium difficile* infection in New Zealand associated with an emerging strain, PCR-ribotype 244. The New Zealand Medical Journal 2013;126:9-14.

[Dmitrienko 2010] Dmitrienko A, Tamhane AC, Bretz, F, editors. Multiple testing problems in pharmaceutical statistics. CRC Press; 2010.

[Dubberke 2010] Dubberke ER, Butler AM, Yokoe DS, Mayer J, Hota B, Mangino JE , et al. Multicenter Study of Surveillance for Hospital-Onset *Clostridium difficile* Infection by the Use of ICD-9-CM Diagnosis Codes. Infect Control Hosp Epidemiol. 2010; 31:262–8.

[EMA 2011] Addendum to the note for guidance on evaluation of medicinal products indicated for treatment of bacterial infections (CPMP/EWP/558/95 REV 2) [EMA/CHMP/776609/2011] [accessed on 2013 Aug 29]. Available from: http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2012/07/WC500129443.pdf

[FDA 2009] FDA. Patient reported outcome measures: Use in Medical Product Development to Support Labeling Claims. Silver Spring, MD: US Dept of Health and Human Services Food and Drug Administration; 2009 [accessed on 2013 Aug 29]. Available from: http://www.fda.gov/downloads/Drugs/Guidances/UCM193282.pdf.

[FDA 2010] FDA. Guidance for Industry: Non-Inferiority Clinical Trials. Draft Guidance. Silver Spring, MD: US Dept of Health and Human Services Food and Drug Administration; 2010 [accessed on 2014 Aug 29]. Available from: http://www.fda.gov/downloads/Drugs/Guidances/UCM202140.pdf.

[Freeman 2010] Freeman J, Bauer MP, Baines SD, Corver J, Fawley WN, Goorhuis B, et al. The changing epidemiology of Clostridium difficile infections. Clin. Microbiol. Rev. 2010;23:529–49.

[Garey 2008] Garey KW, Sethi S, Yadav Y, DuPont HL. Meta-analysis to assess risk factors for recurrent *Clostridium difficile* infection. J Hosp Infect. 2008 Dec;70(4):298–304.

[Hu 2009] Hu MY, Katchar K, Kyne L, Maroo S, Tummala S, Dreisbach V, et al. Prospective Derivation and Validation of a Clinical Prediction Rule for Recurrent *Clostridium difficile* Infection. Gastroenterology 2009;136:1206–14.

[Johnson 2012] Johnson S, et al. Efficacy and Safety of Oral Vancomycin Versus Oral Metronidazole for Treatment of *Clostridium difficile* Associated Diarrhea (CDAD): Pooled Results from Two Randomized Clinical Trials; IDSA 2012 San Diego, CA.

[Kelly 2008] Kelly CP, LaMont T. *Clostridium difficile* - more difficult than ever. N Engl J Med. 2008;359:1932–40.

[Lim 2014] Lim SK, Stuart RL, Mackin KE, Carter GP, Kotsanas D, Francis MJ, et al. Emergence of a ribotype 244 strain of Clostridium difficile associated with severe disease and related to the epidemic ribotype 027 strain. Clin Infect Dis. 2014 Jun15;58(12):1723-30. doi: 10.1093/cid/ciu203. Epub 2014 Apr 4

[Loo 2005] Vivian G. Loo, et al. A Predominantly Clonal Multi-Institutional Outbreak of *Clostridium difficile*–Associated Diarrhea with High Morbidity and Mortality. N Engl J Med. 2005;353:2442–9.

[Louie 2006] Louie T, Peppe J, et al. Tolevamer, a novel nonantibiotic polymer, compared with vancomycin in the treatment of mild to moderately severe Clostridium difficile-associated diarrhea. Clin Infect Dis 2006;43: 411-420.

[Louie 2007] Louie TJ, Gerson M, Grimard D, Johnson S, Poirier A, Weiss K, et al. Results of a phase III study comparing tolevamer, vancomycin and metronidazole in Clostridium difficile-associated diarrhea (CDAD). In: Program and Abstracts of the 47th Interscience Conference on Antimicrobial Agents and Chemotherapy (ICAAC); 17–20 September 2007; Chicago, IL.Washington, DC:ASMPress; 2007. Abstract K-4259.

[Louie 2011] Louie TJ, Miller MA, Mullane KM, Weiss K, Lentnek A, Golan Y, et al. Fidaxomicin versus Vancomycin for *Clostridium difficile* Infection. N Engl J Med. 2011;364:422–31.

[Louie 2013a] Louie TJ et al. Multicentre, double-blind, randomized phase 2 study evaluating the novel antibiotic cadazolid in subjects with *Clostridium difficile*-associated diarrhea; 23rd ECCMID Conference; 2013 April 27–30; Berlin, Germany.

[Louie 2013b] Louie TJ, Miller MA, Crook DW, Lentnek A, Bernard L, High KP, et al. Effect of Age on Treatment Outcomes in *Clostridium difficile* Infection. JAGS 2013;61:222–30.

[Miao He 2012] Miao He et al. Emergence and global spread of epidemic healthcare-associated *Clostridium difficile*. Nature Genetics 2013; 45:109-13.

[Miller 2011] Miller BA, Chen LF, Sexton DJ, Anderson DJ. Comparison of the burdens of hospital-onset, healthcare facility-associated *Clostridium difficile* infection and of healthcare-associated infection due to methicillin-resistant Staphylococcus aureus in community hospitals. Infect Control Hosp Epidemiol. 2011 Apr;32(4):387–90.

[Musher 2005] Musher DM, Aslam S, Logan N, et al. Relatively poor outcome after treatment of *Clostridium difficile* colitis with metronidazole. Clin Infect Dis. 2005; 40:1586–90.

[Newcombe 1998] Newcombe, RG. Improved confidence intervals for the difference between binomial proportions based on paired data. Statistics in Medicine 1998;17(22): 2635–50.

[O'Brien 2007] O'Brien JA, Lahue BJ, Caro JJ, Davidson DM. The emerging infectious challenge of *Clostridium difficile*-associated disease in Massachusetts hospitals: clinical and economic consequences. Infect Control Hosp Epidemiol. 2007; Nov;28(11):1219-27.

[Pachecho 2013] Pachecho SM, Johnson S. Important clinical advances in the understanding of *Clostridium difficile* infection. Curr Opin Gastroenterol. 2013 Jan;29(1):42–8.

[Poutanen 2004] Poutanen SM, Simor AE. *Clostridium difficile*-associated diarrhea in adults. CMAJ 2004;171(1):51–8.

[Reilly 1993] Reilly MC, Zbrozek AS, Dukes E. The validity and reproducibility of a work productivity and activity impairment measure. PharmacoEconomics 1993; 4(5):353–65.

[Tannock 2010] Tannock GW, Young W, Munro K, Byrne B, Taylor C, Emery J, et al. A new macrocyclic antibiotic, fidaxomicin (OPT80), causes less alteration to the bowel microbiota of *Clostridium difficile*-infected patients than does vancomycin. Microbiology 2010;156, 3354–59.

[Vancomycin USPI] VANCOCIN® (Vancomycin hydrochloride capsules, USP). Package Insert. ViroPharma Incorporated [accessed on 2013 Aug 29]. Available from http://www.accessdata.fda.gov/drugsatfda_docs/label/2011/050606s028lbl.pdf

[Voelker 2010] Voelker R. Increased *Clostridium difficile* Virulence Demands New Treatment Approach. JAMA 2010 May;303(20)20:2017–19.

[Weiss 2009] Weiss K. Toxin-binding treatment for Clostridium difficile: a review including reports of studies with tolevamer. Int J Antimicrob Agents. 2009;33(1): 4-7.

[Wilcox 1995] Wilcox MH, Howe R. Diarrhoea caused by *Clostridium difficile*: response time for treatment with metronidazole and vancomycin. J Antimicrob Chemother. 1995 Oct;36(4):673–9.

[Zar 2007] Zar FA, Bakkanagari SR, Moorthi KMLST, Davis MB. A Comparison of Vancomycin and Metronidazole for the Treatment of *Clostridium difficile*-Associated Diarrhea, Stratified by Disease Severity. Clinical Infectious Diseases 2007;45:302–7.

Appendix 1 Equivalence between opiates

By convention, for this study, the following equivalence will be used:

Drug	Route	Equianalgesic dose (mg)	Duration (h)
Morphine	IM/IV/SC	10	3-4
Morphine	PO	20-30	3-6
Codeine	IM	130	4-6
Codeine	PO	200	4-6
Oxycodone	PO	15-20	3-6
Hydrocodone	PO	30	4-8
Oxymorphone	IM/IV/SC	1	3-6
Oxymorphone	PO	15	-
Hydromorphone	IM/IV/SC	1.5	3-4
Hydromorphone	PO	7.5	3-6
Levorphanol	IM/IV/SC	2	4-8
Levorphanol	PO	4	4-8
Meperidine	IM/IV	75	3-5
Meperidine	PO	300	3-5
Methadone	IM/IV/SC	10*	6-8
Methadone	PO	20	4-6
Fentanyl	IV/SC	0.1	0.5-1 IV; 1-2 SC
Buprenorphine	IV/IM	0.3–0.4	6
Buprenorphine	patch	5–10 microgram	7 days

*Extremely variable with chronic dosing; e.g., conversion from oral morphine to oral methadone may range from 4 to 14:1.

Appendix 2 Stool diary

DATE __ __.__ __ __. __ __
d d m m m y y

☐ CHECK IF NO BOWEL MOVEMENT TODAY

ENTRY MADE BY SITE STAFF	BOWEL MOVEMENT		STOOL RECONCILIATION *FOR SITE PERSONNEL ONLY*
☐	1	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	2	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	3	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	4	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	5	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	6	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	7	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	8	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	9	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
☐	10	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐	Time: _ _. _ _ UBM: ☐ NOT UBM: ☐
			TOTAL BOWEL MOVEMENTS: _____ TOTAL UBM(s): _____ (☐ >3 or ☐ ≤3) SITE STAFF INITIALS: _____

Appendix 3 CDAD DaySyms PRO

Instructions: These questions ask about symptoms you may have each day. Please complete the daily diary at about the same time every evening.

For each symptom listed below, **mark with an X the box** that best describes the **worst severity** of that symptom **during the past 24 hours**. Please be sure to answer each question.

	None	Mild	Moderate	Severe	Very Severe
1. Diarrhea	☐	☐	☐	☐	☐
2. Feeling that you needed to empty your bowels right away	☐	☐	☐	☐	☐
3. Needing to go to the bathroom more than usual	☐	☐	☐	☐	☐
4. Passing gas (flatulence)	☐	☐	☐	☐	☐
5. Abdominal cramping	☐	☐	☐	☐	☐
6. Abdominal pain	☐	☐	☐	☐	☐
7. Feeling bloated (feeling like you need to loosen your clothes)	☐	☐	☐	☐	☐
8. Feeling tired	☐	☐	☐	☐	☐
9 Lack of Energy	☐	☐	☐	☐	☐
10. Lightheadedness	☐	☐	☐	☐	☐
11. Dizziness	☐	☐	☐	☐	☐
12. Lack of appetite	☐	☐	☐	☐	☐
13. Nausea	☐	☐	☐	☐	☐

Appendix 4 Definitions of marked abnormalities for safety parameters

The following abnormalities are based on the Common Terminology Criteria for Adverse Events (CTCAE v4.03) – if a new version becomes available this is not implemented in the ongoing study but will be implemented in the following studies or in the Summary of Clinical Safety. Hematology, coagulation and chemistry abnormal values are defined based on the normal ranges from the Central Laboratory [Appendix 5].

An abnormality is considered treatment-emergent if it was not present at baseline and if a worsening occurred, i.e., a shift from normal or to a higher abnormality category, as compared to the corresponding value at baseline.

Safety Parameter	**LL**	**LLL**	**HH**	**HHH**
Hematology				
Hemoglobin (g/L)	< 100	< 80	Increase in (> 20 g/L) above ULN or above baseline if baseline is above ULN	Increase in (> 40 g/L) above ULN or above baseline if baseline is above ULN
Hematocrit (%)	< 28% F < 32% M	< 20%	> 60% M > 55% F	> 65%
Platelet count (10^9 /L)	< 75	< 50	> 600	> 999
Neutrophils (10^9 /L)	< 1.5	< 1.0		
Eosinophils (10^9 /L)			> 5.0	
Lymphocyte (10^9 /L)	< 0.8	< 0.5	> 4.9	> 20
WBC count (10^9 /L)	< 3.0	< 2.0	> 20.0	> 100.0
Coagulation				
INR			> 1.5 × ULN or > 1.5 × above baseline if on anticoagulation	> 2.5 × ULN or > 2.5 × above baseline if on anticoagulation
Chemistry				
AST (U/L)			> 3 ULN	> 5 ULN
ALT (U/L)			> 3 ULN	> 5 ULN
Alkaline phosphatase (U/L)			> 2.5 ULN	>5 ULN
Total bilirubin (umol/L)			> 2 ULN	> 5 ULN
Creatinine (umol/L)			> 1.5 ULN or 1.5 × baseline	> 3 ULN or 3.0 × baseline
Albumin (g/L)	< 30	< 20		
Calcium (mmol/L)	< 2.0	< 1.75	> 2.9	> 3.1

Potassium (mmol/L)	< 3.2	< 3.0	> 5.5	> 6.0
Sodium (mmol/L)		< 130	> 150	> 155
Vital signs				
SBP (mmHg)	SBP < 90 or decrease > 20 from baseline		SBP 140–159 or increase > 20 from baseline	SBP ≥ 160
DBP (mmHg)	DBP < 50 or decrease > 10 from baseline		DBP 90–99 or increase > 10 from baseline	DBP ≥ 100
ECG				
QT, QTc (msec) (absolute)			> 450 (H) > 480 (HH)	> 500
QT, QTc (msec) (change)			> 30	> 60

Appendix 5 Central Laboratory Normal Reference Ranges

"The ranges below are valid at the time of protocol finalization. Any changes to these ranges during the course of the study will be reflected in the ranges displayed in the laboratory reports sent from the Central Laboratory to the investigational sites."

Parameter	Sex	Unit	Normal Range	
			Low	High
Hematology				
Hemoglobin	M	g/L	130	175
	F	g/L	115	160
Hematocrit	M	L/L	0.416	0.541
	F	L/L	0.364	0.489
Platelets	M/F	$^{10^9}$/L	130	400
Erythrocytes	M	$^{10^{12}}$/L	4.50	5.90
	F	$^{10^{12}}$/L	3.80	5.20
Leukocytes	M/F	$^{10^9}$/L	4.5	11.0
Absolute Neutrophil Count	M/F	$^{10^9}$/L	1.8	7.7
Absolute Lymphocyte Count	M/F	$^{10^9}$/L	1.0	4.8
Absolute Monocyte Count	M/F	$^{10^9}$/L	0.1	0.8
Absolute Eosinophil Count	M/F	$^{10^9}$/L	0.0	0.5
Absolute Basophil Count	M/F	$^{10^9}$/L	0.0	0.2
Coagulation (Frozen Plasma)				
Protime	M/F	seconds	9.4	11.4
INR	M/F	N/A	0.90	1.10
aPTT	M/F	seconds	24.0	33.0
Blood chemistry				
Aspartate aminotransferase (AST)	M	U/L	14	39
	F	U/L	14	34
Alanine aminotransferase (ALT)	M	U/L	0	44
	F	U/L	0	33
Alkaline phosphatase	M	U/L	53	129
	F	U/L	42	98

Bilirubin, total	M/F	umol/L	5.1	20.5
Bilirubin, direct	M/F	umol/L	0.0	5.1
Calcium	M/F	mmol/L	2.15	2.55
Creatinine	M	umol/L	62	115
	F	umol/L	44	97
Urea	M/F	mmol/L	3.2	8.2
Sodium	M/F	mmol/L	136	145
Potassium	M/F	mmol/L	3.5	5.1
Chloride	M/F	mmol/L	99	109
Protein	M/F	g/L	64	83
Albumin	M/F	g/L	35	52

Appendix 6 Work Productivity and Activity Impairment Questionnaire

Clostridium difficile-associated diarrhea V2.0 (WPAI:CDAD)

The following questions ask about the effect of your *Clostridium difficile*-associated diarrhea on your ability to work and perform regular activities. *Please fill in the blanks or circle a number, as indicated.*

1. Are you currently employed (working for pay)? _____ NO ___ YES
   If NO, check 'NO' and skip to Question 6.

The next questions are about the **past seven days**, not including today.

2. During the past seven days, how many hours did you miss from work because of problems **associated with your *Clostridium-difficile* associated diarrhea**?

   Include hours you missed on sick days, times you went in late, left early, etc., because of your Clostridium-difficile associated diarrhea. Do not include time you missed to participate in this study.

   _____HOURS

3. During the past seven days, how many hours did you miss from work because of any other reason, such as vacation, holidays, time off to participate in this study?

   _____HOURS

4. During the past seven days, how many hours did you actually work?

   _____HOURS

   If '0', skip to Question 6.

5. During the past seven days, how much did your *Clostridium difficile*-associated diarrhea affect your productivity **while you were working**?

Think about days you were limited in the amount or kind of work you could do, days you accomplished less than you would like, or days you could not do your work as carefully as usual. If *Clostridium difficile*-associated diarrhea affected your work only a little, choose a low number. Choose a high number if *Clostridium difficile*-associated diarrhea affected your work a great deal.

Consider only how much ***Clostridium difficile*-associated diarrhea** affected productivity **while you were working**.

Clostridium difficile-associated diarrhea had no effect on my work	0	1	2	3	4	5	6	7	8	9	10	*Clostridium difficile*-associated diarrhea completely prevented me from working

CIRCLE A NUMBER

6. During the past seven days, how much did your *Clostridium difficile*-associated diarrhea affect your ability to do your regular daily activities, other than work at a job?

   By regular activities, we mean the usual activities you do, such as work around the house, shopping, childcare, exercising, studying, etc. Think about times you were limited in the amount or kind of activities you could do and times you accomplished less than you would like. If Clostridium difficile-associated diarrhea affected your activities only a little, choose a low number. Choose a high number if Clostridium difficile-associated diarrhea affected your activities a great deal.

Consider only how much ***Clostridium-difficile*** **associated diarrhea** affected your ability to do your regular daily activities, other than work at a job.

Clostridium-difficile associated diarrhea had no effect on my daily activities	0	1	2	3	4	5	6	7	8	9	10	*Clostridium-difficile* associated diarrhea completely prevented me from doing my daily activities

CIRCLE A NUMBER

WPAI:CDAD V2.0 (US English)

[Reilly 1993]

Appendix 7 Justification of the non-inferiority margin

Historical evidence to estimate the treatment effect of vancomycin against placebo is obtained from the results of two large randomized, double-blind, and controlled studies demonstrating the superiority of vancomycin over tolevamer [Louie 2007, Bouza 2008, Weiss 2009]. The cadazolid studies will be conducted in a similar population as the studies on tolevamer and the recently conducted studies to compare fidaxomicin versus vancomycin. Therefore, a similar 10% NI margin is justified and is consistent with the draft FDA Guidance for Industry Non-Inferiority Clinical Trials (Draft, March 2010), and the Center for Drug Evaluation and Research document as Statistical Review for the new drug application (NDA) of fidaxomicin (Application 201699Orig1s000).

The FDA has used the results of these two trials to estimate vancomycin's treatment effect while considering tolevamer as putative placebo. In the Statistical Review that the FDA has performed on the fidaxomicin Phase 3 results, additional analyses were performed on the two fidaxomicin trials 303 and 304, on publications by Louie [Louie 2006, Louie 2007], Bouza [Bouza 2008], and Weiss [Weiss 2009] and using the summary of the trials in clinicaltrials.gov. It included also a discussion of similarities and differences between the historical trials and current trials as well as a discussion of evidence from vancomycin placebo trials. Based on these results, the FDA considered the NI margin of 10% for the fidaxomicin trial as acceptable.

The treatment effect of vancomycin was estimated based on the data from two pivotal trials comparing vancomycin to tolevamer and showing the superiority of vancomycin over tolevamer.

Table 1 Summary of clinical success rate of historical trials (from Louie [Louie 2007] and Bouza [Bouza 2008]): Intent-to-Treat analysis [1]

Study	Agent	Clinical Cure rate		Treatment Difference (95% CI)[2]
		Number of subjects cured/Total	%	
301	Tolevamer	124/266	46.44	
	Vancomycin	109/134	80.74	35% (25%, 43%)
302	Tolevamer	112/268	41.64	
	Vancomycin	101/125	80.16	39% (29%, 47%)

1. Intent-to-Treat Set: includes all randomly assigned patients who received at least 1 dose of study drug, with any post-dosing Investigator Evaluation data.
2. Confidence interval was derived using method recommended in Newcombe 1998 and Agresti 2000.
CI = confidence interval.

Results from the Phase 3 fidaxomicin studies and the recently completed Actelion Phase 2 cadazolid study (AC-061A201) support an unchanged effect of vancomycin across studies and time. No decrease in the efficacy of vancomycin as measured by the clinical cure was observed and in the Phase 2 study. In this study, cadazolid had in the 250 mg treatment arm a higher rate of clinical cure than vancomycin on the modified clinical cure endpoint, which is the endpoint with the most similar definition to the primary endpoint used in the fidaxomicin pivotal studies.

Table 2 Summary of Clinical Cure rates of cadazolid and historical trials: mITT analysis set.

Study	**Agent**	**Clinical Cure Rate**		**Treatment Difference (95% CI)**
		Number of subjects cured / Total	**%**	
Cure Rate Fidaxomicin 003	Fidaxomicin	255/289	88.2	
	Vancomycin	258/307	84.0	4.2% (−1.4%, 9.7%)
Cure Rate Fidaxomicin 004	Fidaxomicin	217/253	85.8	
	Vancomycin	219/256	85.5	0.2% (−5.9%, 6.4%)
Modified Cure Rate Cadazolid AC-061201	Cadazolid 250 mg BID	16/17	94.1	
	Vancomycin 125 mg QID	19/22	86.4	

BID = twice daily; CI = confidence interval; QID = four times daily.

The cadazolid studies will be conducted on very similar patient population to the fidoxamicin and the tolevamer trials, and under very similar experimental conditions. Important design features, including patient populations, concomitant medications, and endpoint assessment, for establishing the constancy assumption are outlined and compared in Table 3.

Importantly, overall design characteristics are similar with all studies being international randomized, double-blind studies with vancomycin as the active comparator at a dose of 125 mg every 6 hours for 10 days. All studies are contemporaneous, having been conducted after 2005 when the epidemic hypervirulent strain 027 was identified in North America. One minor difference in the primary endpoint assessment is the Actelion's use of an objective, programmatic assessment of clinical cure at EOT + 2 days as opposed to a more subjective clinician reported outcome used in the other trials.

Table 3 Comparison of current studies to historical studies

Studies	Tolevamer	Fidaxomicin	Cadazolid AC-061A301/302
Overall study design	Randomized, double-blind, parallel groups.	Randomized, double-blind, parallel groups.	Randomized, double-blind, parallel groups.
Number of sites, countries	Study 301: 91 sites in the US and Canada. Study 302: 135 sites in Australia, Canada, and Europe.	Study 003: 75 sites in the US, 23 sites in Canada. Study 004: 30 sites in the US, 11 sites in Canada, 45 sites in Europe.	For each study: up to 100 centers in up to 20 countries (planned) among: AUS, AUT, BEL, BGR, CAN, CHE, CHN, DEU, DNK, ESP, FIN, FRA, GBR, HUN, ITA, MEX, NZL, POL, ROU, SWE, USA.
Treatment arms	Vancomycin Tolevamer Metronidazole	Vancomycin Fidaxomicin	Vancomycin Cadazolid
Randomization scheme	1:2:1	1:1	1:1
Total number of subjects	1420 (ITT)	1105 (mITT)	Approximately 1280 subjects randomized.
Start date-end date	301: March 2005 to February 2007. 302: May 2005 to August 2007.	003: 9 May 2006 to 21 August 2008. 004: 19 April 2007 to 11 December 2009.	3rd Quarter 2013. 4th Quarter 2015.
Duration of treatment	10 days	10 days	10 days
Assessments	Daily Assessments	Daily Assessments	Daily Assessments
Test of Cure	Day 10	EOT to Day 12	EOT + 2 days

Definition of Clinical Cure	Resolution of diarrhea *(not defined in detail)* and absence of severe abdominal discomfort due to CDAD for 2 contiguous days including Day 10.	- Subjects who, in the opinion of the Investigator, require no further CDAD therapy 2 days after completion of study medication. - Subjects who have 3 or fewer unformed stools for 2 consecutive days and remain well prior to the time of study medication discontinuation. - Subjects who at EOT have had a marked reduction in the number of unformed stools and who have residual and mild abdominal discomfort interpreted as recovering bowel, providing no new antiinfective CDAD therapy has been initiated.	- Resolution of Diarrhea ROD (≤ 3 UBMs per day for at least 2 consecutive days) on study treatment and maintained for 2 days after EOT).
Inclusion criteria	- Adult patients - Acute CDAD (mild, moderate, or severe), + Mild: 3–5 bowel movements per day, WBC less or equal to 1500/mm^3 and no abdominal pain, + Moderate: 6–9 bowel movements per day, WBC 1501–20000/mm^3 and no, mild or moderate abdominal pain, + Severe: 10 or more bowel movements per day, WBC greater or equal to 20001/mm^3 and severe abdominal pain. - 1st occurrence or recurrent.	- Male or female ≥ 16 years - CDAD (> 3 UBMs within 24 hours and positive *C. difficile* toxin A/B test within 48 hours of randomization for metronidazole failures, or within 96 hours of randomization for subjects with ≤ 24 hours pretreatment of CDAD). - 1st occurrence or 1st recurrence within 3 months. - Subjects meeting CDAD criteria after at least a full 3 day course of metronidazole.	- Male or female ≥ 18 years - CDAD (> 3 UBMs within 24 hours and positive *C. difficile* toxin A/B test within 72 hours). - 1st occurrence or 1st recurrence within 3 months. - Subjects meeting CDAD criteria (with a positive test within 48 hours) after at least 72 hours of treatment with metronidazole (MTF).

Exclusion criteria	Not known	- Life-threatening or fulminant CDAD, + septic shock, + peritoneal signs, + significant dehydration, + systolic blood pressure < 90mmHg, + WBC count > 30000 /mm^3, + body temperature > 40 °C. - Toxic megacolon. - Likelihood of death within 72 hours from any cause. - History of ulcerative colitis or Crohn's disease. - No more than 24 hours of treatment with metronidazole and/or vancomycin. - Opiates are permitted as needed (PRN) as long as subjects taking them are on stable doses at the time of randomization and expected to maintain these doses during the treatment period. - The anticipated need to continue other antibacterials for a period exceeding 7 days. - Subjects who, in the opinion of the investigator, require other drugs to control diarrhea (e.g., loperamide) or drugs that could affect peristalsis. - Participation in other clinical research studies utilizing an investigational agent within 1 month	- Fulminant or life-threatening CDAD, + septic shock, + peritonitis, + significant dehydration, + systolic blood pressure < 90mmHg, + WBC count > 30000/mm^3, + body temperature > 40 °C, + ileus, + toxic megacolon. - Concurrent life-threatening disease or condition (likelihood of death within 72 hours). - History of ulcerative colitis, microscopic colitis or Crohn's disease, chronic abdominal pain or chronic diarrhea of any etiology. - History of total colectomy. - Vomiting, inability to take oral medication (subjects with feeding tubes are excluded). - Antimicrobial treatment active on CDAD > 24 hours for the current episode, except MTF. - Planned treatment with prohibited concomitant medications: + cholestyramine, fecal transplant, probiotics until Visit 5, + Initiation or change in dose or regimen resulting in an increased opiate effect up to TOC, + Anti-peristaltic medications, kaolin, pectin or charcoal-containing

		before screening or within 5 half-lives of the investigational agent, whichever was longer. - Previous exposure to fidaxomicin. - Subjects the investigator believes are inappropriate for the trial, e.g., subjects with known hypersensitivity to vancomycin.	anti-diarrheals until EOS. - Fecal transplant, treatment with immunoglobulin or another investigational drug within 1 month. - Treatment with monoclonal antibodies against *Clostridium difficile* within 6 months. - Investigational vaccination against *Clostridium difficile*. - Previous participation in a clinical trial with cadazolid. - Known hypersensitivity or contraindication to any excipients of the investigational drug formulation, to an oxazolidinone, a quinolone or to vancomycin.

CDAD = *clostridium difficile*-associated diarrhea; EOS = end-of-study; EOT = end-of-treatment; ITT = intent-to-treat; mITT = modified intent-to-treat; MTF = metronidazole treatment failure; PRN = *pro re nata* (as necessary); ROD = resolution of diarrhea; TOC = test-of-cure; UBM = unformed bowel movement; WBC = white blood cell.

As it is assumed that tolevamer response rates are similar to and no worse than placebo, the estimated treatment effect of vancomycin versus tolevamer presents a conservative estimate of the treatment effect of vancomycin versus placebo in case of a small treatment effect of tolevamer.

A meta-analysis of the results using the DerSimonian and Laird approach (random effect model) gives an estimate of the vancomycin treatment effect of 37% with 95% CI of (30%, 43%).

To account for the uncertainties in the constancy assumption, the lower CI of the vancomycin treatment effect estimate from the tolevamer meta-analysis of 30% is discounted by 10%, resulting in an estimate for the vancomycin treatment effect of M1 = 27% (M1 defined as the entire effect of active control assumed to be present in the study, see FDA 2010) . The proposed NI-margin M2 is 10% which is generally considered clinically acceptable to show non-inferiority in antibacterial trials (M2 is defined as the largest clinically acceptable difference of test drug compared to active

control, see FDA 2010). This still preserves at least 60% (17%/27% = 63.0%) of the vancomycin treatment effect of 27%. In addition, the true treatment effect of vancomycin versus placebo is probably larger than the estimate, since tolevamer may have some limited clinical benefit.

Therefore, taking into account all the above consideration, the proposed NI margin of 10% will preserve more than 60% of the treatment effect of vancomycin, which is the minimal recommendation in [FDA 2010].