Integrated Review

Table 1. Application Information

Application type	NDA
Application number	219491
Priority or standard	PRIORITY
Submit date	4/15/2025
Received date	4/15/2025
PDUFA goal date	12/15/2025
Division/office	Division of Anti-Infectives (DAI)
Review completion date	12/10/2025
Established/proper name	Zoliflodacin
(Proposed) proprietary name	NUZOLVENCE
Pharmacologic class	Spiropyrimidinetrione bacterial type II topoisomerase inhibitor
Other product name(s)	ETX0914, AZD0914
Applicant	ENTASIS THERAPEUTICS INC
Dosage form/formulation	For Oral Suspension: each unit-dose packet of white to off-white granules contains 3 g of zoliflodacin
Dosing regimen	3 g orally, single dose
Applicant-proposed indication(s)/ population(s)	Treatment of uncomplicated gonorrhea due to *Neisseria gonorrhoeae* in adult and pediatric patients 12 years and older, weighing at least 35 kg
SNOMED CT code for proposed indication disease term(s)[1]	15628003 |Gonorrhea (disorder)|
Regulatory action	Approval
Approved dosage (if applicable)	3 g orally, single dose
Approved indication(s)/ population(s) (if applicable)	Treatment of uncomplicated urogenital gonorrhea due to *Neisseria gonorrhoeae* in adults and pediatric patients 12 years of age and older, weighing at least 35 kg
SNOMED CT code for approved indication disease term(s)[1]	15628003 | Gonorrhea (disorder)

[1] For internal tracking purposes only.
Abbreviations: PDUFA, Prescription Drug User Fee Act; SNOMED CT, Systematized Nomenclature of Medicine Clinical Terms

Table of Contents

Table of Tables

Table of Figures

Table of Equations

Glossary

ACKR1	atypical chemokine receptor 1
ADME	absorption, distribution, metabolism, and excretion
AE	adverse event
ALT	alanine aminotransferase
AST	antimicrobial susceptibility testing
AUC	area under the concentration-time curve
$AUC_{0-\infty}$	area under the concentration-time curve estimated to infinity
BA	bioavailability
BCRP	breast cancer resistance protein
CI	clinical inspection
CL	clearance
CLSI	Clinical Laboratory and Standards Institute
C_{max}	maximum plasma concentration
CMC	chemistry, manufacturing, and controls
COVID-19	coronavirus disease of 2019
CYP	cytochrome P450
DANC	Duffy-null associated neutrophil count
DARRTS	Document Archiving, Reporting and Regulatory Tracking System
DDI	drug-drug interaction
DMID	Division of Microbiology and Infectious Diseases
DNDi	Drugs for Neglected Diseases initiative
DUOG	Division of Urology, Obstetrics and Gynecology
ECG	electrocardiogram
EFD	embryo-fetal development
EOS	end-of-study
EPC	established pharmacologic class
f	free-drug
F1	first generation
F2	second generation
F_a	fraction absorbed
FDA	Food and Drug Administration
FE	food effect
FEFD	fertility and embryo-fetal development
F_g	fraction available
$GABA_A$	gamma-aminobutyric acid A
GARDP	Global Antibiotic Research and Development Partnership
GCP	good clinical practice
GD	gestation day
GFOS	granules for oral suspension
GMR	geometric mean ratio
hERG	human ether-a-go-go related gene
HFIM	hollow fiber infection model
IC_{50}	half-maximal inhibitory concentration

ICH	International Council for Harmonisation
IIV	interindividual variability
IM	intramuscular
IND	investigational new drug
iPSP	initial Pediatric Study Plan
IR	information request
IRB	institutional review board
IRT	interdisciplinary review team
IV	intravenous
k_a	absorption rate constant
Ki,u	enzyme competitive inhibition constant for unbound drug
K_p	tissue-to-plasma partition coefficient
LC-MS/MS	liquid chromatography with tandem mass spectrometry
MATE	multidrug and toxin extrusion transporter
MIC	minimum inhibitory concentration
Micro-ITT	microbiological intent-to-treat
Micro-MITT	microbiological modified intent-to-treat
MLST	multilocus sequence typing
MRHD	maximum recommended human dose
MRT	mean residence time
MTT	mean transit time
NAAT	nucleic acid amplification test
NDA	new drug application
NI	noninferiority
NIAID	National Institute of Allergy and Infectious Diseases
NG	*Neisseria gonorrhoeae*
NOAEL	no-observed-adverse-effect level
OAT	organic anion transporter
OATP	organic anion transporting polypeptide
OCMQ	Office of New Drugs custom medical query
OCT2	organic cation transporter 2
OPQ	Office of Pharmaceutical Quality
OSI	Office of Scientific Investigations
OSIS	Office of Study Integrity and Surveillance
PBPK	physiologically based pharmacokinetic
PD	pharmacodynamic
PFOS	powder for oral suspension
P-gp	P-glycoprotein
PI	Prescribing Information
PK	pharmacokinetic
PMR	postmarketing requirement
PO	per oral
PopPK	population pharmacokinetic
PP	per-protocol
PPB	plasma protein binding
PT	preferred term

PTA	probability of target attainment
QC	quality control
QIDP	Qualified Infectious Disease Product
QTc	corrected QT interval
QTcF	QT interval corrected for heart rate using Fridericia's formula
ΔΔQTcF	change of QTcF from Baseline
RBC	red blood cell
SAD	single ascending dose
SAE	serious adverse event
SEE	substantial evidence of effectiveness
SOC	system organ class
STAR	sequence typing for antimicrobial resistance
$t_{1/2}$	half-life
TDD	total daily dose
TEAE	treatment-emergent adverse event
TK	toxicokinetic
T_{max}	time to maximum plasma concentration
TOC	test-of-cure
TQT	thorough QT
ULN	upper limit of normal
Vc	volume of central compartment
V_{ss}	volume of distribution at steady state
WBC	white blood cell
ZoliAZ	zoliflodacin manufactured by AstraZeneca
ZoliDr	zoliflodacin manufactured by Dr Reddy’s Laboratories, Ltd
ZoliPa	zoliflodacin manufactured by Patheon

I. Executive Summary

1. Overview

1.1. Summary of Regulatory Action

On April 15, 2025, Entasis Therapeutics, Inc. (the Applicant) submitted a 505(b)(1) NDA for zoliflodacin, a spiropyrimidinetrione bacterial type II topoisomerase inhibitor intended to treat uncomplicated gonorrhea due to *Neisseria gonorrhoeae* (NG) in adult and pediatric patients 12 years and older, weighing at least 35 kg. Zoliflodacin is a new molecular entity that inhibits bacterial DNA gyrase and topoisomerase IV (type II topoisomerases) and demonstrates in vitro and clinical activity against *N. gonorrhoeae*. Zoliflodacin for oral suspension was developed as a 3 g single dose for oral administration with a half-life ($t_{1/2}$) of about 6 hours.

Zoliflodacin has Qualified Infectious Disease Product (QIDP) designation and underwent Priority Review. The Prescription Drug User Free Act goal date is December 15, 2025. The drug was developed by the following entities under individual IND applications which cross-referenced each other: AstraZeneca under IND 118958, later transferred to Entasis in 2015, the National Institute of Allergy and Infectious Diseases (NIAID) under IND (b) (4), and the Global Antibiotic Research and Development Partnership (GARDP) Foundation under IND 139105. NIAID and the GARDP Foundation conducted the phase 2 (Division of Microbiology and Infectious Diseases [DMID] 14-0014) and phase 3 (STI_Zoli001) trials, respectively.

The primary evidence for clinical efficacy was provided from a phase 3, international, multicenter, open-label, randomized, controlled, noninferiority (NI) trial (Trial STI_Zoli001) conducted in participants 12 years of age and older with body weight ≥35 kg diagnosed with uncomplicated gonorrhea. Participants were randomized 2:1 to receive a single 3 g oral dose of zoliflodacin or a single 500 mg intramuscular (IM) dose of ceftriaxone in combination with a single 1 g oral dose of azithromycin. Participants were eligible for enrollment if they had signs, symptoms, or laboratory confirmation of urethral or cervical gonorrhea, or had unprotected sexual contact within 14 days prior to screening with an individual confirmed to have NG infection.

The highest proportion of enrolled participants in the phase 3 trial were from South Africa (46%), followed by Thailand (29%), the United States (17%) and the European Union (8%). The majority of participants treated with zoliflodacin were male (88%). Participants treated with zoliflodacin identified as Black or African American (56%), Asian (31%), White (11%), American Indian or Alaska Native (1%), or Other (1%). A total of 3% of zoliflodacin treated participants identified as Hispanic or Latino and 22% were living with HIV.

The primary analysis population was the microbiologic intent-to-treat (urogenital) population [micro-ITT (urogenital)], which included all randomized participants with *N. gonorrhoeae* isolated at Baseline from urogenital sites and whose baseline culture result showed susceptibility to either ceftriaxone or azithromycin. The primary endpoint was microbiological cure rate at the test-of-cure (TOC) Visit (Day 6±2) in the micro-ITT (urogenital) population. FDA guidance for

industry *Uncomplicated Gonorrhea: Developing Drugs for Treatment*[1] recommends a NI margin of 10% for gonorrhea clinical trials, but the Division of Anti-Infectives (Division) agreed to a 12% NI margin in view of the significant need for drugs for treatment of gonorrhea,[2] including strains resistant to current standard of care antibacterial therapy.

In Trial STI_Zoli001, the microbiological cure rate was 90.9% (460/506) for zoliflodacin compared to 96.2% (229/238) for the control arm, with a difference of −5.3% (95% CI: −8.6%, −1.4%). Zoliflodacin met the prespecified 12% NI margin and achieved an NI margin of 10% for the primary efficacy endpoint. Although the trial included participants with urogenital, pharyngeal, and rectal gonorrhea, the primary endpoint of the trial was microbiological cure as determined by culture at the urogenital sites. No strategies were implemented to control overall type I error for testing of the secondary endpoints, which included microbiological cure at the pharyngeal and rectal sites. In addition, the analyses of secondary endpoints at the pharyngeal and rectal sites were not powered and were based on relatively small sample sizes. Thus, in the Prescribing Information (PI), the Division modified the Applicant's proposed indication to indicate zoliflodacin for the treatment of uncomplicated urogenital gonorrhea due to *N. gonorrhoeae* in adults and pediatric patients 12 years of age and older, weighing at least 35 kg.

Confirmatory evidence was obtained from a phase 2, randomized, open-label trial (Trial DMID 14-0014), in which a single oral dose of either 2 g or 3 g of zoliflodacin was compared to a single 500 mg IM dose of ceftriaxone for the treatment of uncomplicated gonorrhea in adult participants. The primary endpoint and the primary analysis population were identical to the phase 3 trial. The phase 2 trial results were supportive, with cure rates of 96.5% (55/57) and 96.4% (54/56) for the 2 g and 3 g zoliflodacin doses, respectively, compared to 100% (28/28) for ceftriaxone.

The safety database was composed of 995 participants across eight phase 1 to 3 clinical studies and trials who received zoliflodacin; 782 participants received zoliflodacin at the proposed single oral dose of 3 g, including 686 participants in the phase 2 and phase 3 trials and 96 healthy participants in phase 1 studies. The safety profile of zoliflodacin in the clinical development program was well-characterized and acceptable, with the adverse events (AEs) of neutropenia, headache, leukopenia, dizziness, nausea, and diarrhea occurring in greater than or equal to 2% of participants in the zoliflodacin arm of Trial STI_Zoli001.

However, zoliflodacin demonstrated reproductive toxicity in nonclinical repeat-dose studies, including embryo-fetal toxicity (exencephaly) in gravid mice, reduced fertility in male rats administered zoliflodacin prior to mating, increased embryonic loss in untreated female rats mated with male rats administered zoliflodacin, and testicular toxicity (decreased spermatogenesis and degeneration) in male rats and dogs. The fertility effects were reversible

[1] FDA guidance for industry *Uncomplicated Gonorrhea: Developing Drugs for Treatment* (August 2015). Guidances are updated periodically. For the most recent version of a guidance, check the FDA guidance web page at https://www.fda.gov/regulatory-information/search-fdaguidance-documents.

[2] Hiruy, H, S Bala, JM Byrne, KG Roche, SH Jang, P Kim, S Nambiar, D Rubin, Y Yasinskaya, LH Bachmann, K Bernstein, R Botgros, S Cammarata, RL Chaves, CD Deal, GL Drusano, EM Duffy, AE Eakin, S Gelone, T Hiltke, EW Hook Iii, AE Jerse, CJ McNeil, L Newman, S O'Brien, C Perry, HEL Reno, RA Romaguera, J Sato, M Unemo, TEC Wi, K Workowski, GA O'May, SJ Shukla, and JJ Farley, 2024, FDA, CDC, and NIH Co-sponsored Public Workshop Summary-Development Considerations of Antimicrobial Drugs for the Treatment of Gonorrhea, Clin Infect Dis, https://www.ncbi.nlm.nih.gov/pubmed/39045871.

within 28 days, but testicular histopathology showed only partial recovery 3 months following cessation of zoliflodacin administration.

While available data from the phase 2 and 3 trials indicate that zoliflodacin's safety profile is acceptable for its intended use as a single-dose treatment, potential safety risks related to the nonclinical signals will be described in zoliflodacin's labeling along with approaches to risk mitigation. These include Warnings and requirements for pregnancy testing in females of reproductive potential prior to zoliflodacin administration, contraception for males with female partners of reproductive potential for 3 months following administration of zoliflodacin, and a Medication Guide. Additionally, postmarketing requirements (PMRs) for a descriptive pregnancy safety study to evaluate pregnancy outcomes in females exposed to zoliflodacin during pregnancy, a human sperm study to evaluate the impact of zoliflodacin administration on sperm count and viability, a clinical lactation study, and microbiological surveillance for the development of resistance to zoliflodacin will be issued.

A population pharmacokinetic (PopPK) analysis based on pharmacokinetic (PK) data from phase 1 studies and the phase 3 trial showed that weight is a significant covariate of zoliflodacin pharmacokinetics in humans, with an inverse relationship between body weight and zoliflodacin exposure. Further, exposure is increased when administered with food. Thus, zoliflodacin will be administered as a single 3 g dose of zoliflodacin granules for suspension with food, in patients weighing ≥50 kg and without food in patients weighing 35 kg to <50 kg. Additionally, concentration-dependent corrected QT interval (QTc) prolongation was observed in the thorough QT (TQT) study. Although the totality of evidence suggests that clinically significant QTc prolongation is not anticipated for a single dose of 3 g of zoliflodacin when administered with or without food to patients weighing >35 kg, the Applicant proposed that administration of zoliflodacin under fasted conditions would reduce the likelihood of QTc prolongation in patients weighing >35 kg and <50 kg. The Applicant's proposal is acceptable to the review team.

Overall, the review team and signatory authority have concluded that the benefits of zoliflodacin outweigh its associated risks for the treatment of uncomplicated urogenital gonorrhea due to *N. gonorrhoeae* in adult and pediatric patients 12 years and older, weighing at least 35 kg, and recommend approval.

1.2. Conclusions on Substantial Evidence of Effectiveness

Substantial evidence of effectiveness (SEE) was established with one adequate and well-controlled clinical investigation and confirmatory evidence.

Substantial evidence of effectiveness (SEE) for zoliflodacin for the treatment of uncomplicated urogenital gonorrhea was established based on one randomized, active-controlled phase 3 clinical trial (STI_Zoli001), with confirmatory evidence from the phase 2 trial (DMID 14-0014).

In Trial STI_Zoli001, zoliflodacin was noninferior to dual therapy with ceftriaxone and azithromycin for the treatment of uncomplicated urogenital gonorrhea. The primary endpoint was the microbiological cure rate at the urogenital site at the TOC Visit (Day 6±2) in the micro-ITT (urogenital) population (participants with *N. gonorrhoeae* isolated at Baseline from urogenital sites with baseline testing showing susceptibility to ceftriaxone or azithromycin). The

microbiological cure rate was 90.9% (460/506) for zoliflodacin compared to 96.2% (229/238) for the control arm, with a difference of −5.3% (95% CI: −8.6%, −1.4%). Zoliflodacin met the prespecified 12% margin and achieved an NI margin of 10% for the primary efficacy endpoint.

Confirmatory evidence of efficacy for treatment of uncomplicated urogenital gonorrhea was obtained from Trial DMID 14-0014. In Trial DMID 14-0014, the primary endpoint and the primary efficacy population were identical to the phase 3 trial. The trial results were supportive, with cure rates of 96.5% (55/57) and 96.4% (54/56) for the 2 g and 3 g zoliflodacin doses, respectively, compared to 100% (28/28) for ceftriaxone.

In summary, SEE for zoliflodacin for the treatment of uncomplicated urogenital gonorrhea has been provided by statistical noninferiority in the micro-ITT (urogenital) population of Trial STI_Zoli001 with confirmatory evidence from Trial DMID 14-0014.

2. Benefit-Risk Assessment

2.1. Benefit-Risk Framework

Table 2. Benefit-Risk Framework

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Analysis of condition	• Gonorrhea is caused by the bacterium *Neisseria gonorrhoeae*. • *N. gonorrhoeae* is a strictly human pathogen transmitted primarily by sexual contact but also via vertical transmission from mother to infant during vaginal delivery. The bacterium may cause symptomatic or asymptomatic infections.[3] • Gonorrhea is the second most commonly reported bacterial sexually transmitted infection (STI) in the United States with over 600,000 cases reported in 2023.[4] • The Centers for Disease Control and Prevention (CDC) estimate approximately 1.6 million new infections occur each year.[5] • If left untreated, gonorrhea can cause serious and permanent health problems such as pelvic inflammatory disease (PID) and ectopic pregnancy in women, and infertility and disseminated infections in both men and women.[6]	Gonorrhea is an STI that impacts both males and females. If left untreated, this infection can lead to complications including PID and infertility.

[3] WHO. "Gonorrhoea (*Neisseria gonorrhoeae* infection)." 4 July 2024. https://www.who.int/news-room/fact-sheets/detail/gonorrhoea-(neisseria-gonorrhoeae-infection)

[4] CDC. "National Overview of STIs in 2023." November 12, 2024. https://www.cdc.gov/sti-statistics/annual/summary.html.

[5] Workowski, K. A., Bachmann, L. H., Chan, P. A., Johnston, C. M., Muzny, C. A., Park, I., Reno, H., Zenilman, J. M., & Bolan, G. A. (2021). Sexually Transmitted Infections Treatment Guidelines, 2021. MMWR Recomm Rep, 70(4), 1-187. https://doi.org/10.15585/mmwr.rr7004a1

[6] Quilter LAS, St Cyr SB, Barbee LA. The Management of Gonorrhea in the Era of Emerging Antimicrobial Resistance: What Primary Care Clinicians Should Know. Med Clin North Am. 2024 Mar;108(2):279-296. doi: 10.1016/j.mcna.2023.08.015. Epub 2023 Sep 14. PMID: 38331480; PMCID: PMC11150008.

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Current treatment options	• The first line treatment for gonococcal infections in the United States is one 500 mg IM dose of ceftriaxone.[7] – Cefixime as a single 800 mg oral dose is an alternative regimen if ceftriaxone is unavailable. Cefixime is less efficacious than ceftriaxone for pharyngeal gonorrhea.[8] – For individuals with a cephalosporin allergy, treatment with one 240 mg IM dose of gentamicin and one 2 g dose of oral azithromycin is recommended. • In 2013, the CDC classified cephalosporin-resistant *N. gonorrhoeae* as an urgent public health threat.[9] • The first multidrug resistant (MDR) *N. gonorrhoeae* strain was identified in Japan in 2015 and has been reported in several European and Asian countries.[10] • Antimicrobial resistance may complicate selection of appropriate antibacterial treatment for gonorrhea.	The recommended first-line treatment option for gonorrhea infections in the United States is ceftriaxone. Antibacterial resistant strains of *N. gonorrhoeae* are on the rise with limited effective treatment options.
Benefit	• The clinical efficacy of zoliflodacin was demonstrated in one phase 3 adequate and well-controlled trial (Trial STI_Zoli001). • The primary efficacy endpoint was the microbiological cure rate of uncomplicated urogenital gonorrhea at TOC (6±2 days) in the micro-ITT (urogenital) population. • The microbiological cure rate was 90.9% (460/506) for zoliflodacin compared to 96.2% (229/238) for the control arm, with a difference of −5.3% (95% CI: −8.6%, −1.4%). • In STI_Zoli001, zoliflodacin met the prespecified 12% NI margin and achieved an NI margin of 10% for the primary efficacy endpoint at TOC in the micro-ITT (urogenital) population. • Confirmatory evidence was obtained from a phase 2, randomized, controlled clinical trial (DMID 14-0014) that demonstrated microbiologic cure rates of 96.5% (55/57) and 96.4% (54/56) in the 2 g and 3 g arms of zoliflodacin, respectively, compared to 100% (28/28) in the comparator arm (ceftriaxone 500 mg IM).	Zoliflodacin was noninferior to ceftriaxone and azithromycin for the treatment of uncomplicated urogenital gonorrhea.

[7] CDC. "Gonococcal Infections Among Adolescents and Adults." September 21, 2022. https://www.cdc.gov/std/treatment-guidelines/gonorrhea-adults.htm

[8] Yang KJ, Kojima N, Bristow CC, Klausner JD. Effectiveness of Cefixime for the Treatment of *Neisseria gonorrhoeae* Infection at 3 Anatomic Sites: A Systematic Review and Meta-Analysis. Sex Transm Dis. 2023 Mar 1;50(3):131-137. doi: 10.1097/OLQ.0000000000001742. Epub 2022 Dec 13. PMID: 36729626; PMCID: PMC9906985.

[9] CDC. "Drug-resistant Gonorrhea." https://www.cdc.gov/gonorrhea/hcp/drug-resistant/index.html.

[10] Raccagni AR, Ranzenigo M, Bruzzesi E, Maci C, Castagna A, Nozza S. *Neisseria gonorrhoeae* Antimicrobial Resistance: The Future of Antibiotic Therapy. Journal of Clinical Medicine. 2023; 12(24):7767. https://doi.org/10.3390/jcm12247767

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Risk and risk management	• The safety database contained 995 participants who received oral zoliflodacin, including 782 who received the 3 g dose in the phase 1 studies and the phase 2 and 3 trials. • There were no serious adverse events (SAE) and no deaths in the phase 1 studies and the phase 2 and 3 trials. • The most common adverse events were low neutrophil and white cell counts, headache, dizziness, diarrhea and nausea. • In nonclinical studies, testicular toxicity and decreased spermatogenesis were found in male rats and dogs dosed for 2 days to 4 weeks. Fertility was fully reversible at 28 days; however, the histopathology was only partially reversible at 2 to 3 months. Additionally, in rat studies, pregnancy losses in female rats increased when females were mated with males dosed with zoliflodacin. • In nonclinical mouse studies, the fetuses of gravid mice dosed with zoliflodacin during organogenesis had a higher rate of exencephaly as compared to historical or concurrent controls. • Surveillance studies and the clinical trials (phase 2 and 3) showed that the upper bound of the wild-type population of zoliflodacin MIC is 0.5 mg/L.	The safety profile for zoliflodacin in humans is acceptable for the treatment of uncomplicated urogenital gonorrhea in adult and pediatric patients 12 years of age and older, weighing 35 kg. A Warning will be included in labeling to communicate the potential risk of male testicular toxicity and adverse effects on male fertility based on findings in nonclinical studies. Based on the same nonclincial studies, another Warning will be included to communicate the risk of embyonic loss in female partners of reproductive potential of males treated with zoliflodacin. Additionally, male contraception will be recommended for 3 months following treatment with zoliflodacin. A Warning will be included in labeling to communicate the risk of embryo-fetal toxicity, including exencephaly, in pregnant women treated with zoliflodacin. A recommendation to obtain a pregnancy test prior to administration of zoliflodacin will be included in labeling. A Medication Guide for patients will be required to ensure all patients receive information on

Dimension	Evidence and Uncertainties	Conclusions and Reasons
		the potential risks associated with zoliflodacin. A descriptive pregnancy safety study (DPSS),a human sperm study, a lactation study, and a microbiological surveillance study to assess for the development of resistance to zoliflodacin will be postmarketing requirements.

Abbreviations: AE, adverse event; IM, intramuscular; NI, noninferiority; TOC, test-of-cure

2.2. Conclusions Regarding Benefit-Risk

In NDA 219491, the Applicant submitted safety and efficacy data to support the use of zoliflodacin for the treatment of uncomplicated urogenital gonorrhea caused by *N. gonorrhoeae* in adult and pediatric patients 12 years and older, weighing at least 35 kg. Zoliflodacin is a novel first-in-class spiropyrimidinetrione bacterial type II topoisomerase inhibitor that inhibits bacterial DNA gyrase and topoisomerase IV. Zoliflodacin addresses a need for additional options for the treatment of *N. gonorrhoeae*, including multidrug-resistant strains, potentially providing an important alternative for treatment as resistance to current treatments continues to emerge.

Evidence of clinical efficacy was provided by Trial STI_Zoli001, a phase 3, adequate and well-controlled clinical trial of zoliflodacin at the proposed single dose of 3 g compared with the standard of care treatment at the time the trial was designed, i.e., ceftriaxone 500 mg IM and azithromycin 1 g per oral (PO). Only participants with isolates with known or suspected susceptibility to ceftriaxone or azithromycin on culture were randomized to a treatment arm. Although the FDA guidance specifies a 10% NI margin for clinical trials for uncomplicated gonorrhea,[11] the Division agreed to a 12% NI margin in view of the need for treatment options for this infection.[12]

The primary efficacy endpoint assessed microbiological response at TOC (6±2 days) in participants with uncomplicated urogenital gonorrhea. The microbiological cure rate was 90.9% (460/506) in the zoliflodacin arm and 96.2% (229/238) in the ceftriaxone-azithromycin arm with a treatment difference of −5.3% (95% CI: −8.6%, −1.4%). Thus, zoliflodacin demonstrated noninferiority to the comparator as it met the pre-specified 12% NI margin and achieved a 10% NI margin. It was noted that zoliflodacin was statistically inferior to ceftriaxone-azithromycin, with a treatment difference of −5.3% (95% CI: −8.6%, −1.4%). Given the need for additional treatments for gonorrhea, including drug-resistant strains, the use of two potentially effective drugs in the comparator arm, the overall high efficacy rate of >90% in both treatment arms, and meeting a 10% NI margin (narrower than the pre-specified 12% NI margin), the imbalance in failure rates disfavoring zoliflodacin was considered to be clinically acceptable.

Confirmatory evidence was provided from a phase 2, randomized, controlled clinical trial, with microbiologic cure rates of 96.5% (55/57) and 96.4% (54/56) in the 2 g and 3 g arms of zoliflodacin, respectively, compared to 100% (28/28) in the comparator arm (ceftriaxone 500 mg IM). No formal statistical hypothesis testing was performed for this trial. All efficacy endpoints were summarized descriptively.

The safety database included 995 participants who received at least one dose of zoliflodacin, ranging from 200 mg to 4 g. In total, 782 participants received the 3 g dose of zoliflodacin, including 686 participants from the phase 2 and phase 3 trials and 96 healthy participants in the phase 1 studies. The available data indicate that zoliflodacin's safety profile is acceptable for the

[11] See Footnote 1

[12] Hiruy, H, S Bala, JM Byrne, KG Roche, SH Jang, P Kim, S Nambiar, D Rubin, Y Yasinskaya, LH Bachmann, K Bernstein, R Botgros, S Cammarata, RL Chaves, CD Deal, GL Drusano, EM Duffy, AE Eakin, S Gelone, T Hiltke, EW Hook Iii, AE Jerse, CJ McNeil, L Newman, S O'Brien, C Perry, HEL Reno, RA Romaguera, J Sato, M Unemo, TEC Wi, K Workowski, GA O'May, SJ Shukla, and JJ Farley, 2024, FDA, CDC, and NIH Co-sponsored Public Workshop Summary-Development Considerations of Antimicrobial Drugs for the Treatment of Gonorrhea, Clin Infect Dis, https://www.ncbi.nlm.nih.gov/pubmed/39045871.

treatment of uncomplicated urogenital gonorrhea in adults and adolescents weighing 35 kg or more. Adverse events observed in the clinical trials were mostly mild and self-limited, with low neutrophil counts and headaches being the most commonly reported events (10.1% and 9.9%, respectively) versus 13% and 5%, respectively, in the comparator arm. Additional AEs with an incidence between 2% and 5% in the zoliflodacin arm included low white blood cell count, dizziness, nausea, and diarrhea.

In the nonclinical studies, zoliflodacin demonstrated reproductive toxicity in nonclinical repeat-dose general toxicity and developmental and reproductive studies, including:

- Embryo-fetal toxicity (exencephaly) in gravid mice
- Reduced fertility in male rats administered zoliflodacin prior to mating at 4-fold or greater than the maximum recommended human dose
- Increased embryonic loss in female rats and mice administered zoliflodacin
- Testicular toxicity (decreased spermatogenesis and degeneration) in male rats and dogs dosed for 2 days to 4 weeks
- Increase in fetal loss in rat studies where untreated females were mated with males treated with zoliflodacin
- Decreased litter weights in rat and mouse pregnancy studies

The male fertility effects were fully reversible within 28 days, but testicular histopathology showed only partial recovery 3 months following cessation of zoliflodacin administration. There were no animal or human data on potential secretion of zoliflodacin into breast milk. The potential safety risks related to the nonclinical signals will be described in zoliflodacin's labeling along with approaches to risk mitigation, including Warnings, requirements for pregnancy testing in females of reproductive potential prior to zoliflodacin administration, contraception for males with female partners of reproductive potential for 3 months following administration of zoliflodacin, and a Medication Guide. Additionally, postmarketing requirements (PMRs) for a descriptive pregnancy safety study to evaluate pregnancy outcomes in females exposed to zoliflodacin during pregnancy, a human sperm study to evaluate the impact of zoliflodacin administration on sperm count and viability, a clinical lactation study, and microbiological surveillance to assess for the development of resistance to zoliflodacin will be issued.

Based on the review of all efficacy and safety data, NDA 219491 for zoliflodacin provides substantial evidence of effectiveness and a favorable benefit-risk profile for the treatment of uncomplicated urogenital gonorrhea in patients 12 years of age and older, weighing at least 35 kilograms.

II. Interdisciplinary Assessment

3. Introduction

Zoliflodacin is a first-in-class oral spiropyrimidinetrione bacterial type II topoisomerase inhibitor. This drug inhibits DNA gyrase and topoisomerase IV, resulting in the inhibition of bacterial DNA replication. The Applicant's proposed indication is treatment of uncomplicated gonorrhea in male and female adult and pediatric patients 12 years of age and older who weigh at least 35 kilograms. Gonorrhea is the second most common bacterial sexually transmitted infection in the United States, with approximately 1.5 million new infections occurring each year.[13] When the phase 3 trial (STI_ Zoli001) was designed, the recommended treatment for gonorrhea was a combination of single doses of IM ceftriaxone and PO azithromycin; thus, the comparator arm included both drugs. In the United States currently, the recommended treatment for uncomplicated gonococcal infections is one IM dose of ceftriaxone. For individuals with cephalosporin allergies, a single dose of IM gentamicin plus a single dose of PO azithromycin is recommended while a single dose of PO cefixime is recommended for those who cannot tolerate injections.

The Division granted zoliflodacin Fast Track designation on April 11, 2019, and QIDP designation for the treatment of uncomplicated gonorrhea on April 16, 2019. This application received a priority review due to its QIDP designation and limited treatment options for uncomplicated gonorrhea.

One phase 3, global, multicenter, open-label, randomized, active-controlled, noninferiority trial (STI_Zoli001) was conducted, in which a single 3 g oral dose of zoliflodacin was compared to a single IM dose of ceftriaxone and single PO dose of azithromycin for the treatment of uncomplicated gonorrhea to assess the safety and effectiveness of zoliflodacin. This trial met the pre-specified NI margin of 12% for the primary endpoint of microbiologic cure at TOC (6±2 days) in patients with uncomplicated urogenital gonorrhea and achieved an NI margin of 10%.

One phase 2, randomized, open-label trial (DMID 14-0014) was conducted to compare a single PO dose of either 2 g or 3 g of zoliflodacin to a single IM dose of ceftriaxone for the treatment of uncomplicated gonorrhea in adult participants and provides confirmatory evidence of efficacy and safety for zoliflodacin.

The safety database was composed of at least 995 participants who received zoliflodacin across 8 clinical trials and studies, including 782 participants who received zoliflodacin at a single oral dose of 3 g proposed for the treatment of uncomplicated urogenital gonorrhea. Several key efficacy and safety review issues were identified during the review of the NDA, as discussed below.

[13] See Footnote 7

3.1. Review Issue List

3.1.1. Key Efficacy Review Issues

3.1.1.1. Imbalance in Rescue Treatment and Post-TOC Infections in Trial STI_Zoli001

A greater number of participants in the zoliflodacin arm, compared to the ceftriaxone-azithromycin arm, were administered concomitant systemic antibacterial drugs. There was also a greater number of infections identified after the TOC Visit in the zoliflodacin arm. Exploratory analyses with alternate treatment failure definitions including these events indicated consistency with findings from the primary efficacy analysis. Refer to section 6.3 for further details.

3.1.1.2. Comparative Efficacy of Zoliflodacin and Comparator in Trial STI_Zoli001

More participants in the zoliflodacin arm had microbiologic treatment failure, compared to the ceftriaxone-azithromycin arm, and required retreatment. However, both treatment arms had microbiological cure rates of >90% and the difference in treatment failure rates is within the clinically acceptable noninferiority margin of 10%. Given the unmet need for additional treatments for gonorrhea, including drug-resistant strains, the use of potentially two effective drugs in the comparator arm, the overall high efficacy in both treatment arms, and meeting a 10% NI margin (narrower than the pre-specified NI margin (12%)), the imbalance in failure rates disfavoring zoliflodacin was considered to be clinically acceptable. Refer to section 6.3 for further details.

3.1.2. Key Safety Review Issues

3.1.2.1. Male Reproductive Toxicity

Based on data from animal toxicity and fertility studies, zoliflodacin may cause testicular toxicity and impair male fertility. Testicular toxicity in these males may adversely affect sperm and consequently induce a potential risk of embryo-fetal toxicity related to males with female partners of reproductive potential (see 3.1.2.3). Minimal to mild decreased spermatogenesis and minimal to moderate testicular degeneration or exfoliation of germinal epithelial cells/immature sperm were observed in rats and dogs administered zoliflodacin for 2 days and up to 4 weeks at doses approximately 3-to 10-times the exposures of the maximum recommended human dose (MRHD). In rats, these findings were associated with a fully reversible decrease in male fertility; however, the underlying testicular histopathology was only partially reversible after 2 to 3 months. Because of the implications for treatment with zoliflodacin in men and women of reproductive potential, two Warnings were added to the PI and will be discussed further in sections 7.7.1, 13.1.5.4 and 23.

3.1.2.2. Embryo-Fetal Toxicity

Based on data from animal reproduction studies, zoliflodacin may cause fetal malformations (exencephaly) or increased embryo-fetal loss when administered to a pregnant female at clinically relevant doses. Fetal exencephaly was observed in fetuses of gravid mice administered zoliflodacin during organogenesis at animal exposures approximately 1.6-times the clinical exposures at the MRHD. Zoliflodacin administered orally to gravid mice and rats during organogenesis was associated with increased embryo-fetal loss at and above exposures of 1.6-fold and 6-fold the MRHD, respectively. Because of the implications for treatment with zoliflodacin in women of reproductive potential, a Warning was added to the PI and will be discussed further in sections 7.7.2, 13.1.5.1 and 23.

3.1.2.3. Potential Risk Related to Males with Female Partners of Reproductive Potential

Increase in fetal loss was reported in rat studies where untreated females were mated with males treated with zoliflodacin for 4 weeks prior to mating. This effect was no longer present after a 4-week recovery period. The review team evaluated the available nonclinical safety data and proposed edits to the Applicant's draft labeling to communicate a potential developmental toxicity risk for males with female partners of reproductive potential, and to recommend contraceptive use for 3 months - the duration of the human sperm cycle - following treatment with zoliflodacin. Refer to sections 7.7.3 and 23 for further details.

3.1.2.4. Additional Analysis of Neutropenia

Low neutrophil cell counts were reported as adverse events in more than 10% of trial participants in both treatment arms. Further analysis of reported laboratory data confirmed this finding. The majority of participants who developed neutropenia were Black or African American and were from South Africa. There was no difference between treatment arms in occurrence of neutropenia and there was no correlation identified between neutropenia and HIV status. Please refer to section 7.6.1.6.1 for further details.

3.2. Approach to the Clinical Review

Evidence of efficacy and safety for zoliflodacin was primarily evaluated in the pivotal clinical trial (STI_Zoli001) in male and female adult and pediatric patients 12 years of age and older with uncomplicated gonococcal infection. Confirmatory evidence was obtained from a phase 2 clinical trial in adult patients (DMID 14-0014). Because of differences in trial design between the two trials, safety data were evaluated from each clinical trial separately. Safety from phase 1 studies was also evaluated. The clinical studies submitted to NDA 219491 were adequate to evaluate the safety and efficacy of zoliflodacin and are summarized in Table 3 below.

3.3. Approach To Establishing Substantial Evidence of Effectiveness

1. Indication granted:

   Treatment of uncomplicated urogenital gonorrhea due to *Neisseria gonorrhoeae* in adults and pediatric patients 12 years of age and older, weighing at least 35 kg

2. SEE was established with

   a. Adequate and well-controlled clinical investigation(s):

      i. ☐ Two or more adequate and well-controlled clinical investigations, **OR**
      ii. ☐ One adequate and well-controlled clinical investigation with highly persuasive results that is considered to be the scientific equivalent of two clinical investigations

      OR

   b. ☒ One adequate and well-controlled clinical investigation and confirmatory evidence[14,15,16]

      OR

   c. ☐ Evidence that supported SEE from a prior approval (e.g., 505(b)(2) application relying only on a previous determination of effectiveness; extrapolation; over-the-counter switch)

3. Complete response, if applicable

   a. ☒ SEE was established
   b. ☐ SEE was not established

[14] FDA draft guidance for industry *Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products* (December 2019). When final, this guidance will represent the FDA's current thinking on this topic. For the most recent version of a guidance, check the FDA guidance web page at https://www.fda.gov/regulatory-information/search-fda-guidance-documents.

[15] FDA guidance for industry *Providing Clinical Evidence of Effectiveness for Human Drug and Biological Products* (May 1998).

[16] FDA draft guidance for industry *Demonstrating Substantial Evidence of Effectiveness With One Adequate and Well-Controlled Clinical Investigation and Confirmatory Evidence* (September 2023).

Table 3. Clinical Trials Submitted in Support of Efficacy and Safety Determinations[1] for Zoliflodacin

Trial Identifier	Trial Population	Trial Design	Regimen (Number Treated), Duration	Primary and Key Secondary Endpoints	No. of Participants Planned; Actual Randomized	No. of Centers and Countries
STI_Zoli001	≥12-year-old male and female participants with uncomplicated gonorrhea	phase 3, R, 2:1 ratio, OL, MC, active control, parallel group, noninferiority	Single PO dose of zoliflodacin 3 g; or single PO dose of azithromycin 1 g plus single IM dose of ceftriaxone 500 mg Single dose trial with a 30-day follow-up Total: N=927 Zoliflodacin 3 g: N=619 Ceftriaxone + Azithromycin: N=308	**Primary:** • Microbiological cure as determined by culture at urethral or cervical sites at TOC (Day 6±2). **Secondary:** • Incidence, severity, causality, and seriousness of treatment-emergent adverse events and the evaluation of changes from Baseline in safety laboratory test results and physical examinations. • Proportion of participants with microbiological cure as determined by culture at pharyngeal and rectal sites at TOC (Day 6±2). • Proportion of male participants with clinical cure at TOC (Day 6±2). • Proportion of female and male participants with microbiological cure as determined by culture at cervical or urethral site at TOC (Day 6±2). • Antimicrobial susceptibility testing (AST) profile of gonococcal strains isolated at Baseline and at TOC (Day 6±2).	Planned: 928 Actual: 930	Centers: 16 Countries: 5

Trial Identifier	Trial Population	Trial Design	Regimen (Number Treated), Duration	Primary and Key Secondary Endpoints	No. of Participants Planned; Actual Randomized	No. of Centers and Countries
DMID 14-0014	≥18- to 55-year-old male and female participants with uncomplicated gonorrhea	phase 2, R, 7:7:4 ratio, OL, MC, active control, parallel group	Single PO dose of zoliflodacin 2 g or 3 g; or single IM dose of ceftriaxone 500 mg Single dose trial with a 30-day follow-up Total: N=179 Zoliflodacin 2 g: N=72 Zoliflodacin 3 g: N=67 Ceftriaxone: N=40	**Primary Efficacy:** • The proportion of participants with microbiological cure at urethral or cervical sites in each trial arm at Visit 2 (Day 6±2) after study drug administration. **Primary Safety:** • The primary safety endpoint was the proportion of participants reporting AEs and SAEs considered product related following dose of study product through Visit 3 (Day 31±2). **Secondary Efficacy:** • The proportion of participants with microbiological cure at rectal sites in each trial arm at Visit 2 after study drug administration. • The proportion of participants with microbiological cure at pharyngeal sites in each trial arm at Visit 2 after study drug administration. • The proportion of participants with no detectable *N. gonorrhoeae* nucleic acid in urethral, cervical, rectum, and pharynx specimens in each trial arm at Baseline and Visit 2. • The proportion of participants with clinical cure in each trial arm at Visit 2. • The in vitro MICs against zoliflodacin and ceftriaxone of gonococcal isolates from cultures taken at Baseline and Visit 2.	Actual: 180	Centers: 5 Countries: 1

Source: Clinical Study Report and adsl.xpt
[1] Includes all submitted clinical trials, even if not reviewed in-depth, except for phase 1 and pharmacokinetic studies.
Abbreviations: AE, adverse event; IM, intramuscular; MC, multicenter; MIC, minimal Inhibitory concentration; N, number of participants; OL, open-label; PO, by mouth; R, randomized; SAE, serious adverse event; TOC, test-of-cure

4. Patient Experience Data

Table 4. Patient Experience Data Submitted or Considered

Data Submitted in the Application		
Check if Submitted	**Type of Data**	**Section Where Discussed, if Applicable**
Clinical Outcome Assessment Data Submitted in the Application		
☐	Patient-reported outcome	Section 6
☐	Observer-reported outcome	
☒	Clinician-reported outcome	
☐	Performance outcome	
Other Patient Experience Data Submitted in the Application		
☐	Patient-focused drug development meeting summary	
☐	Qualitative studies (e.g., individual patient/caregiver interviews, focus group interviews, expert interviews, Delphi Panel)	
☐	Observational survey studies	
☐	Natural history studies	
☐	Patient preference studies	
☐	Other: (please specify)	
☐	If no patient experience data were submitted by Applicant, indicate here.	
Data Considered in the Assessment (But Not Submitted by Applicant)		
Check if Considered	**Type of Data**	**Section Where Discussed, if Applicable**
☐	Perspectives shared at patient stakeholder meeting	
☐	Patient-focused drug development meeting summary report	
☐	Other stakeholder meeting summary report	
☐	Observational survey studies	
☐	Other: (please specify)	

5. Pharmacologic Activity, Pharmacokinetics, and Clinical Pharmacology

5.1. Nonclinical Assessment of Potential Effectiveness

Nonclinical Zoliflodacin PK-PD Information

The Applicant examined the nonclinical pharmacokinetic-pharmacodynamic (PK-PD) of zoliflodacin in a dynamic in vitro hollow fiber infection model (HFIM), with *N. gonorrhoeae.* The HFIM studies evaluated zoliflodacin's activity against *N. gonorrhoeae* reference strains WHO-F and WHO-X. Strain WHO-F is susceptible to all relevant antimicrobials with zoliflodacin agar and microbroth minimum inhibitory concentrations (MICs) of 0.06 mg/L and 0.12 mg/L, respectively. Strain WHO-X is extensively drug-resistant, including resistance to ceftriaxone with zoliflodacin agar and microbroth MICs of 0.12 mg/L and 0.25 mg/L,

respectively. Zoliflodacin's activity was evaluated in HFIM studies using humanized zoliflodacin exposures, i.e., simulated plasma $t_{1/2}$ and free (non-protein-bound fraction) drug concentration-time profiles for zoliflodacin reported in humans. HFIM studies included both dose-ranging (both strains) and dose-fractionation studies (with the WHO-X strain) with a duration of up to 7 days. The findings from these studies are as follows:

1. The dose ranging study (single doses of 0.5, 1, 2, 3, 4, 6, and 8 g of zoliflodacin) demonstrated that single doses of >1 g and >2 g were sufficient to prevent regrowth of strains WHO-F and WHO-X, respectively, within 7 days.
2. The dose fractionation study (total daily doses [TDD] of 1, 2, 3, and 4 g of zoliflodacin divided into once, twice, and three-times-a-day dosing schedules) demonstrated that both single and divided doses of a TDD of >2 g were sufficient to prevent regrowth of WHO-X strain for 7 days, confirming that the activity of zoliflodacin was more concentration-dependent than time-dependent.
3. The ratio of zoliflodacin free-drug (*f*) area under the time-concentration curve from zero to infinity ($AUC_{0-\infty}$) relative to the minimal inhibitory concentration (MIC) was best correlated with bacterial burden reduction and the suppression of emergence of resistance.
4. An $fAUC_{0-\infty}$/MIC ratio of 70.6 was associated with the suppression of emergence of resistance for N. gonorrhoeae (see Section 14.1.1 for more details).

The Applicant utilized this nonclinical PK-PD target information along with probability of target attainment (PTA) analyses to inform the proposed dose and administration with respect to food instruction for the to-be-marketed product.

Because *N. gonorrhoeae* is human-specific and grows poorly in animals, the Applicant did not submit any PK-PD information from *N. gonorrhoeae* in vivo infection model studies.

5.2. Clinical Pharmacology/Pharmacokinetics

Clinical pharmacology properties of zoliflodacin are summarized in Table 5.

Table 5. Summary of Clinical Pharmacology and Pharmacokinetics

Characteristic	Drug Information
	Pharmacologic Activity
Established pharmacologic class (EPC)	Zoliflodacin is a spiropyrimidinetrione bacterial type II topoisomerase inhibitor.
Mechanism of action	Zoliflodacin inhibits bacterial type II topoisomerase enzymes (DNA gyrase and topoisomerase IV), which are required for bacterial DNA synthesis.
Active moieties	Zoliflodacin is the active moiety.
QT prolongation	A concentration-dependent increase in the QT corrected for heart rate (QTc) interval was observed in the thorough QT (TQT) study. However, based on the observed relationship between zoliflodacin concentrations and QTc interval, clinically significant QTc interval prolongation is not expected at the maximum recommended single dose of 3 g zoliflodacin administered with or without food to patients weighing 35 kg or more. This assessment is based on the totality of data review that excludes a clinically significant QTc prolongation for a single dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more. The following are key review findings; see the memorandum dated Jun 26, 2025, from the Interdisciplinary Review Team for Cardiac Safety Studies for additional details: • The predicted mean (CI) QTc for the worst-case concentration scenario is 7.8 (5.1 to 10.6) msec and 11.2 (7.3 to 15.1) msec at the geometric mean C_{max} and the 95th percentile C_{max}, respectively. While these predictions are based on extrapolation, we consider the extrapolation to be reasonable in this case as the mechanism of QTc prolongation is likely to be hERG-mediated and thus the concentration-QTc (C-QTc) is expected to be linear. • The worst-case concentration scenario is the simulated C_{max} for female patients weighing $\geq$35 kg to <50 kg inadvertently taking zoliflodacin 3 g in fed conditions. • No clinically significant QTc prolongation was observed in the food effect study, which included 24-hr Holter recordings and evaluated doses up to 4 g under fed conditions, covering the C_{max} of the worst-case scenario. • No clinically concerning cardiovascular adverse events were reported in the phase 3 trial. TQT study data summary: The effect of zoliflodacin was evaluated in a phase 1, randomized, double-blind, four-period crossover, placebo- and positive-controlled study conducted in healthy male and female participants. The corrected electrocardiogram interval from onset of QRS complex to end of T-wave using Fridericia's formula (QTcF) effect of zoliflodacin or moxifloxacin (positive control) was studied following single doses of 2 g or 4 g zoliflodacin or 400 mg PO moxifloxacin, administered under fasted conditions. Concentration-QTc analysis of data from the TQT study indicated concentration-dependent QTc prolongation. The upper bound of the 95% one-sided CI of the

Characteristic	Drug Information
	estimated mean placebo-adjusted change of QTcF from Baseline (ΔΔQTcF) was predicted to exclude 10 msec at a total-drug plasma concentration of approximately 42.1 µg/mL and lower.
	General Information
Bioanalysis	Validated HPLC-MS/MS methods were used to determine the concentrations of zoliflodacin in human plasma and urine (as applicable to individual studies). The bioanalytical methods validation and performance met the criteria recommended in the ICH M10 Bioanalytical Method Validation and Study Sample Analysis guidance document.[17]
Healthy participants versus participants with uncomplicated urogenital gonorrhea	Participants (n=24) with uncomplicated confirmed or suspected urogenital gonorrhea enrolled in the PK substudy of the phase 3 trial exhibited higher exposure (approximately 138% and 131% higher $AUC_{0-\infty}$ and C_{max}, respectively) than healthy participants enrolled in the phase 1 studies. The observed difference in exposure between the phase 3 participants with uncomplicated urogenital gonorrhea and phase 1 healthy participants may be explained by the lower overall body weight of the phase 3 substudy participants with uncomplicated urogenital gonorrhea compared to the phase 1 healthy participants, as body weight was found to be a significant covariate with respect to exposure in the population PK analysis.
Drug exposure following a single dose of 3 g zoliflodacin	Parameter — Geometric Mean (%GCV)* $AUC_{0-\infty}$ (µg•hr/mL) — 353 (24.1%) C_{max} (µg/mL) — 28.5 (21.6%) *Predicted geometric mean (%GCV) estimates based on post hoc parameters from 24 participants enrolled in the phase 3 trial (weight range 48.2 to 112.3 kg) who received zoliflodacin 3 g after a low/moderate fat meal.
Range of effective dose(s) or exposure	The pivotal phase 3 trial, STI_Zoli001, evaluated a 3 g single dose of zoliflodacin administered under fasted or fed conditions. The supportive phase 2 trial, DMID 14-0014, evaluated a 2 or 3 g single dose of zoliflodacin administered under fasted conditions. In both studies, the microbiological cure rate at test of cure for the urogenital site was >90% for the zoliflodacin arms at all doses evaluated in the micro-ITT population.
Maximally tolerated dose (MTD) or exposure	An MTD was not determined. The highest evaluated single dose in humans was 4 g under fed and fasted conditions.
Dose proportionality	Zoliflodacin, as single doses, generally displayed dose-proportional increases in exposure up to 800 mg (0.27 times the recommended dosage). Increases above 800 mg led to slightly less than dose-proportional increases in exposure up to 4 g (1.3 times the recommended dosage).
Accumulation	Not applicable for the recommended single-dose administration.
Time to achieve steady-state	Not applicable for the recommended single-dose administration.
Bridge between to-be-marketed and clinical trial/study formulations	The to-be-marketed drug product, zoliflodacin oral granules for suspension, was shown to have comparable zoliflodacin bioavailability (i.e., met the bioequivalent criteria under fed and fasted conditions) with the formulation used in the phase 3 clinical trials. The to-be-marketed drug product is manufactured at a different site with a different process than that used for the manufacturing of the phase 3 clinical trial formulation.

[17] ICH guidance for industry M10 Bioanalytical Method Validation and Study Sample Analysis (November 2022), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/m10-bioanalytical-method-validation-and-study-sample-analysis.

Characteristic	Drug Information
	Absorption
Bioavailability	Absolute bioavailability was not assessed in clinical trials.
T_{max}	The median time of maximal concentration was observed at 2.5 hours under fasted conditions and 4 hours under fed conditions.
Food effect (fed/fasted) Geometric least square mean and 90% CI	Parameter / Fed GM (%GCV) / Fasted GM (%GCV) / $Ratio_{Fed/Fasted}$ (%) (90% CI) $AUC_{0-\infty}$ (µg•h/mL) / 241 (27.8) / 157 (28.8) / 152.901 (145.20, 161.01) C_{max} (µg/mL) / 26.700 (25.4) / 17.9 (29.4) / 149.182 (138.48, 161.71) Parameter / Fed / Fasted T_{max} (h)* / 4.0 (3.0 to 5.5) / 2.5 (1.0 to 4.0) *Median (min to max) At the 3 g dose, both C_{max} and AUC estimate was approximately 1.5-fold when given with a moderate fat meal versus fasted condition. Moderate fat meal contained approximately 400 to 500 kcal with 51%, 10%, and 39% calories from carbohydrates, protein, and fat, respectively.
	Distribution
Apparent volume of distribution	The geometric mean (%GCV) is 177 L (26.6) when administered under fasted conditions and 98.7 L (24.1) under fed conditions.
Plasma protein binding	Plasma protein binding (PPB) was estimated to be approximately 83% (range 82.8% to 83.1%) in in vitro assessments. PPB estimates were independent of concentrations.
Drug as substrate of transporters	Zoliflodacin is a substrate of drug transporters P-gp and BCRP.

Characteristic	Drug Information
	Elimination
Mass balance results	Following a single 3 g radiolabeled (~500 µCi) zoliflodacin dose, the mean cumulative radioactivity recovery was 98% over the 192-hour collection period. Cumulative radiolabeled excretion data indicate that elimination is primarily nonrenal, with the majority of the radioactive dose recovered in the feces. Ratios of blood to plasma total radioactivity were approximately 0.7, and zoliflodacin was the most abundant circulating component (57.5% of total radioactivity by $AUC_{0-\infty}$) in plasma.
Apparent clearance	The geometric mean (%GCV) is 19.1 L/h (28.8) when administered under fasted conditions and 12.5 L/h (27.8) under fed conditions.
Half-life	Zoliflodacin demonstrated a monophasic pharmacokinetic profile. Based on the terminal/elimination phase, the estimated geometric mean (%GCV) half-life is 6.4 hrs (20.4) when administered under fasted conditions and 5.5 hrs (14.0) under fed conditions.
Metabolic pathway(s)	The major clearance mechanism for zoliflodacin is metabolism, as approximately 5% of unmetabolized parent drug was observed in the urine and feces in a radiolabeled mass balance study. Zoliflodacin is metabolized by CYP- and non-CYP-mediated pathways. CYP-mediated metabolism is predominantly via CYP 3A4/5 enzymes, with lesser contributions from CYP1A2, CYP2C9, CYP2C8, and CYP2C19.
Primary excretion pathways (% dose recovered in 192 hours post dose)	Following a single 3 g radiolabeled (~500 µCi) zoliflodacin dose, 18.2% of the dose was recovered in urine, 2.5% of which was recovered as unchanged zoliflodacin; 79.6% of the dose was recovered in feces, 1.5% of which was recovered as unchanged zoliflodacin.
	Intrinsic Factors and Specific Populations
Body weight	The available information suggests no dosage adjustments are required in patients weighing 35 kg or greater. A population PK analysis based on the PK data from phase 1 studies and the phase 3 trial show that weight is a significant covariate of zoliflodacin pharmacokinetics in participants with an inverse relationship between body weight and zoliflodacin exposure. The weights of participants in the population PK database ranged from 48.2 to 112 kg. No PK data were available in participants <48.2 kg. However, participants enrolled in the phase 3 trial receiving a single 3 g dose of zoliflodacin weighed between 36.5 to 138.6 kg.
Age	Age (19 to 55 years old) was not a significant predictor of the variability in pharmacokinetics of zoliflodacin. The available information suggests that after body weight was considered, no dosage adjustments are required in adolescent patients aged 12 to <18 years of age or elderly patients aged >65 years of age.
Renal impairment	Zoliflodacin is eliminated primarily by a nonrenal route. Less than 25% of the parent drug and metabolites are detected in urine in a mass balance study of radiolabeled zoliflodacin. Additionally, NUZOLVENCE is a single-dose drug product, therefore no dosage adjustments are recommended for patients with renal impairment.
Hepatic impairment	Zoliflodacin clearance is mainly via metabolism by CYP- and non-CYP-mediated pathways. Based on the totality of data and considering NUZOLVENCE is a single-dose drug product, no dosage adjustments are recommended for patients with hepatic impairment. See Section 8.1 for additional details.

Characteristic	Drug Information
	Drug Interaction Liability (Drug as Perpetrator)
Inhibition/induction of metabolism	Zoliflodacin demonstrated inhibition potential of CYPs 2C8, 2C9, and 2C19 in vitro. However, based on mechanistic static model predictions, zoliflodacin has a low risk of a clinically relevant drug interaction as a direct inhibitor of CYPs 2C8, 2C9, and 2C19 in vivo. No reversible or time-dependent inhibition of CYP3A4/5 was observed in the in vitro drug-drug interaction (DDI) studies at <500µM (50× maximum clinical concentration). The anticipated maximum zoliflodacin concentration in the GI tract is estimated to be 1538 µM after considering maximum reported solubility. However, zoliflodacin's potential to inhibit intestinal CYP3A4/5 is considered to be transient, if any, due to rapid absorption and administration as a single dose. See Section 8.2 for additional details. Zoliflodacin demonstrated induction of CYP1A2 in vitro. However, the drug interaction potential is unlikely to be clinically significant as NUZOLVENCE is a single-dose drug product.
Inhibition/induction of transporter systems	Zoliflodacin demonstrated inhibition of P-gp, BCRP, OATP1 B1/B3, OAT1/3, MATE1, and MATE2K drug transporters in vitro. However, as NUZOLVENCE is a single-dose drug product (with a short half-life of 5.5 hrs to 6.4 hrs), the overall DDI risk is low.

Source: FDA reviewer's summary and assessment
AUC, area under the concentration-time curve; $AUC_{0-\infty}$/MIC, area under the concentration-time curve estimated to infinity relative to MIC; BCRP, breast cancer resistance protein; CI, confidence interval; C_{max}, maximum plasma concentration; C-QTc, concentration-QTc; CYP, cytochrome P-450; GCV, geometric coefficient of variation; GM, geometric mean; hERG, human ether-à-go-go-related gene; HPLC-MS/MS, high pressure liquid chromatography with tandem mass spectrometry; ICH, International Council for Harmonisation; MATE, multidrug and toxin extrusion transporter; max, maximum; MIC, minimum inhibitory concentration; min, minimum; mITT, microbiologic intent-to-treat; OAT, organic anion transporter; OATP, organic anion transporting polypeptide; P-gp, P-glycoprotein; PK, pharmacokinetic; PO, by mouth

6. Efficacy (Evaluation of Benefit)

6.1. Assessment of Dose and Potential Effectiveness

The Applicant's proposed zoliflodacin dosage is a single 3 g dose of zoliflodacin for oral suspension with administration instructions based on patient weight:

For patients weighing >35 kg and <50 kg: Administer under fasted conditions, i.e., take on an empty stomach, 1 hour before or 2 hours after food.

For patients weighing >50 kg: Administer under fed conditions, i.e., take with food.

The proposed dosage was supported by safety and efficacy data from the pivotal phase 3 trial (STI_Zoli001). Additionally, the proposed dosage was supported by the safety and efficacy data from the phase 2 trial (DMID 14-0014) as well as the PTA analyses based on the nonclinical PK-PD target.

Phase 3 Trial, STI_Zoli001

The pivotal phase 3 trial (STI_Zoli001) evaluated a single 3 g dose of zoliflodacin for oral suspension administered under either fasted (n=64) or fed (n=555) conditions and compared it with dual therapy with ceftriaxone and azithromycin for the treatment of uncomplicated gonorrhea. A single 3 g dose under fed or fasted conditions was selected primarily based on the phase 2 trial tolerability and efficacy data.

In Trial STI_Zoli001, zoliflodacin administration with respect to food was not governed by participant weight as proposed in the labeling. Instead, the initial version of the trial protocol allowed administration of zoliflodacin without respect to food. A subsequent protocol revision specified dosing under fed conditions. All participants with weight <50 kg were dosed under fed conditions. The primary endpoint in STI_Zoli001 was microbiological cure, defined as a negative culture for *N. gonorrhoeae* at the urethral or cervical site at TOC (Day 6±2 days) among participants included in the micro-ITT urogenital population. The primary efficacy results for microbiological cure at the urethral or cervical site at TOC demonstrated noninferiority of zoliflodacin compared to IM ceftriaxone and PO azithromycin using a 12% noninferiority margin. In general, the zoliflodacin dosage was well tolerated. For more details on the efficacy and safety data see Sections 6.2.2 and Section 7, respectively.

Phase 2 Trial, DMID 14-0014

Trial DMID 14-0014 evaluated two zoliflodacin dosages, either a single 2 g or 3 g dose of zoliflodacin, administered without food, in participants with uncomplicated urogenital gonorrhea. The primary endpoint for DMID 14-0014 was microbiological cure at urethral or cervical sites at TOC (Visit 2 at Day 6±2) in the micro-ITT population. At TOC in the micro-ITT population, the microbiological cure rate at the urogenital location exceeded 96% for both doses evaluated. Additionally, both the doses were well tolerated. For more details on the efficacy and safety data from the phase 2 trial see Section 6.2.3 and Section 7, respectively.

Nonclinical PK-PD Data and PTA

Through Monte Carlo simulation and PTA, the Applicant integrated the nonclinical zoliflodacin PK-PD information (see Section 5.1) with clinical zoliflodacin PK information to support selection of the proposed dosage (a single 3 g dose of zoliflodacin granules for oral suspension with administration instructions based on patient weight).

A PK-PD target (fAUC$_{0-\infty}$/MIC =70.6) was associated with the suppression of emergence of resistance for *N. gonorrhoeae*, identified in the in vitro HFIM study with *N. gonorrhoeae* strains WHO-F and WHO-X with zoliflodacin agar and microbroth MICs of 0.12 mg/L and 0.25 mg/L, respectively. This PK-PD target was utilized to support the proposed dosage for marketing approval including the administration instructions based on patient weight, as discussed further below.

Evaluation of Administration Instructions Based on Patient Weight

Patient body weight was found to be a significant covariate in the population PK analysis and predicted exposure as both maximum plasma concentration (C_{max}) and AUC$_{0-\infty}$ had an inverse relationship to body weight. Zoliflodacin bioavailability is significantly increased when taken in the fed state (see Section 8.2 for more details). The Applicant's rationale for the proposed zoliflodacin dosage based on a patient's weight as well as administration instruction in relation to food was guided by two principles:

1. To not exceed a C_{max} which was predicted to be associated with QTc prolongation and,
2. To ensure sufficient exposure to meet a fAUC$_{0-\infty}$/MIC target associated with the suppression of emergence of resistance for N. gonorrhoeae.

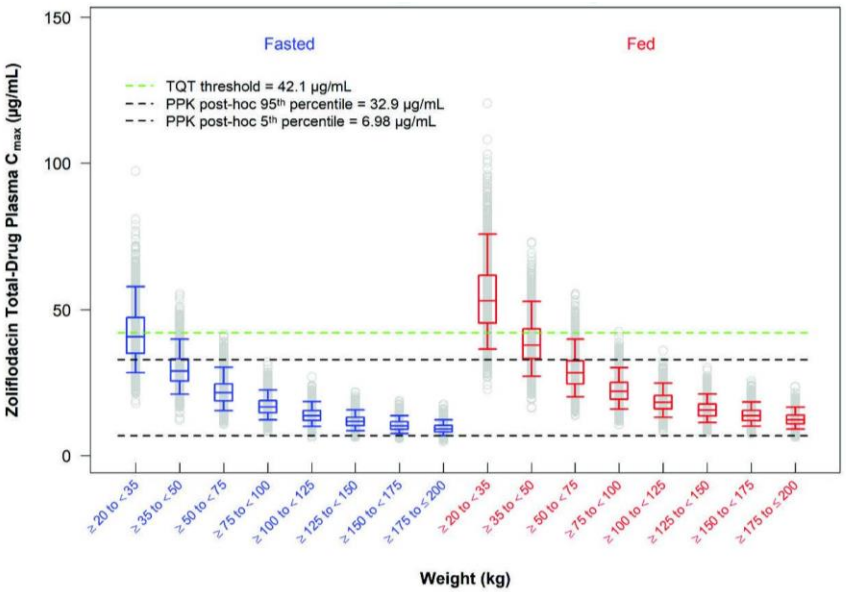
QT Prolongation Risk

The Applicant simulated exposure, both AUC$_{0-\infty}$ and C_{max}, in male and female participants with uncomplicated gonorrhea by body weight group, after administration of a single PO dose of zoliflodacin 3 g under both fasted and fed conditions. The Applicant compared the simulated C_{max} to a concentration threshold (42.1 µg/mL) predicted to be associated with QTc prolongation identified in the TQT study, as shown in Figure 1 below (see Section 14.2.5 for additional information). Based on the predicted mean C_{max} for patients weighing ≥35 kg and <50 kg under fed conditions exceeding the threshold, the Applicant proposed a single 3 g dose of zoliflodacin be administered under fasted conditions for patients weighing ≥35 kg and <50 kg. All other body weight groups evaluated were below the threshold under either fed or fasted conditions when administered a single 3 g dose of zoliflodacin.

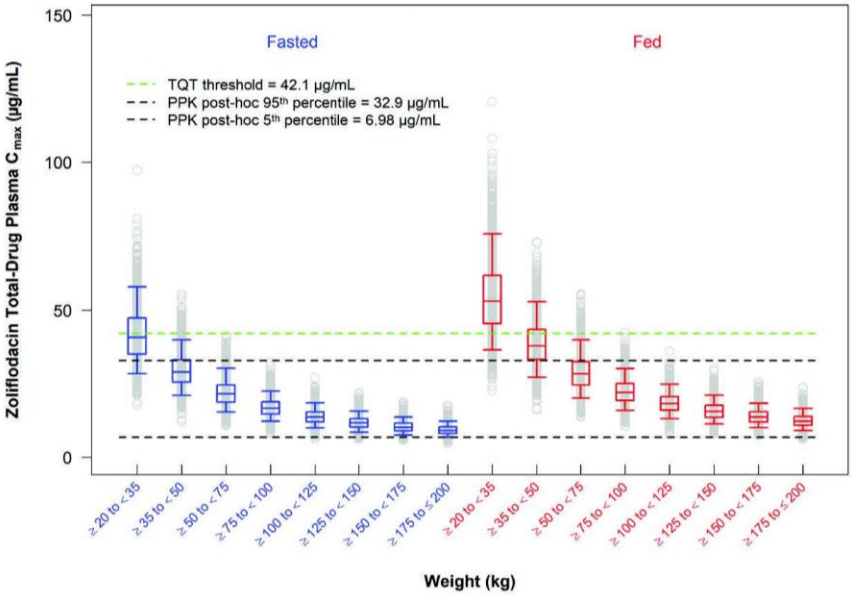
The totality of evidence suggested that clinically significant QTc prolongation is not anticipated for a single dose of 3 g of zoliflodacin when administered with or without food to patients weighing ≥35 kg. Nevertheless, given the concentration-dependent QTc prolongation observed in the TQT study, administration of zoliflodacin under fasted conditions would reduce the likelihood of QTc prolongation in patients weighing ≥35 kg and <50 kg. Therefore, the Applicant's proposal is acceptable to the review team given it is based on a conservative approach utilizing a theoretical QT safety threshold. See Section 14.2.9 for additional details.

Figure 1. Box-and-Whisker Plots of Zoliflodacin Total-Drug Plasma C_{max} in Comparison to the TQT Threshold Among Simulated Fasted or Fed Male (A) and Female (B) Participants With Uncomplicated Gonorrhea After Administration of a Single Oral Dose of Zoliflodacin 3 g

A. Simulated Female Patients by Meal Status by Body Weight Group Assuming Phase 3 Bioavailability



B. Simulated Female Patients by Meal Status by Body Weight Group Assuming Phase 3 Bioavailability



Source: Figure 8 Report Number ICPD 00734-2
Abbreviations: C_{max}, maximum plasma concentration; PPK, population pharmacokinetic; TQT, thorough QT

PTA Analysis

Utilizing this simulated $AUC_{0-\infty}$ data described above, the Applicant evaluated the PTA rates for the selected PK-PD target (i.e., $fAUC_{0-\infty}$/MIC =70.6) by body weight group and by MIC for *N. gonorrhoeae* based on in vitro surveillance and phase 3 data. The Applicant utilized a criteria

of PTA ≥90% for PK-PD target as a threshold for efficacy. When administered a single 3 g dose of zoliflodacin, all body weight groups evaluated under fed conditions, and all body weight groups <175 kg evaluated under fasted conditions were above the PTA threshold, for all MICs ≤0.25 mg/L, i.e., the proposed susceptible breakpoint. These findings support the Applicant's proposal of administering a single 3 g dose of zoliflodacin with food for patients weighing ≥50 kg.

With respect to data supporting the recommended lowest weight cutoff of ≥35 kg, the lowest participant body weights evaluated in the zoliflodacin arms in the pivotal phase 3 trial and supportive phase 2 trial were 36.5 kg and 51.7 kg, respectively. Additionally, the pivotal phase 3 trial enrolled adolescent participants with uncomplicated gonorrhea, and the lowest age enrolled in the zoliflodacin arm was 16 years of age. Therefore, efficacy and safety were extrapolated to support use of zoliflodacin using the proposed dosage down to a weight of 35 kg and down to an age of 12 years by relying on the safe and effective zoliflodacin exposures range identified in the phase 3 trial. See Pharmacometrics and PTA analysis in section 14.5.1 for further details.

6.2. Clinical Studies/Trials Intended To Demonstrate Efficacy

6.2.1. Results of Pooled Analyses

In this NDA submission, one phase 3 clinical trial, STI_Zoli001, provides the primary efficacy data for zoliflodacin, with the phase 2 clinical trial, DMID 14-0014 providing confirmatory evidence. Due to the differences in study designs, a pooled analysis was not conducted.

6.2.2. Trial STI_Zoli001

6.2.2.1. Design, Trial STI_Zoli001

STI_Zoli001 (clinicaltrials.gov identifier NCT03959527) was an international, multicenter, randomized, open-label, noninferiority trial that enrolled participants ≥12 years of age, weighing at least 35 kg, to compare the efficacy and safety of zoliflodacin to dual therapy with ceftriaxone and azithromycin, for the treatment of uncomplicated urogenital gonorrhea. The trial enrolled participants between November 6, 2019 and March 16, 2023 from 16 study centers across five countries. Participants were randomized 2:1 to receive one 3 g PO dose of zoliflodacin or combination therapy with a single IM 500 mg dose of ceftriaxone and single 1 g PO dose of azithromycin.

Participants were enrolled for 30±2 days, with trial visits to assess safety, efficacy and microbiological endpoints occurring at Baseline (Day 1), TOC (Day 6±2) and end-of-study (EOS) (Day 30±2). The primary efficacy endpoint was microbiological cure as determined by culture at urethral or cervical sites at TOC.

The trial utilized a noninferiority design and was designed in accordance with the Agency's current uncomplicated gonorrhea guidance (August 2015)."[18] In view of the significant unmet public health need for antibacterial drugs to treat *N. gonorrhoeae*, including resistant strains, FDA agreed to accept a prespecified noninferiority margin of 12%.

In total, 930 participants were enrolled, with 744 participants (506 and 238 for zoliflodacin and ceftriaxone-azithromycin, respectively) included in the micro-ITT (urogenital) population. Of note, due to a serious breach of Good Clinical Practice at a center in South Africa (710-005), all data obtained from the 31 participants enrolled at this site were excluded from the analyses.

In addition to the primary endpoint of the microbiological cure at TOC, secondary endpoints included the following:

1. Incidence, severity, causality and seriousness of treatment-emergent adverse events (TEAEs) and the evaluation of changes from Baseline in safety laboratory test results and physical examinations.
2. Proportion of participants with microbiological cure as determined by culture at pharyngeal sites at TOC (Day 6±2).
3. Proportion of participants with microbiological cure as determined by culture at rectal sites at TOC (Day 6±2).
4. Proportion of male participants with clinical cure at TOC (Day 6±2).
5. Proportion of female and male participants with microbiological cure as determined by culture at cervical or urethral sites at TOC (Day 6±2).
6. Proportion of participants with microbiological cure as determined by culture at urethral or cervical sites at the TOC Visit and for whom the baseline antimicrobial susceptibility test (AST) profile indicated pre-existing resistance to antibiotics commonly used for NG treatment (including to ceftriaxone, to azithromycin alone and to both).
7. AST profile of gonococcal strains isolated at Baseline and at TOC (Day 6±2).
8. Proportion of participants with a negative *N. gonorrhoeae* nucleic acid amplification test (NAAT) from urethral or cervical sites at TOC (Day 6±2).
9. Proportion of participants with a negative *N. gonorrhoeae* NAAT from oropharyngeal sites at TOC (Day 6±2).
10. Proportion of participants with a negative *N. gonorrhoeae* NAAT from rectal sites at TOC (Day 6±2)
11. Plasma PK parameters of zoliflodacin in adult participants (≥18 years old).
12. Plasma PK parameters of zoliflodacin in HIV negative adolescents (≥12 and ≤18 years old).

Of note, there were no strategies implemented to control overall type I error for testing of the secondary endpoints, which included microbiological cure at the pharyngeal and rectal sites.

[18] See Footnote 1

6.2.2.2. Eligibility Criteria, Trial STI_Zoli001

Participants were eligible for enrollment into Trial STI_Zoli001 if they fulfilled all the following inclusion criteria:

1. ≥12 years old (if enrollment of minors was in agreement with local regulations and ethics guidance).
2. ≥35 kilograms.
3. Signs and symptoms consistent with urethral or cervical gonorrhea OR, urethral or cervical uncomplicated gonorrhea as determined by either a positive culture or NAAT or Gram stain or methylene blue/gentian violet stain in the past 14 days prior to screening OR, unprotected sexual contact with an individual confirmed to be infected with NG in the past 14 days prior to screening (confirmation by positive NAAT, Gram stain, methylene blue/gentian violet stain or culture).
4. For females of child-bearing potential, a negative urine pregnancy test at screening.
5. For females of child-bearing potential, use of a highly effective method of contraception at the time of trial drug administration on Day 1 and until at least 28 days after treatment. Females on oral contraceptives also had to use a barrier contraception method during participation in the trial.
6. For males with a female partner of child-bearing age, willingness to delay conception during the trial and for 28 days after treatment.
7. Willingness to comply with trial protocol.
8. For participants in the PK substudy: willingness to undergo HIV testing.
9. Willingness to abstain from sexual intercourse or use condoms for vaginal, anal and oral sex from enrollment until the end-of-trial Visit.
10. Willingness and ability to give written informed consent or be consented by a legal representative, or provide assent and parental consent (for minors, as appropriate).

6.2.2.3. Statistical Analysis Plan, Trial STI_Zoli001

The following analysis populations were included in the trial:

1. Randomized Population: All participants who were randomized.
2. Micro-ITT population: All randomized participants who had a positive *N. gonorrhoeae* culture from the relevant anatomical site (urogenital, rectal, pharyngeal) at Baseline and whose baseline AST result showed susceptibility to ceftriaxone or azithromycin.
3. Modified micro-ITT population: All participants who had a positive *N. gonorrhoeae* culture from the relevant anatomical site at Baseline (regardless of pre-existing resistance).
4. Clinical cure population: Included all participants assigned male at birth who were included in the micro-ITT (urogenital) population and who had at least one sign or symptom of urethral gonorrhea at Baseline.
5. Safety population: All randomized participants who received any part of trial treatment.
6. PK population: Included all participants randomized to the zoliflodacin group who consented to participate in the PK subtrial and from whom at least one valid PK post-treatment sample was obtained.
7. Evaluable population: Defined separately for urogenital, rectal, or pharyngeal body sites; included all randomized participants with a positive *N. gonorrhoeae* culture at Baseline and whose baseline AST result showed no pre-existing resistance to both ceftriaxone and

azithromycin, who did not vomit within 30 minutes of administration of zoliflodacin or azithromycin and who had a *N. gonorrhoeae* culture result at the TOC Visit.

8. Per-protocol (PP) population: Defined separately for urogenital, rectal, or pharyngeal body sites; included all participants in the micro-ITT population who met all inclusion/exclusion criteria, complied with trial treatment, did not vomit within 30 minutes of administration of zoliflodacin or azithromycin, did not receive any systemic antibiotic with known activity against *N. gonorrhoeae* prior to TOC Visit, did not receive any of the prohibited medications, abstained from sexual intercourse or used condoms for vaginal, anal and oral sex prior to TOC and returned to the trial site for the TOC Visit within the specified window (Visit 4, Day 6±2).
9. Clinical-PP population: Included participants assigned male at birth in the PP population with at least one sign or symptom of gonorrhea at Baseline and with an evaluable clinical outcome.

The micro-ITT (urogenital) population was the primary analysis population.

The primary efficacy endpoint was microbiological cure as determined by culture at urethral or cervical sites at TOC (Day 6±2). The noninferiority of zoliflodacin to ceftriaxone-azithromycin was evaluated using a two-sided 95% Newcombe score confidence interval, with a noninferiority margin of 12%.

Secondary efficacy endpoints included the following:

1. Proportion of participants with microbiological cure as determined by culture at pharyngeal sites at TOC.
2. Proportion of participants with microbiological cure as determined by culture at rectal sites at TOC.
3. Proportion of male participants with clinical cure at TOC.
4. Proportion of female and male participants with microbiological cure as determined by culture at cervical or urethral sites at TOC.
5. Proportion of participants with microbiological cure as determined by culture at urethral or cervical sites at the TOC Visit and for whom the baseline antimicrobial susceptibility profile indicated pre-existing resistance to antibiotics commonly used for treatment of *N. gonorrhoeae.*
6. Antimicrobial susceptibility profile of gonococcal strains isolated at Baseline and at TOC.
7. Proportion of participants with a negative NG NAAT from urethral or cervical sites at TOC.
8. Proportion of participants with a negative NG NAAT from oropharyngeal sites at TOC.
9. Proportion of participants with a negative NG NAAT from rectal sites at TOC (Day 6±2).

Secondary endpoints were summarized descriptively. No strategies were implemented to control the overall type I error rate for testing secondary endpoints, including analyses of results at the pharyngeal and rectal sites, and the trial was not statistically powered for these analyses.

6.2.2.4. Results of Analyses, Trial STI_Zoli001

Data from Site 710-005 were removed from all analyses due to good clinical practice (GCP) noncompliance. After excluding Site 710-005, a total of 1011 participants were screened. Among them, 930 participants were randomized in the trial and 927 participants were treated. In this

trial, the micro-ITT population and modified micro-ITT populations were identical for each body site.

Table 6. Participant Disposition, Trial STI_Zoli001

Parameter	Zoliflodacin N=621	Ceftriaxone-Azithromycin N=309
Randomized population	621 (100)	309 (100)
Safety population	619 (99.7)	308 (99.7)
Micro-ITT (urogenital) population	506 (81.5)	238 (77.0)
Micro-ITT rectal population	79 (12.7)	35 (11.3)
Micro-ITT pharyngeal population	53 (8.5)	28 (9.1)
Modified micro-ITT (urogenital) population	506 (81.5)	238 (77.0)
Modified micro-ITT rectal population	79 (12.7)	35 (11.3)
Modified micro-ITT pharyngeal population	53 (8.5)	28 (9.1)
Evaluable urogenital population	475 (76.5)	229 (74.1)
Evaluable rectal population	72 (11.6)	31 (10.0)
Evaluable pharyngeal population	46 (7.4)	23 (7.4)
Per-protocol urogenital population	452 (72.8)	219 (70.9)
Per-protocol rectal population	64 (10.3)	31 (10.0)
Per-protocol pharyngeal population	46 (7.4)	23 (7.4)
Clinical cure population	460 (74.1)	220 (71.2)
Clinical-PP population	410 (66.0)	201 (65.0)
PK population	24 (3.9)	0
Discontinued trial	50 (8.1)	24 (7.8)
Lost to follow-up	31 (5.0)	15 (4.9)
Participant did not meet eligibility criteria	10 (1.6)	5 (1.6)
Withdrawal by participant	1 (0.2)	1 (0.3)
Other	8 (1.3)	3 (1.0)

Source: FDA Analysis; adsl.xpt; excluding participants from site 710-005
Abbreviations: micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; PP, per-protocol

Demographic characteristics are listed in Table 7. The two groups had similar distributions in these characteristics. The majority of participants were male at birth (88%), not Hispanic (97%), and African American (55%). Among all randomized participants, 158 participants were from the United States. The average age of participants from the United States was 35.4 years, with ages ranging from 19 to 67 years. Participants from the United States were Black or African American (53%), White (33%), or of other races (14%).

Table 7. Baseline Demographics and Clinical Characteristics, Randomized Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=621	Ceftriaxone-Azithromycin N=309
Sex assigned at birth, n (%)		
Female	77 (12.4)	38 (12.3)
Male	544 (87.6)	271 (87.7)
Gender, n (%)		
Female	78 (12.6)	40 (12.9)
Male	543 (87.4)	269 (87.1)
Age, years		
Mean (SD)	30.0 (9.56)	29.2 (9.13)
Median	28.0	26.0
Min, max	16.0, 73.0	15.0, 67.0

Characteristic	Zoliflodacin N=621	Ceftriaxone-Azithromycin N=309
Age group (years), n (%)		
<18	12 (1.9)	2 (<1)
≥18 to <65	606 (97.6)	306 (99.0)
≥65	3 (<1)	1 (<1)
Race, n (%)		
Black or African American	349 (56.2)	165 (53.4)
Asian	193 (31.1)	92 (29.8)
White	66 (10.6)	47 (15.2)
American Indian or Alaska Native Asian	8 (1.3)	1 (<1)
Native Hawaiian or Other Pacific Islander	2 (<1)	1 (<1)
Multiple	1 (<1)	2 (<1)
Other	2 (<1)	1 (<1)
Ethnicity, n (%)		
Hispanic	18 (2.9)	13 (4.2)
Not Hispanic	603 (97.1)	296 (95.8)
Baseline height (cm)		
Mean (SD)	172.2 (8.51)	171.7 (8.39)
Median	172.0	171.0
Min, max	150.0, 201.0	148.0, 196.5
Missing	2	2
Baseline weight (kg)		
Mean (SD)	70.4 (15.54)	68.6 (13.58)
Median	67.5	66.8
Min, max	36.5, 138.6	43.4, 119.0
Missing	3	1
BMI (kg/m^2)		
Mean (SD)	23.7 (4.71)	23.3 (4.57)
Median	22.7	22.4
Min, max	13.4, 46.0	15.0, 47.0
Missing	3	2
Region, n (%)		
Asia	181 (29.1)	89 (28.8)
Europe	55 (8.9)	23 (7.4)
South Africa	278 (44.8)	146 (47.2)
United States	107 (17.2)	51 (16.5)
Country of participation, n (%)		
Belgium	6 (1.0)	3 (1.0)
Netherlands	49 (7.9)	20 (6.5)
Thailand	181 (29.1)	89 (28.8)
United States	107 (17.2)	51 (16.5)
South Africa	278 (44.8)	146 (47.2)
History of sexually transmitted infection(s), n (%)		
Yes	343 (55.2)	179 (57.9)
No	278 (44.8)	130 (42.1)
HIV status, n (%)		
Negative	455 (73.3)	234 (75.7)
Positive	134 (21.6)	65 (21.0)
Missing	32 (5.2)	10 (3.2)

Source: FDA Analysis; adsl.xpt; excluding participants from site 710-005
Abbreviations: max, maximum; min, minimum; N, number of participants in treatment arm; n, number of participants with given characteristic; SD, standard deviation

Microbiological Cure at TOC at Urogenital Site (Primary Efficacy Endpoint)

Table 8 displays analysis results for the primary efficacy endpoint of microbiological cure at TOC evaluated in the micro-ITT (urogenital) population. Treatment with zoliflodacin demonstrated noninferiority to the active control of ceftriaxone-azithromycin with a noninferiority margin of 12% in the primary analysis. Noninferiority was also demonstrated using a more conservative margin of 10%, as recommended in the Agency's current uncomplicated gonorrhea guidance.[19]

It was noted that zoliflodacin was statistically inferior to ceftriaxone-azithromycin with the confidence interval for the treatment difference completely below zero. The comparative efficacy of zoliflodacin is discussed in more detail in Section 6.3.2.

Most failures in the primary analysis were due to nonassessable outcomes. However, there were 15 confirmed positive culture results in the zoliflodacin arm at TOC but no confirmed positive cultures in the ceftriaxone-azithromycin arm. To evaluate the impact of nonassessable outcomes, the reviewer conducted the following additional sensitivity analyses, with details summarized in Table 8. First, the primary analysis was conducted including out-of-window TOC culture results (out-of-window TOC results were collected from Day 9 to Day 20). Results were similar to the original primary analysis, demonstrating noninferiority of zoliflodacin to the active control.

Second, a worst-case scenario analysis was conducted. All nonassessable outcomes in the zoliflodacin arm were considered as failures, while all nonassessable outcomes in the ceftriaxone-azithromycin arm were considered as cures. In this analysis, zoliflodacin still demonstrated noninferiority to the active control with a margin of 12%.

Third, a sensitivity analysis was conducted excluding all participants with nonassessable outcomes. The noninferiority of zoliflodacin to ceftriaxone-azithromycin was still demonstrated.

Table 8. Microbiological Cure Rate at TOC at Urogenital Site, Micro-ITT (Urogenital) Population, Trial STI_Zoli001

Primary Analysis	**Zoliflodacin N=50 n (%)**	**Ceftriaxone-Azithromycin N=23 n (%**	**Difference % (95% CI[a])**
Cure (Negative for NG)	460 (90.9)	229 (96.2)	−5.3% (−8.6%, −1.4%)
Failure	46 (9.1)	9 (3.8)	
Positive for NG	15 (3.0)	0	
Nonassessable	31 (6.1)	9 (3.8)	
Missed TOC Visit	15 (3.0)	6 (2.5)	
Unobtainable specimen	4 (0.8)	1 (0.4)	
TOC out of window[b]	12 (2.4)	2 (0.8)	
Include OOW Results	**Zoliflodacin N=506**	**Ceftriaxone-Azithromycin N=238**	**Difference % (95% CI)**
Cure	471 (93.1)	231 (97.1)	−4.0% (−6.9%, −0.4%)
Worst Case Scenario	**Zoliflodacin N=506**	**Ceftriaxone-Azithromycin N=238**	**Difference % (95% CI)**
Cure	460 (90.9)	238 (100)	−9.1% (−11.9%, −6.4%)

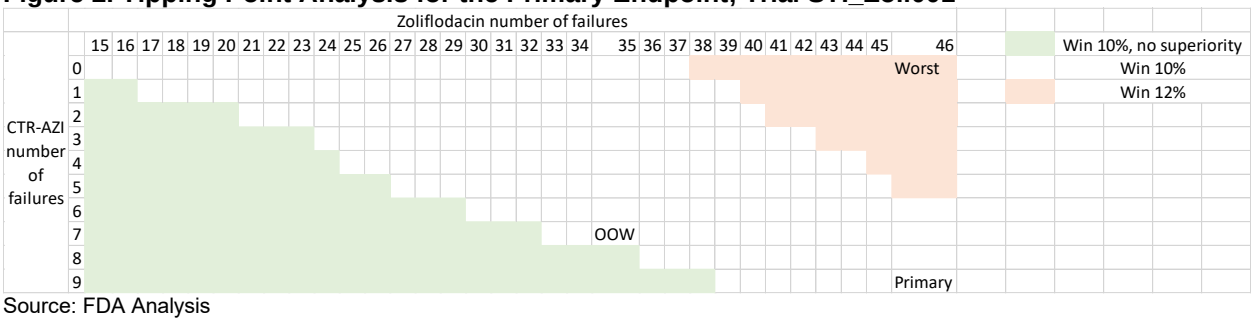
[19] See Footnote 1

Nonassessable Results Removed	Zoliflodacin N=475	Ceftriaxone-Azithromycin N=229	Difference % (95% CI)
Cure	460/475 (96.8)	229/229 (100)	−3.2% (−5.1%, −1.1%)

Source: FDA Analysis; adcure.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
[b] One participant from the zoliflodacin arm whose TOC Visit was out of window had an unobtainable specimen. All 13 other participants whose TOC Visits were out of window had negative culture results at that visit.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; NG, *Neisseria gonorrhoeae*; OOW, out-of-window; TOC, test-of-cure

The reviewer also conducted the following tipping point analysis to explore all possible outcomes of the nonassessable cases. In all cases, zoliflodacin still demonstrated noninferiority to the active control with a margin of 12%. In certain cases, zoliflodacin could demonstrate noninferiority with a more conservative margin of 10%. See details in Figure 2.

Figure 2. Tipping Point Analysis for the Primary Endpoint, Trial STI_Zoli001



Source: FDA Analysis
Green area indicating demonstrating noninferiority with a 10% margin and not showing superiority of ceftriaxone-azithromycin. White area indicating demonstrating noninferiority with a 10% margin but showing superiority of ceftriaxone-azithromycin. Orange area indicating demonstrating noninferiority with a 12% margin but showing superiority of ceftriaxone-azithromycin.
Worst indicates worst-case scenario sensitivity analysis.
Primary indicates primary efficacy analysis.
Abbreviations: CTR-AZI, ceftriaxone-azithromycin; OOW, out-of-window

Table 9 below summarizes the primary endpoint by subgroup. Larger than 11% numerical cure rate differences between the two treatment arms were observed in the Black or African American race subgroup and the South Africa subgroup by region.

Table 9. Microbiological Cure Rate at TOC at Urogenital Site by Subgroup, Micro-ITT (Urogenital) Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=506 n/N (%)	Ceftriaxone-Azithromycin N=238 n/N (%)	Difference % (95% CI[a])
Overall	460/506 (90.9%)	229/238 (96.2%)	−5.3% (−8.6%, −1.4%)
Sex assigned at birth			
Female	48/50 (96.0%)	16/18 (88.9%)	7.1% (−5.3%, 29.0%)
Male	412/456 (90.4%)	213/220 (96.8%)	−6.5% (−9.9%, −2.4%)
Age group (years)			
<18	9/9 (100%)	1/1 (100%)	
≥18 to <65	449/495 (90.7%)	227/236 (96.2%)	−5.5% (−8.9%, −1.5%)
≥65	2/2 (100%)	1/1 (100%)	

Characteristic	Zoliflodacin N=506 n/N (%)	Ceftriaxone-Azithromycin N=238 n/N (%)	Difference % (95% CI[a])
Race			
Black or African American	241/278 (86.7%)	125/128 (97.7%)	−11.0% (−15.7%, −5.4%)
Asian	158/161 (98.1%)	65/67 (97.0%)	1.1% (−3.0%, 8.5%)
White	51/57 (89.5%)	34/38 (89.5%)	0 (−12.4%, 14.7%)
American Indian or Alaska Native	6/6 (100%)	1/1 (100%)	
Native Hawaiian or Other Pacific Islander	1/1 (100%)	1/1 (100%)	
Multiple	1/1 (100%)	2/2 (100%)	
Other	2/2 (100%)	1/1 (100%)	
Ethnicity			
Hispanic	15/15 (100%)	12/13 (92.3%)	7.7% (−13.7%, 33.3%)
Not Hispanic	445/491 (90.6%)	217/225 (96.4)	−5.8% (−9.2%, −1.8%)
Region			
Asia	146/149 (98.0%)	63/64 (98.4%)	−0.5% (−4.4%, 6.4%)
Europe	42/45 (93.3%)	15/17 (88.2%)	5.1% (−8.9%, 28.1%)
South Africa	188/217 (86.6%)	111/113 (98.2%)	−11.6% (−16.9%, −5.7%)
United States	84/95 (88.4%)	40/44 (90.9%)	−2.5% (−12.2%, 10.6%)
History of sexually transmitted infection(s)			
Yes	260/284 (91.5%)	126/130 (96.9%)	−5.4% (−9.6%, −0.1%)
No	200/222 (90.1%)	103/108 (95.4%)	−5.3% (−10.6%, 1.3%)
HIV status			
Negative	345/374 (92.2%)	177/185 (95.7%)	−3.4% (−7.2%, 1.2%)
Positive	95/105 (90.5%)	46/47 (97.9%)	−7.4% (−14.7%, 2.6%)
Missing	20/27 (74.1%)	6/6 (100%)	−25.9% (−44.7%, 15.1%)

Source: FDA Analysis; adsl.xpt; adcure.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with given characteristic; TOC, test-of-cure

Given the higher percentage of missing culture results compared to confirmed positive culture results among the microbiological failures, further analyses were conducted on the culture positivity rates in subgroups by race and region, as shown in Table 10 below. There were no positive cultures in the ceftriaxone-azithromycin arm in the micro-ITT (urogenital) population, so there were no subgroup differences in culture positivity rates in the ceftriaxone-azithromycin arm in this population. Additional analyses were conducted to evaluate the subgroup differences in culture positivity rates in the zoliflodacin arm. Among participants from the zoliflodacin arm, 13/263 (4.9%) Black or African American participants had positive cultures at TOC, compared to 2/223 (0.9%) in non-Black or African American participants (two-sided Fisher's Exact test p-value=0.015). Additionally, 11/207 (5.3%) participants from South Africa had positive cultures at TOC, compared to 4/279 (1.4%) in participants from the rest of the world (two-sided Fisher's Exact test p-value=0.031).

Although the differences in culture positivity rates between Black or African American and non-African American and between South Africa and rest of the world were nominally significant in the zoliflodacin arm, those results should be interpreted with caution given that no multiplicity adjustment was applied in these post hoc analyses. In the comparison between the Black or African American group and the non-Black or African American group, the imbalance was

driven by the low culture positivity rate of the Asian subgroup at TOC (1/161, 0.6%). The White subgroup had a smaller sample size with a culture positivity rate of 1/52 (1.9%). There was a high correlation between race and region, with all 207 participants from South Africa in the zoliflodacin arm being Black or African American. This high correlation introduces additional challenges to the interpretation of results.

Table 10. *Neisseria gonorrhoeae* Culture Positivity Rate at TOC Including OOW Results at Urogenital Site by Subgroup, Micro-ITT (Urogenital) Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=486 n/N (%)	Ceftriaxone-Azithromycin N=231 n/N (%)	Difference % (95% CI[a])
Overall	15/486 (3.1%)	0/231	3.1% (1.1%, 5.0%)
Race			
Black or African American	13/263 (4.9%)	0/126	4.9% (1.4%, 8.3%)
South Africa	11/207 (5.3%)	0/111	
United States	2/50 (4.0%)	0/13	
Europe	0/6	0/2	
Asian	1/161 (0.6%)	0/65	0.6% (−5.0%, 3.4%)
White	1/52 (1.9%)	0/35	1.9% (−8.1%, 10.1%)
American Indian or Alaska Native	0/6	0/1	
Native Hawaiian or Other Pacific Islander	0/1	0/1	
Multiple	0/1	0/2	
Other	0/2	0/1	
Region			
Asia	1/149 (0.7%)	0/63	0.7% (−5.1%, 3.7%)
Europe	1/43 (2.3%)	0/16	2.3% (−17.1%, 12.1%)
South Africa	11/207 (5.3%)	0/112	5.3% (1.3%, 9.3%)
United States	2/87 (2.3%)	0/40	2.3% (−6.6%, 8.0%)
Black or African American	2/50 (4.0%)	0/13	

Source: FDA Analysis; adsl.xpt; excluding participants from site 710-005. Participants with no confirmed (positive or negative) culture results were excluded.
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with given characteristic; OOW, out-of-window; TOC, test-of-cure

Microbiological Cure at TOC at Pharyngeal Site

The microbiological cure rates at the pharyngeal site were similar between the zoliflodacin arm and the ceftriaxone-azithromycin arm. See Table 11 for details. This analysis was based on a relatively small sample size.

Table 11. Microbiological Cure Rate at TOC at Pharyngeal Site, Micro-ITT Pharyngeal Population, Trial STI_Zoli001

Outcome	Zoliflodacin N=53 n (%)	Ceftriaxone-Azithromycin N=28 n (%)	Difference % (95% CI[a])
Cure	42 (79.2)	22 (78.6)	0.7% (−16.3%, 20.8%)
Negative	40 (75.5)	21 (75.0)	0.5% (−17.5%, 21.2%)
Indeterminate	2 (3.8)	1/28 (3.6)	
Failure	11 (20.8)	6 (21.4)	
Positive	4 (7.5)	1 (3.6)	
Nonassessable	7 (13.2)	5 (17.9)	
Missed TOC Visit	1 (1.9)	1 (3.6)	
Unobtainable specimen	3 (5.7)	2 (7.1)	
TOC out of window	3 (5.7)	2 (7.1)	
OOW negative	2 (3.8)	1 (3.6)	
OOW indeterminate	1 (1.9)	0	
OOW unobtainable	0	1 (3.6)	

Source: FDA Analysis; adcure.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in trial arm; n, number of participants with outcome; TOC, test-of-cure; OOW, out-of-window

Microbiological Cure at TOC at Rectal Site

The microbiological cure rates at the rectal site were similar between the zoliflodacin arm and the ceftriaxone-azithromycin arm. See Table 12 for details. This analysis was based on a relatively small sample size.

Table 12. Microbiological Cure Rate at TOC at Rectal Site, Micro-ITT Rectal Population, Trial STI_Zoli001

Outcome	Zoliflodacin N=79 n (%)	Ceftriaxone-Azithromycin N=35 n (%)	Difference % (95% CI[a])
Cure (negative)	69 (87.3)	31 (88.6)	1.2% (−14.4%, 12.6%)
Failure	10 (12.7)	4 (11.4)	
Positive	3 (3.8)	0	
Nonassessable	7 (8.9)	5 (14.3)	
Missed TOC Visit	3 (3.8)	1 (2.9)	
Unobtainable specimen	1 (1.3)	1 (2.9)	
TOC out of window	3 (3.8)	2 (5.7)	
OOW negative	2 (2.5)	2 (5.7)	
OOW unobtainable	1 (1.3)	0	

Source: FDA Analysis; adcure.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; TOC, test-of-cure; OOW, out-of-window

NG NAAT Result at TOC

The *N. gonorrhoeae* NAAT negative rates were similar between the zoliflodacin arm and the ceftriaxone-azithromycin arm at TOC. See Table 13 for details.

Table 13. *Neisseria gonorrhoeae* NAAT Results at TOC among *Neisseria gonorrhoeae* NAAT Baseline Positive Participants, Micro-ITT Population, Trial STI_Zoli001

NG NAAT Result at Urogenital Site, Micro-ITT (Urogenital) Population	Zoliflodacin N=492[b] n (%)	Ceftriaxone-Azithromycin N=233[b] n (%)
Negative	405 (82.3)	190 (81.5)
Positive	65 (13.2)	30 (12.9)
Indeterminate	5 (1.0)	6 (2.6)
Missing	17 (3.5)	7 (3.0)
Difference in negative rate between two arms % (95% CI[a])	0.8% (−5.0%, 7.1%)	

NG NAAT Result at the Pharyngeal Site, Micro-ITT Pharyngeal Population	Zoliflodacin N=45[b] n (%)	Ceftriaxone-Azithromycin N=24[b] n (%)
Negative	28 (62.2)	12 (50.0)
Positive	11 (24.4)	8 (33.3)
Indeterminate	3 (6.7)	2 (8.3)
Missing	3 (6.7)	2 (8.3)

NG NAAT Result at the Rectal Site, Micro-ITT Rectal Population	Zoliflodacin N=73[b] n (%)	Ceftriaxone-Azithromycin N=30[b] n (%)
Negative	60 (82.2)	27 (90.0)
Positive	8 (11.0)	1 (3.3)
Indeterminate	1 (1.4)	1 (3.3)
Missing	4 (5.5)	1 (3.3)

Source: FDA Analysis; adsl.xpt; admb.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
[b] Participant who tested negative for NG NAAT at Baseline were excluded from this analysis. Out of window test results were included in this analysis using the observed outcomes.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; NAAT, nucleic acid amplification test; NG, *Neisseria gonorrhoeae*; TOC, test-of-cure

Clinical Cure Rate in Male Participants at TOC

Clinical cure in participants was defined as participants assigned male at birth experiencing resolution at TOC of signs and symptoms of urogenital gonococcal infection that were present at Baseline. It was assessed in participants who have at least one sign or symptom of urethral gonorrhea at Baseline (e.g., urethral discharge and dysuria). This analysis was only defined for males because females were likely to be asymptomatic.

A lower clinical cure rate was observed in the zoliflodacin arm compared to the active control arm, although both arms had higher than 80% clinical cure rates. The difference between the two arms in clinical cure rates was similar to the result of the primary efficacy endpoint, with the upper bound of the 95% confidence interval below zero. A higher percentage of the clinical cures in the zoliflodacin arm were collected during out-of-window TOC Visits.

Table 14. Clinical Cure Rate at TOC, Clinical Cure Population, Trial STI_Zoli001

Outcome	Zoliflodacin N=460 n (%)	Ceftriaxone-Azithromycin N=220 n (%)	Difference % (95% CI[a])
Cured	375 (81.5)	194 (88.2)	−6.6% (−11.9%, −0.7%)
TOC at Day 4 – Day 8	364 (79.1)	193 (87.7)	−8.6% (−14.0%, −2.5%)
TOC at Day 9 – Day 13	7 (1.5)	0	
TOC at Day 14 – Day 20	4 (0.9)	1 (0.5)	
Not cured			
Improved	66 (14.3)	18 (8.2)	
Failure	1 (0.2)	0	
Unknown or missing	18 (3.9)	8 (3.6)	

Source: FDA Analysis; adcure.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; N, number of participants in treatment arm; n, number of participants with outcome; TOC, test-of-cure

6.2.3. Trial DMID 14-0014

6.2.3.1. Design, Trial DMID 14-0014

Trial DMID 14-0014 (clinicaltrials.gov identifier NCT02434503) was a phase 2, multicenter, randomized, open-label trial to compare a single dose of zoliflodacin to a single dose of ceftriaxone for treatment of uncomplicated gonorrhea in adult men and women, ages 18 to 55 years. Eligible participants were randomly assigned 70:70:40 to receive a single oral dose of 2000 mg of zoliflodacin, 3000 mg of zoliflodacin or an IM dose of 500 mg of ceftriaxone, respectively. The trial enrolled participants between November 10, 2014, and December 2, 2015, from five sites in the United States.

Participants were enrolled for 30±2 days, with trial visits to assess safety, efficacy and microbiological endpoints occurring at Baseline (Day 1), TOC (Day 6±2) and EOS (Day 30±2). The primary endpoints were the proportion of participants with microbiological cure at urethral or cervical sites in each trial arm at TOC after study drug administration and the proportion of participants reporting AEs and serious adverse events (SAEs) considered product related, following the dose of study product through EOS.

In addition to the primary endpoints of efficacy and safety, secondary endpoints included the following:

The proportion of participants with microbiological cure at rectal sites in each trial arm at TOC after study drug administration.

The proportion of participants with microbiological cure at pharyngeal sites in each trial arm at TOC after study drug administration.

The proportion of participants with no detectable *N. gonorrhoeae* nucleic acid in urethral, cervical, rectum, and pharynx specimens in each trial arm at Baseline and TOC.

The proportion of participants with clinical cure in each trial arm at TOC.

The in vitro MICs against zoliflodacin and ceftriaxone of gonococcal isolates from cultures taken at Baseline and TOC.

6.2.3.2. Eligibility Criteria, Trial DMID 14-0014

Participants had to meet the following criteria to be eligible for the trial:

Untreated participants with signs and symptoms of urethral or cervical gonorrhea or confirmed urethral or cervical gonorrhea (as defined by positive culture, NAAT, or Gram stain) or any type of sexual contact with an infected individual in the 14 days prior to enrollment.

Participant able to give voluntary written informed consent before any study-related procedure was performed.

Willingness to comply with all protocol requirements.

Male or non-pregnant female 18 to 55 years of age, inclusive.

Female participants must have had a negative urine pregnancy test at Visit 1 prior to receiving study drug.

Participants were willing to abstain from anal, oral, and vaginal sexual intercourse or use condoms for 7 days following study drug dosing to prevent potential gonococcal re-infection.

Male participants must have been surgically sterilized or have agreed to use condoms for 7 days following study drug dosing.

Female participants must have been of non-childbearing potential* or if of childbearing potential, participants must have been using a highly effective method of birth control**.

*Non-childbearing potential was defined as being post-menopausal for at least two years, status post-bilateral oophorectomy or status post-hysterectomy.

**Female participants must have avoided becoming pregnant by using one of the following acceptable methods of birth control for 30 days prior to study drug dosing:

Intrauterine contraceptive device; OR

Oral contraceptives; OR

Implanon®, Nexplanon®, DepoProvera®, contraceptive skin patch or NuvaRing®; OR

Tubal ligation; OR

Abstinence AND

For 30 days following dosing, any method above should have been used plus the required use of a barrier method (condom) by the male partner (even if vasectomized)

Participants who met any of the following specified criteria were excluded from the study, i.e., confirmed or suspected, complicated or systemic gonorrhea such as pelvic inflammatory disease, testicular pain, epididymitis, arthritis, conjunctivitis, endocarditis, or clinical proctitis.

6.2.3.3. Statistical Analysis Plan, Trial DMID 14-0014

The primary efficacy endpoint was microbiological cure at urethral or cervical sites on Day 6/TOC in the micro-ITT efficacy analysis population; this was repeated as a secondary analysis in the PP efficacy analysis population.

The following analysis populations were included in the trial:

Micro-ITT Population: all randomized participants who had *N. gonorrhoeae* isolated at Baseline.

Clinical-intent-to-treat Population: participants in the micro-ITT population with at least one sign or symptom of gonorrhea at Baseline.

PP Efficacy Population: all participants in the micro-ITT population who met all inclusion/exclusion criteria, complied with study treatment, were not diagnosed with a concomitant infection besides *Chlamydia* or bacterial vaginosis, did not receive any systemic antibiotic other than the study regimen prior to TOC, and returned to the study site for the Test of Cure Visit within the specified window (TOC, Study Day 6±2 relative to the first day of treatment).

Clinical-PP Population: participants in the PP population with at least one sign or symptom of gonorrhea at Baseline and with an evaluable clinical cure outcome.

Safety Population: all randomized participants who received study treatment.

The primary efficacy endpoint was the proportion of participants with microbiological cure at urethral or cervical sites in each trial arm at TOC after study drug administration, evaluated in the micro-ITT population. Microbiological Cure was defined as *N. gonorrhoeae* not detectable by culture. Participants who did not comply with study treatment, did not return for the TOC Visit, had a non-evaluable culture result at the TOC Visit, or who received antibacterial therapy with activity against *N. gonorrhoeae* for a coinfection other than *Chlamydia,* were classified as treatment failures.

The secondary efficacy endpoints for the trial were:

The proportion of participants with microbiological cure at rectal sites in each trial arm at TOC after study drug administration.

The proportion of participants with microbiological cure at pharyngeal sites in each trial arm at TOC after study drug administration.

The proportion of participants with no detectable *N. gonorrhoeae* nucleic acid in urethral, cervical, rectum, and pharynx specimens in each trial arm at Baseline and TOC.

The proportion of participants with clinical cure in each trial arm at TOC.

The in vitro MICs against zoliflodacin and ceftriaxone of gonococcal isolates from cultures taken at Baseline and TOC.

No formal statistical hypothesis testing was performed for this trial. All efficacy endpoints were summarized descriptively.

6.2.3.4. Results of Analyses, Trial DMID 14-0014

A total of 181 participants were screened. Among them, 180 participants were enrolled in the trial and 179 participants were treated. One individual was enrolled twice, and all analyses considered these two enrollments as separate individuals.

Table 15. Participant Disposition, Trial DMID 14-0014

Parameter	Zoliflodacin 2000 mg N=72 n (%)	Zoliflodacin 3000 mg N=67 n (%)	Ceftriaxone 500 mg N=41 n (%)
Participants randomized	72 (100)	67 (100)	41 (100)
Safety population	72 (100)	67 (100)	40 (97.6)
Micro-ITT urethral/cervical population	57 (79.2)	56 (83.6)	28 (68.3)
Micro-ITT pharyngeal population	8 (11.1)	11 (16.4)	4 (9.8)
Micro-ITT rectal population	5 (6.9)	7 (10.4)	3 (7.3)
Per-protocol urethral/cervical population	49 (68.1)	47 (70.1)	21 (51.2)
Per-protocol pharyngeal population	6 (8.3)	9 (13.4)	4 (9.8)
Per-protocol rectal population	4 (5.6)	6 (9.0)	3 (7.3)
Clinical-ITT population	57 (79.2)	49 (73.1)	27 (65.9)
Clinical-PP population	49 (68.1)	41 (61.2)	21 (51.2)
Discontinued trial	4 (5.6)	1 (1.5)	2 (4.9)
Enrolled but treatment not administered	0	0	1 (2.4)
Lost to follow-up	4 (5.6)	1 (1.5)	0
Voluntary withdrawal by participant	0	0	1 (2.4)

Source: FDA analysis; adsl.xpt
Abbreviations: micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with disposition; PP, per-protocol

Demographic characteristics are listed in Table 16. This trial was conducted in the United States. The majority of participants were male (93%), not Hispanic (93%), and African American (59%).

Table 16. Baseline Demographics and Clinical Characteristics, Safety Population, Trial DMID 14-0014

Characteristic	Zoliflodacin 2000 mg N=72	Zoliflodacin 3000 mg N=67	Ceftriaxone 500 mg N=40
Sex, n (%)			
Female	2 (2.8)	8 (11.9)	2 (5.0)
Male	70 (97.2)	59 (88.1)	38 (95.0)
Age, years			
Mean (SD)	29.2 (8.86)	28.3 (7.63)	29.3 (8.16)
Median	26.0	26.0	28.0
Min, max	18.0, 53.0	18.0, 52.0	18.0, 53.0
Race, n (%)			
Black or African American	44 (61.1)	40 (59.7)	22 (55.0)
White	24 (33.3)	19 (28.4)	15 (37.5)
Asian	1 (1.4)	2 (3.0)	0
American Indian or Alaska Native	1 (1.4)	0	0
Native Hawaiian or Other Pacific Islander	0	0	1 (2.5)
Multiracial	2 (2.8)	5 (7.5)	2 (5.0)
Other/Unknown	0	1 (1.5)	0
Ethnicity, n (%)			
Hispanic	5 (6.9)	4 (6.0)	4 (10.0)
Not Hispanic	67 (93.1)	63 (94.0)	36 (90.0)

Source: FDA analysis; adsl.xpt
Abbreviations: max, maximum; min, minimum; N, number of participants in treatment arm; n, number of participants with characteristic; SD, standard deviation

Microbiological Cure at TOC (Including the Primary Efficacy Endpoint)

Table 17 displays results for microbiological cure at TOC in the micro-ITT population. All three arms had cure rates above 90% at the urethral/cervical site. The number of participants in the pharyngeal and rectal site groups was small.

Table 17. Proportion of Participants With Microbiological Cure at TOC, Micro-ITT Population, Trial DMID 14-0014

Microbiological Cure at the Urethral/Cervical Site	Zoliflodacin 2000 mg N=57	Zoliflodacin 3000 mg N=56	Ceftriaxone 500 mg N=28
Cure, n (%)	55 (96.5)	54 (96.4)	28 (100)
Negative, TOC at Day 4 – Day 8	51 (89.5)	48 (85.7)	22 (78.6)
Negative, TOC at Day 9 (OOW)	1 (1.8)	5 (8.9)	2 (7.1)
Negative, TOC at Day 11 – Day 28 (OOW)	3 (5.3)	1 (1.8)	4 (14.3)
Difference in cure rate between zoliflodacin and ceftriaxone % (95% CI[a])	−3.5 (−11.9, 8.8)	−3.6 (−12.1, 8.8)	
Difference in cure rate between zoliflodacin (pooled) and ceftriaxone % (95% CI[a])		−3.5 (−8.7, 8.7)	
Microbiological Cure at the Pharyngeal Site	**Zoliflodacin 2000 mg N=8**	**Zoliflodacin 3000 mg N=11**	**Ceftriaxone 500 mg N=4**
Cure, n (%)	4 (50.0)	8 (81.8)	4 (100)
Microbiological Cure at the Rectal Site	**Zoliflodacin 2000 mg N=5**	**Zoliflodacin 3000 mg N=7**	**Ceftriaxone 500 mg N=3**
Cure, n (%)	5 (100)	7 (100)	3 (100)

Source: FDA Analysis; adsl.xpt
[a] 95% CI was calculated with Newcombe score method.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; OOW, out-of-window; TOC, test-of-cure

The primary efficacy results did not impute any out-of-window culture results. A higher percentage of negative out-of-window culture results was observed in the ceftriaxone arm.

***Neisseria gonorrhoeae* Nucleic Acid Results at TOC**

Table 18 displays *N. gonorrhoeae* nucleic acid test results at TOC in the micro-ITT population. All three arms had lower than 20% positive *N. gonorrhoeae* nucleic acid rates in the urethral/cervical sites. The number of participants in the pharyngeal and rectal site groups was small.

Table 18. *Neisseria gonorrhoeae* Nucleic Acid Result at TOC, Micro-ITT Population, Trial DMID 14-0014

NG Nucleic Acid Result at the Urethral/Cervical Site	Zoliflodacin 2000 mg N=57	Zoliflodacin 3000 mg N=56	Ceftriaxone 500 mg N=27[b]
Negative, n (%)	48 (84.2)	42 (75.0)	24 (88.9)
Positive, n (%)	9 (15.8)	10 (17.9)	3 (11.1)
Indeterminate, n (%)	0	4 (7.1)	0
Difference in negative rate between zoliflodacin and ceftriaxone % (95% CI[a])	−4.7 (−18.3, 13.8)	−13.9 (−28.5, 5.5)	
Difference in negative rate between zoliflodacin (pooled) and ceftriaxone % (95% CI[a])		−9.2 (−20.3, 8.9)	

NG Nucleic Acid Result at the Pharyngeal Site	Zoliflodacin 2000 mg N=8	Zoliflodacin 3000 mg N=11	Ceftriaxone 500 mg N=4
Negative, n (%)	2 (25.0)	6 (54.5)	2 (50.0)
Positive, n (%)	6 (75.0)	5 (45.5)	2 (50.0)
NG Nucleic Acid Result at the Rectal Site	**Zoliflodacin 2000 mg N=5**	**Zoliflodacin 3000 mg N=7**	**Ceftriaxone 500 mg N=3**
Negative, n (%)	5 (100.0)	7 (100.0)	3 (100.0)
Positive, n (%)	0	0	0

Source: FDA Analysis; adsl.xpt
[a] 95% CI was calculated with Newcombe score method.
[b] One Ceftriaxone participant who tested negative for NG Nucleic Acid at Baseline was excluded from this analysis.
Abbreviations: CI, confidence interval; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; NG, *Neisseria gonorrhoeae*; TOC, test-of-cure

Table 19 displays clinical cure results at TOC in the clinical-intent-to-treat population. All three arms had above 90% clinical cure rates in this trial.

Table 19. Proportion of Participants With Clinical Cure at TOC, Clinical-ITT Population, Trial DMID 14-0014

Outcome	Zoliflodacin 2000 mg N=57	Zoliflodacin 3000 mg N=49	Ceftriaxone 500 mg N=27
Cure, n (%)	52 (91.2)	45 (91.8)	26 (96.3)
Non-cure, n (%)	5 (8.8)	4[b] (8.2)	0
Missing, n (%)	0	0	1 (3.7)
Difference in cure rate between zoliflodacin and ceftriaxone % (95% CI[a])	−5.1 (−15.7, 10.3)	−4.5 (−15.9, 10.9)	
Difference in cure rate between zoliflodacin (pooled) and ceftriaxone % (95% CI[a])		−4.8 (−12.3, 10.3)	

Source: FDA Analysis; adsl.xpt
[a] 95% CI was calculated with Newcombe score method.
[b] One ceftriaxone 3000 mg participant who vomited within 30 minutes of study product administration and administered additional antimicrobial treatment, was classified as a treatment failure in this analysis.
Abbreviations: CI, confidence interval; ITT, intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; TOC, test-of-cure

6.3. Key Efficacy Review Issues

6.3.1. Imbalance in Rescue Treatment and Post-TOC Infections in Trial STI_Zoli001

Issue

A greater number of participants in the zoliflodacin arm, compared to the ceftriaxone-azithromycin arm, were administered concomitant systemic antibacterials. There was also a greater number of infections captured after the TOC visit in the zoliflodacin arm.

Background

In Trial STI_Zoli001, there was an imbalance between the two arms in use of systemic antibacterials as concomitant medications, with a greater number of participants in the

zoliflodacin arm receiving these medications. This imbalance may reflect treatment failures not captured in the primary efficacy analysis or participants with concurrent *Chlamydia* infections which would not be covered by zoliflodacin but would be covered by azithromycin in the comparator arm. Additionally, there was also an imbalance in infections observed at the EOS Visit, with more reported in the zoliflodacin arm. These appeared to be new infections necessitating antibacterial treatment, or infections not identified using a culture-based primary endpoint at the TOC visit.

Assessment

In Trial STI_Zoli001, key efficacy analyses were focused on the micro-ITT populations for urogenital, pharyngeal, and rectal sites. Participants could have more than one site of infection. Many participants also had coinfection with *Chlamydia trachomatis*. Zoliflodacin has no activity against *C. trachomatis* while the comparator arm included azithromycin which conveys antibacterial coverage for possible coinfection with *C. trachomatis*. Table 20 below summarizes the number of participants with different infection sites or coinfections.

Table 20. Participant Populations, Trial STI_Zoli001

Populations	Zoliflodacin	Ceftriaxone-Azithromycin
Any-Micro-ITT population[a]	519	247
Micro-ITT (urogenital) population, n (%[b])	506 (97.5)	238 (96.4)
Micro-ITT pharyngeal population, n (%[b])	53 (10.2)	28 (11.3)
Micro-ITT rectal population, n (%[b])	79 (15.2)	35 (14.2)
Single site infection at Baseline, n (%[b])	414 (79.8)	204 (82.6)
Two sites of infections at Baseline, n (%[b])	91 (17.5)	32 (13.0)
Three sites of infections at Baseline, n (%[b])	14 (2.7)	11 (4.5)
Any culture positive at TOC, n (%[b])	20 (3.9%)	1 (0.4)
Any CT Coinfection at Baseline in Any-Micro-ITT Population, n (%[b])	150 (28.9)	75 (30.4)

Source: FDA Analysis; adsl.xpt; excluding participants from site 710-005
[a] Any-Micro-ITT population includes participants who were in any-Micro-ITT population of urogenital, pharyngeal, or rectal sites.
[b] Percentages were calculated over Any-Micro-ITT population.
Abbreviations: CT, *Chlamydia trachomatis*; micro-ITT, microbiological intent-to-treat; n, number of participants in specified population or group; TOC, test-of-cure

Given the fact that participants were treated based on the overall infection status regardless of site, the Any-Micro-ITT population is defined in this section of the review to include participants who were in any of the micro-ITT populations for urogenital, pharyngeal, or rectal sites.

Among those in the Any-Micro-ITT population, approximately 30% were coinfected with *C. trachomatis* at Baseline. Azithromycin was included in the control arm, as the combination of ceftriaxone and azithromycin was standard of care at the time of trial design. It is active against strains of *N. gonorrhoeae* as well as *C. trachomatis*. However, zoliflodacin does not have known clinical efficacy against *C. trachomatis*. In the Any-Micro-ITT population, 123 (23.7%) participants in the zoliflodacin arm reported use of azithromycin as a concomitant medication as compared to 8 (3.6%) in the control arm; 65 (12.5%) participants in the zoliflodacin arm reported use of doxycycline/doxycycline hyclate as a concomitant medication as compared to 7 (2.8%) in the control arm. These imbalances likely reflect additional coverage needed for *C. trachomatis* coinfection in the zoliflodacin arm.

However, IM ceftriaxone is the recommended first-line treatment for uncomplicated gonorrhea and cefixime is an alternative oral cephalosporin option when ceftriaxone administration is not available or not feasible.[20] Imbalances in the concomitant administration of ceftriaxone/cefixime in Trial STI_Zoli001 could raise concern regarding the efficacy of zoliflodacin. Table 21 summarizes concomitant use of ceftriaxone/cefixime in Trial STI_Zoli001.

Table 21. Concomitant Use of Ceftriaxone/Cefixime, Any-Micro-ITT Population, Trial STI_Zoli001

Use of Ceftriaxone/Cefixime	Zoliflodacin N=519	Ceftriaxone-Azithromycin N=247
Any use of ceftriaxone/cefixime n (%[a])	46 (8.9)	8 (3.2)
Used before TOC, TOC results all negative	1 (0.2)	2 (0.8)
Used on/after TOC, TOC any positive culture	17 (3.3)	0
Used on/after TOC, TOC no positive culture	28 (5.4)	6 (2.4)
Ceftriaxone/cefixime use on Day 6 – Day 9	5 (1.0)	0
Ceftriaxone/cefixime use on Day 14 – Day 22	8 (1.5)	2 (0.8)
Ceftriaxone/cefixime use Day 25 – Day 44	15 (2.9)	4 (1.6)

Source: FDA Analysis; adsl.xpt, adcm.xpt, admb.xpt; excluding participants from site 710-005
[a] Percentages were calculated over Any-Micro-ITT population.
Abbreviations: micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants who used ceftriaxone/cefixime; TOC, test-of-cure

At TOC, 20 participants in the zoliflodacin arm and one participant in the control arm had a positive *N. gonorrhoeae* culture at one or more sites. Among them, 17 participants from the zoliflodacin arm reported use of ceftriaxone/cefixime on or after the TOC Visit, which could be considered rescue therapy. Another 28 (5.4%) participants in the zoliflodacin arm and six (2.4%) participants in the control arm reported use of ceftriaxone/cefixime around the TOC Visit (Day 6 to Day 9), after the TOC Visit but before the EOS Visit (Day 14 to Day 22), and on or after the EOS Visit (Day 25 to Day 44). Most of those participants were treated based on positive NAAT results and/or symptoms at TOC and/or EOS. These additional treatments were provided either for treatment failures of the original infection or for treatment of new infections.

Table 22 summarizes outcomes that may lead to consideration of retreatment. Note that EOS tests were not required for all participants. However, higher percentages of positive culture/positive NAAT/signs or symptoms at EOS were observed in participants from the zoliflodacin arm as compared to the active control arm.

[20] See Footnote 7

Table 22. Summarized Outcomes in Any-Micro-ITT Population, Trial STI_Zoli001

Characteristics n (%[a])	Zoliflodacin N=519	Ceftriaxone-Azithromycin N=247
TOC any culture positive	20 (3.9)	1 (0.4)
TOC any NAAT positive	108 (20.8)	55 (22.3)
TOC any signs/symptoms present	120 (23.1)	51 (20.6)
TOC Visit missing	15 (2.9)	6 (2.4)
EOS any signs/symptoms present	36 (6.9)	4 (1.6)
EOS any culture positive	11 (2.1)	0
EOS any NAAT positive	26 (5.0)	3 (1.2)
EOS Visit missing	37 (7.1)	17 (6.9)

Source: FDA Analysis; adsl.xpt, adrs.xpt, admb.xpt; excluding participants from site 710-005
[a] Percentages were calculated over Any-Micro-ITT population.
Abbreviations: EOS, end-of-study; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with characteristic; NAAT, nucleic acid amplification test; TOC, test-of-cure

The reviewer conducted the following exploratory analyses in the Any-Micro-ITT population to evaluate potential treatment failures not captured in the key efficacy analyses. Two scenarios were explored:

Scenario 1: Failure is defined as any TOC *N. gonorrhoeae* culture positive, or any EOS *N. gonorrhoeae* culture positive, or any reported use of ceftriaxone/cefixime with all available TOC and EOS *N. gonorrhoeae* cultures negative.

Scenario 2: Failure is defined as any TOC *N. gonorrhoeae* culture positive, or any EOS *N. gonorrhoeae* culture or *N. gonorrhoeae* NAAT positive, or any reported use of ceftriaxone/cefixime with all available TOC and EOS *N. gonorrhoeae* cultures/NAATs negative.

Table 23. Exploratory Analyses on Treatment Failures, Any-Micro-ITT Population, Trial STI_Zoli001

Scenario 1	Zoliflodacin N=519	Ceftriaxone-Azithromycin N=247
Failure n (%)	52 (10.0)	9 (3.6)
TOC NG culture positive, any site	20 (3.9)	1 (0.4)
EOS NG culture positive, any site, TOC NG culture not positive	10 (1.9)	0
Use of ceftriaxone/cefixime, no observed positive NG culture at TOC and EOS	22 (4.2)	8 (3.2)
Difference in failure rate between two arms % (95% CI[a])	6.4% (2.5%, 9.7%)	

Scenario 2	Zoliflodacin N=519	Ceftriaxone-Azithromycin N=247
Failure n (%)	63 (12.1)	12 (4.9)
TOC NG culture positive, any site	20 (3.9)	1 (0.4)
EOS NG culture or NAAT positive, any site, TOC NG culture not positive	24 (4.6)	3 (1.2)
Use of ceftriaxone/cefixime, no observed positive NG culture at TOC, no observed positive NG culture/NAAT at EOS	19 (3.7)	8 (3.2)
Difference in failure rate between two arms % (95% CI[a])	7.3% (3.0%, 11.0%)	

Source: FDA Analysis; adsl.xpt; admb.xpt; adrs.xpt; adcm.xpt; excluding participants from site 710-005
[a] 95% CI was calculated with Newcombe score method
[b] Participant who tested negative for NG NAAT at Baseline were excluded from this analysis. Out of window test results were included in this analysis using the observed outcomes.
Abbreviations: CI, confidence interval; EOS, end-of-study; micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with outcome; NAAT: nucleic acid amplification test; NG, *Neisseria gonorrhoeae*; TOC, test-of-cure

In both scenarios, zoliflodacin was inferior to ceftriaxone-azithromycin, but the upper bounds of the 95% CI of failure rate differences are below 12%. This is consistent with the conclusion that the zoliflodacin treatment effect is non-inferior to ceftriaxone-azithromycin if using a 12% margin. However, the trial was not blinded, so the exploratory analyses should be interpreted with caution.

Conclusion

The treatment failure rate of zoliflodacin was higher than ceftriaxone-azithromycin in Trial STI_Zoli001 based on additional exploratory analyses. However, the difference in treatment failure rates is still within a clinically acceptable margin of 12%.

6.3.2. Comparative Efficacy of Zoliflodacin and Comparator in Trial STI_Zoli001

Issue

Zoliflodacin demonstrated noninferiority with a margin of 10%, to the active control of ceftriaxone-azithromycin in Trial STI_Zoli001, but the active control was superior to zoliflodacin in this trial.

Background

As shown in Table 9, in Trial STI_Zoli001, the microbiological cure rates at the urogenital site at TOC were 90.9% in the zoliflodacin arm and 96.2% in the ceftriaxone-azithromycin arm in the micro-ITT population, with a difference of −5.3% (95% CI: −8.7%, −1.4%). In this case, zoliflodacin demonstrated noninferiority to the active control of ceftriaxone-azithromycin in Trial STI_Zoli001, but the active control was also superior to zoliflodacin in this trial, as the confidence interval is completely below zero.

Assessment

In Trial STI_Zoli001, the lower bound of the 95% confidence interval for the difference in microbiological cure rates in the primary efficacy analysis is −8.7%, demonstrating noninferiority of treatment with zoliflodacin to the active control of ceftriaxone-azithromycin with a noninferiority margin of 12%. Noninferiority could also be demonstrated if a more conservative margin of 10% was used, as recommended in the Agency's current uncomplicated gonorrhea guidance.[21]

However, the confidence interval was completely below zero, showing that the active control of ceftriaxone-azithromycin was superior to zoliflodacin in this trial. From a statistical perspective, this does not invalidate the demonstration of noninferiority for the primary analysis, because a drug may be within the clinically acceptable noninferiority margin relative to the control even though the control is statistically superior.

Both the zoliflodacin and ceftriaxone-azithromycin arms had treatment success rates of >90% in Trial STI_Zoli001. Given the unmet need for additional treatments for gonorrhea, including drug-resistant strains, the use of potentially two effective drugs in the comparator arm, the overall high efficacy, and the achievement of a tighter NI margin (10%) than the pre-specified NI margin (12%), the imbalance in failure rates disfavoring zoliflodacin was considered to be clinically acceptable.

Conclusion

The imbalance in failure rates disfavoring zoliflodacin in Trial STI_Zoli001 was considered to be clinically acceptable.

7. Safety (Risk and Risk Management)

7.1. Potential Risks or Safety Concerns Based on Nonclinical Data

Nonclinical safety studies submitted to support the safety evaluation of zoliflodacin included pharmacology studies (primary, secondary, and safety pharmacology), pharmacokinetics studies (absorption, distribution, metabolism, and excretion), toxicology studies (repeat-dose studies in rats [up to 4 weeks] and dogs [up to 4 weeks]), genetic toxicology assays (in vitro and in vivo),

[21] See Footnote 1

and reproductive and developmental toxicity studies in rats and mice. Additional details are available in Section 13. See Section 19 for a review of the primary pharmacology studies.

Secondary Pharmacology and Safety Pharmacology

In an in vitro human ether-a-go-go related gene (hERG) study, zoliflodacin inhibited hERG currents in a concentration-dependent manner with a half-maximal inhibitory concentration (IC_{50}) of 449µM, which is about 8-fold higher than the C_{max} calculated from clinical studies. In a safety pharmacology study in dogs, an oral 4-week repeat dose toxicity study in dogs, and an intravenous (IV) study in anesthetized guinea pigs, other reported cardiac effects included decreased blood pressure, increased heart rate, increase in cardiac contractility, and decreased cardiac relaxation. No arrhythmias or adverse QTc prolongation were detected in the IV-infusion safety pharmacology study in dogs.

In in vitro radioligand binding assays, adenosine transporter (guinea pig), gamma-aminobutyric acid A ($GABA_A$) chloride channel, tert-butylbicyclophosphorothionate (rat), $GABA_A$ chloride channel, tert-butylbicyclo-ortho-benzoate (rat), and serotonin (5-hydroxytryptamine) 5-HT2B receptor were inhibited by zoliflodacin with IC_{50s} of 13.4, 22.2, 26.2, and 58.2 µg/mL, respectively.

In a safety pharmacology functional observational battery screen in rats, to evaluate subtle neurological changes with a 250 mg/kg IV infusion of zoliflodacin (plasma concentrations at 2-hours about 4-fold the clinical C_{max}), changes in pupil size, body temperature, rolling gait, hunched posture, decreased grip strength, decreased spontaneous activity, and traction response were observed at time points between 15 and 120 minutes post-infusion.

Nonadverse large increases in water consumption were noted in rodent studies (up to 3 times the consumption of control animals). Water consumption was not measured in the dog studies. A dedicated safety pharmacology study in male rats found diuretic effects, but zoliflodacin did not affect plasma electrolytes in repeat-dose oral studies in rats or dogs or urinalysis parameters in dogs, and no adverse kidney effects were found in either species.

ADME/PK

Oral bioavailability in mice, rats, and dogs was 46%, 28%, and 42% to 95%, respectively. In an IV dosed tissue distribution study with radiolabeled zoliflodacin in rats, the highest levels were detected in kidneys and liver, and the zoliflodacin concentration was not higher in pigmented tissues. Small amounts of radioactivity were detectable in the brain up to 1 hour after administration. Radioactivity was detectable in the liver up to 24 hours after administration. The drug $t_{1/2}$ in repeat-dose studies in rats orally administered zoliflodacin increased from about 2 hours at 200 mg/kg/day to about 9 hours at 1000 mg/kg/day. The $t_{1/2}$ in dogs increased from about 3 hours at 100 mg/kg/day to about 5 hours at 5000 mg/kg/day. In mice, the $t_{1/2}$ was 1 to 2 hours and did not appear to be dose dependent. In rats, the only reported metabolite in the plasma was M3 (10.3%). In bile duct cannulated rats orally administered zoliflodacin, excretion was via the bile (52%), feces (24%), and urine (15%). Elimination was similar in rats with IV administration.

Target Organs of Toxicity

Male Reproductive Organ Toxicity and Fertility

Toxicology studies with zoliflodacin assessing toxicity targeting male reproductive organs and male fertility included histopathology assessments in a 4-week repeat dose rat study with a 3-month recovery period, histopathology assessments in a 4-week repeat dose dog study with a 3-month recovery period, and a fertility study in male rats.

After 4 weeks of zoliflodacin administration in male rats, fertility decreased 100% at the 1000 mg/kg/day dose (about 8-fold the MRHD by area under the concentration-time curve [AUC_{0-24}]) and decreased 31% at the 500 mg/kg/day dose (about 4-fold the MRHD by AUC_{0-24h}), with no effect on fertility at the 200 mg/kg/day dose (about 2-fold the MRHD by AUC_{0-24h}). Fertility recovered completely in all rats after a 4-week recovery period.

Male reproductive histopathology was reported in rat fertility and rat and dog repeat-dose toxicity studies. In the repeat-dose rat toxicity study, at the end of 4 weeks of oral dosing, minimal to mild testicular degeneration was reported at the 1000 mg/kg/day dose (about 11-fold the MRHD by AUC_{0-24h}), minimal to mild cellular debris in the epididymides was reported at and above the 500 mg/kg/day (about 7-fold the MRHD by AUC), and mild to moderate secretory depletion of seminal vesicles was seen in all test article groups (about 3-fold the MRHD by AUC_{0-24h}). There was incomplete recovery of the histopathological changes at the end of the 3-month recovery period.

At the end of 4 weeks of oral dosing in the dog study, one male of three in the 500 mg/kg/day (about 8-fold the MRHD by AUC_{0-24h}) dose group had moderate testicular degeneration and moderate cellular debris in the epididymides, and one male of three in the 200 mg/kg/day (about 8-fold the MRHD by AUC_{0-24h}) dose group had mild testicular degeneration. At the end of a 3-month recovery period, recovery was incomplete as hypospermatogenesis was observed in two of three high dose males. In the male rat fertility study, following 5 weeks of dosing and a 57-day recovery period, histopathological changes were noted in the 500 and 1000 mg/kg/day dose groups (about 4-fold and 8-fold the MRHD by AUC_{0-24h}, respectively) in the epididymides (minimal cellular debris associated with testicular degeneration) and testes (minimal to marked tubular degeneration, minimal tubular vacuolation, and minimal cellular debris). No male reproductive organ histopathology was reported in 14-day IV studies in rat or dog up to the highest doses tested, i.e., 250 mg/kg/day in the rat (about 6-fold the MRHD by AUC_{0-24h}) and 100 mg/kg/day in the dog (about 3-fold the MRHD by AUC_{0-24h}).

Female Reproduction and Developmental Toxicity

Reproductive toxicology studies with zoliflodacin included a male rat fertility study, a female rat fertility and embryo-fetal development (FEFD) study, a mouse embryo-fetal development (EFD) study, and a rat pre- and postnatal development study.

Increased pre-implantation and post-implantation losses were reported in untreated female rats mated with males dosed with 500 mg/kg/day zoliflodacin. Early intrauterine deaths (dead implants and early embryonic deaths) were increased 3-fold in female rats dosed from 2 weeks prior to mating with untreated males and through organogenesis (rat FEFD study) at doses of 500 mg/kg/day zoliflodacin and above. A reduction in mean numbers of implants and mean live

implants was reported in gravid mice administered 500 mg/kg/day zoliflodacin and above during organogenesis.

Litter weights in the rat FEFD study were reduced dose-dependently in all dose groups and fetal weights were reduced in the mouse EFD study in the 1000 mg/kg/day dose group. A decrease in skeletal ossification was found in litters in the rat FEFD study from all dose groups and in the mouse EFD study in the 500 and 1000 mg/kg/day dose groups.

In the mouse EFD study, exencephaly, a neural tube malformation, was present in litters in the 500 and 1000 mg/kg/day groups at levels above current and historical control rates.

No malformations in first- or second-generation animals were reported in the rat pre- and postnatal development study; however, the highest dose tested was 200 mg/kg/day (up to 2-fold the clinical exposure during organogenesis), which was a lower maximum dose than in the other developmental studies. In an open field test, ambulatory counts and distance traveled were higher in 200 mg/kg/day dose group first generation (F1) males and 100 and 200 mg/kg/day dose group F1 females compared to controls. Maternal exposures measured at the end of lactation were lower than those measured during gestation. The learning and memory testing in this study was not adequately conducted so no conclusions can be drawn on these endpoints.

Table 24. Safety Margins

Study	NOAEL (mg/kg/day)	Nonclinical Exposure (µg*h/mL) [Exposure Multiples[a]]	Adverse Effects (Lowest Dose With Findings [Exposure Multiples[a]])
General Toxicity			
4-week oral rat	200	1075 [3]	• Testicular toxicity (minimal to mild tubular degeneration) (1000 mg/kg/day [11]) • Epididymal cellular debris (minimal to mild) (500 mg/kg/day [7])
4-week oral dog	100	848 [3]	• One female was terminated early (500 mg/kg/day) [9] • Reduced RBC parameters (males: 6% to 16%, females: 9% to 11%) (200 mg/kg/day [7]). Increased reticulocytes (100 mg/kg/day [3]) • Moderate testicular degeneration and cellular debris in the epididymides (500 mg/kg/day [8]) • Mild testicular degeneration (200 mg/kg/day [8])
14-day IV rat	250	2774 [9]	None
14-day IV dog	100	972 [3]	None
14-day rat oral (impurities)	500	1410 [5]	None

Study	NOAEL (mg/kg/day)	Nonclinical Exposure (µg*h/mL) [Exposure Multiples[a]]	Adverse Effects (Lowest Dose With Findings [Exposure Multiples[a]])
Fertility and Developmental Toxicity Studies			
Male rat FEED	200	495 [2]	• Decreased fertility (500 mg/kg/day [4]) • Increased early pregnancy loss (500 mg/kg/day [4])
Female rat FEFD	(200)[b]	(1082) [3[c]]	• Increased intrauterine deaths (500 mg/kg/day [6[c]]) • Reduced fetal and litter weights (200 mg/kg/day [3[c]]) • Decreased ossification (200 mg/kg/day [3[c]])
Mouse EFD	250	204 [0.6]	• Fewer implants and mean live implants (500 mg/kg/day [2]) • Fetal exencephaly, a rare malformation, was present (500 mg/kg/day [2]). • Decreased fetal weights (1000 mg/kg/day [3]) • Decreased ossification (500 mg/kg/day [2])
Rat PPND	50[d]	42 [0.1[e]]	In an open field test, ambulatory counts and distance traveled were higher in 200 mg/kg/day dose group F1 males and 100 and 200 mg/kg/day dose group F1 females compared to controls [0.2 to 2].

Source: Reviewer generated table
[a] Compared to clinical $AUC_{0\text{-}24h}$ =327 µg*h/mL from report ICPD 00734-1.
[b] LOAEL
[c] No direct measure was available. AUC extrapolated from same dose in the dose-range finding rat embryo fetal development study.
[d] Not considering learning and behavior testing which was not adequate.
[e] Exposure as measured on lactation Day 21.
Abbreviations: AUC, area under the concentration-time curve; EFD, embryo-fetal development; FEFD, fertility and embryo-fetal development; IV, intravenous; LOAEL, lowest-observed adverse effect level; NOAEL, no-observed-adverse-effect level; PPND, pre- and postnatal development; RBC, red blood cell

7.2. Potential Risks or Safety Concerns Based on Drug Class or Other Drug-Specific Factors

Zoliflodacin is a novel, bactericidal, first-in-class spiropyrimidinetrione bacterial type II topoisomerase inhibitor that impedes bacterial DNA replication through binding to, and inhibition of the *GyrB* subunit of topoisomerase II (DNA gyrase and topoisomerase IV). While other antibacterials, i.e., fluoroquinolones and gepotidacin are also topoisomerase inhibitors, they differ structurally from zoliflodacin and bind to the *GyrA* subunit of the bacterial topoisomerase enzyme. Because of this difference in binding site, no cross-resistance has been detected between zoliflodacin and either gepotidacin or the fluoroquinolones.

The Applicant did not identify adverse events of special interest specific to zoliflodacin. Of note, known and labeled AEs associated with fluroquinolones include musculoskeletal events (e.g., tendinitis, tendon rupture), QTc prolongation, hepatotoxicity and *Clostridioides difficile* infection. AEs associated with gepotidacin include acetylcholinesterase inhibition, QTc prolongation and *C. difficile* infection. Although, zoliflodacin is structurally distinct from gepotidacin and the fluoroquinolone class of drugs, the FDA review team evaluated for potential

overlap in the drugs' safety profiles. A comparison of TEAEs in fluroquinolones, gepotidacin, and zoliflodacin is detailed in Section 7.6.1.5.1.

7.3. Potential Risks or Safety Concerns Identified Through Postmarket Experience

Not applicable. Zoliflodacin is not approved or marketed in any country at this time.

7.3.1. Adverse Events Identified in Postmarket Experiences

Not applicable.

7.3.2. Expectations on Safety

Not applicable.

7.3.3. Additional Safety Issues From Other Disciplines

Please refer to Section 7.1 for the non-clinical safety signals identified. Based on the nonclinical findings detailed there, the Division of Urology, Obstetrics and Gynecology (DUOG) and the Division of Pediatrics and Maternal Health (DPMH) were consulted for recommendations regarding PMRs and labeling recommendations. DUOG recommended labeling to address potential male infertility and testicular toxicity as well as a human sperm study as a PMR.

DPMH recommended labeling to address the potential for embryo-fetal toxicity in pregnant women and recommended pregnancy testing prior to treatment with zoliflodacin. Additionally, a Descriptive Pregnancy Safety Study and a Clinical Lactation Study were recommended as PMRs.

Please refer to Section 23 for key labeling changes and Section 24 for postmarketing requirements.

7.4. FDA Approach to the Safety Review

The clinical development program for zoliflodacin included eight clinical studies, including one phase 3, randomized, open-label, active-controlled trial and one randomized, open-label, active-controlled phase 2 trial assessing efficacy and safety. The safety review was primarily focused on the data from the phase 3 trial (STI_Zoli001).

Safety data from the phase 3 and phase 2 trials were reviewed separately due to differences in study design and differences in the zoliflodacin formulation administered to participants. Please refer to section 6.2.2 and 6.2.3 for information regarding study design and section 14.2.1 for additional information regarding the zoliflodacin formulations. There were six phase 1 studies, in which a variety of zoliflodacin PO doses and formulations ranging from 200 mg to 4 g were

administered to healthy volunteers. See section 17.1 for additional information about the completed phase 1 studies.

No major data quality or integrity issues were identified that would have precluded performing a safety review for this NDA. Coding of verbatim terms to the Medical Dictionary for Regulatory Activities preferred terms (PTs) was acceptable. TEAEs were protocol-defined by the Applicant as an adverse event that had a start date on or after the first dose date of the study drug, or an AE that had a start date before the date of the first dose of the study drug but that worsened in severity on or after the first dose of study drug until 30 days after administration. All AEs in the reviewed trials were graded using Common Terminology Criteria for Adverse Events version 5.[22]

The clinical safety data scientist performed standard safety analyses including Office of New Drugs custom medical queries (OCMQs) to identify safety signals.

7.5. Adequacy of the Clinical Safety Database

The safety database is adequate. Trial STI_Zoli001 included 619 participants who received the single 3-g dose of zoliflodacin for the treatment of uncomplicated gonorrhea. Trial DMID 14-0014 added an additional 139 participants who received either 2 g (n=72) or 3 g (n=67) of zoliflodacin for the treatment of uncomplicated gonorrhea.

Additional safety data are available from 237 healthy participants from the six phase 1 clinical studies who received oral formulations of zoliflodacin (Table 25).

The numbers of participants, dosing, and duration of exposure in Trials STI_Zoli001 and DMID 14-0014 are sufficient to conduct a safety review.

Table 25. Zoliflodacin Safety Database

Clinical Trial/Study Groups N=1343	Phase of Trial/Study	Zoliflodacin N=995	Active Control N=348
STI_Zoli001*	3	619	308^
DMID 14-0014**	2	139	40^^
D4930C00001 (Entasis)**	1	54	N/A
D4930C00003 (Entasis)**	1	6	N/A
DMID 16-0110 (NIAID)*	1	71	N/A
DMID 16-0118 (NIAID)*	1	8	N/A
STI_Zoli002 (GARDP)*	1	48	N/A
STI_Zoli003 (GARDP)*	1	50	N/A

Source: Reviewer Table
*Granules for oral suspension.
**Powder for oral suspension.
^Active comparator was IM ceftriaxone and oral azithromycin.
^^Active comparator was IM ceftriaxone.
Abbreviations: GARDP, Global Antibiotic Research and Development Partnership; IM, intramuscular; N, number of participants in treatment arm; N/A, not applicable; NIAID, National Institute of Allergy and Infectious Diseases

Baseline Demographic and Clinical Characteristics

In Trial STI_Zoli001, baseline demographic and clinical characteristics were similar between treatment arms (Table 26). The majority of participants in both treatment arms were male

[22] CTEP, Common Terminology Criteria for Adverse Events (CTCAE), Version 5.0, Place Published: Cancer Therapy Evaluation Program. https://dctd.cancer.gov/research/ctep-trials/for-sites/adverse-events/ctcae-v5-5x7.pdf.

(roughly 87%). Most participants were between 18 to 64 years old (>97%) with the median age in the zoliflodacin arm being 28 years and 26 years in the comparator arm. In both arms, over 50% of participants were Black or African American. In the zoliflodacin arm, 31.2% of participants were Asian and 10.5% were White. In the comparator arm, 29.9% of participants were Asian and 15.3% were White. In the zoliflodacin arm, 44.7% of participants lived in South Africa, 29.2% of participants lived in Thailand, 17.3% of participants lived in the United States, and 8.7% of participants lived in the European Union. The proportions of enrolled participants in the comparator arm were similar.

Table 26. Baseline Demographic and Clinical Characteristics, Safety Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=619	Ceftriaxone + Azithromycin N=308
Sex, n (%)		
Male	542 (87.6)	270 (87.7)
Female	77 (12.4)	38 (12.3)
Age, years		
Mean (SD)	30 (9.6)	29.1 (9.1)
Median (min, max)	28 (16, 73)	26 (15, 67)
Age group, years, n (%)		
<18	12 (1.9)	2 (0.6)
18 to 64	604 (97.6)	305 (99.0)
≥65	3 (0.5)	1 (0.3)
Age group ≥75, years, n (%)		
≥75	0	0
Race, n (%)		
Black or African American	348 (56.2)	164 (53.2)
Asian	193 (31.2)	92 (29.9)
White	65 (10.5)	47 (15.3)
American Indian or Alaska Native	8 (1.3)	1 (0.3)
Multiple	1 (0.2)	2 (0.6)
Native Hawaiian or Other Pacific Islander	2 (0.3)	1 (0.3)
Other	2 (0.3)	1 (0.3)
Ethnicity, n (%)		
Not Hispanic or Latino	601 (97.1)	295 (95.8)
Hispanic or Latino	18 (2.9)	13 (4.2)
Country of participation, n (%)		
South Africa	277 (44.7)	145 (47.1)
Thailand	181 (29.2)	89 (28.9)
United States	107 (17.3)	51 (16.6)
Others	54 (8.7)	23 (7.5)
Is in United States, n (%)		
Not in United States	512 (82.7)	257 (83.4)
United States	107 (17.3)	51 (16.6)

Source: adsl.xpt; Software: R
Abbreviations: max, maximum; min, minimum; N, number of participants in treatment arm; n, number of participants with given characteristic; SD, standard deviation

7.6. Safety Results

7.6.1. Safety Results, Trial STI_Zoli001

7.6.1.1. Overview of Treatment-Emergent Adverse Events, Trial STI_Zoli001

As shown in Table 27, the overall incidence of TEAEs was equal between both treatment arms. Most TEAEs were of mild or moderate severity. There were 21 participants (3.4%) in the zoliflodacin arm and 18 participants (5.8%) in the ceftriaxone-azithromycin arm who experienced severe AEs. There were no SAEs reported in either treatment arm, neither were there AEs that led to discontinuation, modification, or reduction of the study drug.

Table 27. Overview of Treatment-Emergent Adverse Events[1], Safety Population, Trial STI_Zoli001

Event Category	Zoliflodacin N=619 n (%)	Ceftriaxone + Azithromycin N=308 n (%)
SAE	0	0
Any AE	286 (46.2)	143 (46.4)
Severe and worse	21 (3.4)	18 (5.8)
Moderate	109 (17.6)	43 (14.0)
Mild	156 (25.2)	82 (26.6)

Source: adae.xpt; Software: R
[1] Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Severity as assessed by the investigator.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants with at least one event; SAE, serious adverse event

7.6.1.2. Deaths, Trial STI_Zoli001

There were no deaths in Trial STI_Zoli001.

7.6.1.3. Serious Treatment-Emergent Adverse Events, Trial STI_Zoli001

There were no SAEs in Trial STI_Zoli001.

7.6.1.4. Adverse Events and OCMQs Leading to Treatment Discontinuation, Trial STI_Zoli001

Treatment discontinuation due to AEs was not applicable in the zoliflodacin arm given that the treatment consisted of a single dose. However, there were also no participants who discontinued treatment early in the comparator arm.

7.6.1.5. Treatment-Emergent Adverse Events, Trial STI_Zoli001

TEAEs occurred in 46.2% of participants in the zoliflodacin arm and 46.4% of participants in the ceftriaxone-azithromycin arm (Table 28). Low neutrophil cell count, headache, and low white blood cell count were the three most commonly reported TEAEs in the zoliflodacin arm. However, the comparator arm had a higher percentage of participants with low neutrophil counts compared to zoliflodacin. Additionally, diarrhea was reported at a higher frequency in the ceftriaxone-azithromycin arm (7.1%) when compared to the zoliflodacin arm (2.4%).

Table 28. Participants With Common Treatment-Emergent Adverse Events[1] Occurring at ≥1% Frequency in the Zoliflodacin Arm, Safety Population, Trial STI_Zoli001

Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone + Azithromycin N=308 n (%)
Any AE	286 (46.2)	143 (46.4)
Low neutrophil cell counts	63 (10.1)	39 (12.7)
Headache	61 (9.9)	15 (4.9)
Low white blood cell count	30 (4.8)	9 (2.9)
Dizziness	21 (3.4)	5 (1.6)
Nausea	16 (2.6)	12 (3.9)
Diarrhea	15 (2.4)	22 (7.1)
Alanine aminotransferase increased	12 (1.9)	5 (1.6)
Renal impairment	12 (1.9)	4 (1.3)
Abdominal pain	9 (1.5)	3 (1.0)
Malaise	8 (1.3)	5 (1.6)
Hyperbilirubinemia	8 (1.3)	2 (0.6)
Oropharyngeal pain	6 (1.0)	1 (0.3)
Pyrexia	6 (1.0)	4 (1.3)

Source: adae.xpt; Software: R, MS Excel
[1] Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Definition of preferred terms: low neutrophil cell count includes neutropenia and neutrophil count decreased; headache includes headache and tension headache; low white blood cell count includes leukopenia and white blood cell count decreased; renal impairment includes renal impairment, creatinine renal clearance decreased, blood creatinine increased, glomerular filtration rate decreased; abdominal pain includes abdominal pain, lower abdominal pain, upper abdominal pain; hyperbilirubinemia includes blood bilirubin increased, conjugated bilirubin increased, hyperbilirubinemia.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Coded as MedDRA version 26.0 preferred terms.
Abbreviations: AE, adverse event; MedDRA, Medical Dictionary for Regulatory Activities; N, number of participants in treatment arm; n, number of participants with adverse event

Low Neutrophil Cell Count

The number of participants with investigator-classified PTs of low neutrophil cell counts was higher in the comparator arm (39/308; 12.7%), compared to the zoliflodacin arm (63/619; 10.1%). Based on the Common Terminology Criteria for Adverse Events, in the zoliflodacin arm, 1.8% of participants had mildly low neutrophil cell counts (1.5 to 2.0×10^9 cells/L), 5.3% of participants had moderately low neutrophil cell counts (1.0 to 1.5×10^9 cells/L), 2.9% of participants developed severely low neutrophil cell counts (0.5 to 1.0×10^9 cells/L) and 0.2% of participants developed life threateningly low neutrophil cell count ($<0.5\times10^9$ cells/L). At the time of trial completion, 6.3% of participants had either resolving or complete resolution of low neutrophil count, while 3.9% of participants had ongoing low neutrophil cell counts. No

participants required treatment. HIV status did not correlate with the occurrence of low neutrophil count.

FDA analysis noted differences between the investigator-classified PTs for low neutrophil cell counts and the objective laboratory measures of neutrophil cell counts. More participants were found to have conventionally-defined neutropenia than low neutrophil counts classified by investigators as TEAEs. An information request was sent to the Applicant for clarification of this finding. The Applicant noted that the PT of low neutrophil count was reported as an AE if the investigator determined it to be clinically significant and with clinical consequence rather than all out-of-range laboratory values. Further analysis of observed changes in laboratory values is discussed in section 7.6.1.6.

Headache

Headache was more common in the zoliflodacin arm, occurring in 9.9% of participants, compared to 4.9% in the comparator arm. In the zoliflodacin arm, most headaches were considered mild (7.6%) or moderate (2.3%). In the comparator arm, most headaches were mild (2.9%) or moderate (1.6%). In the zoliflodacin arm, all headaches resolved by EOS with 3.1% requiring treatment. Given the short half-life of zoliflodacin (~ 6 hours), it is unlikely that headaches occurring at or after TOC were related to study drug.

Low White Blood Cell Count

The incidence of low white blood cell (WBC) count was higher in the zoliflodacin arm, occurring in 4.8% of participants compared to 2.9% in the comparator arm. There were 18 participants who developed low WBC count within 8 days of zoliflodacin administration and 12 participants who developed low WBC count at end-of-study (Day 27 to Day 31). Most cases of low WBC count were mild or moderate in severity; however, one participant did have a severe adverse event (grade 3) of low WBC count in the zoliflodacin arm (0.2%). The participant's low WBC count was noted on follow-up Visit 5 (Day 27). This participant did not receive any interventions.

Similar differences were noted between investigator reported PTs for low WBC count and objective laboratory values for WBC counts. More participants were found to have low WBC counts based on laboratory values than those reported by investigators as TEAEs. See Section 7.6.1.6 for further analysis of observed changes in laboratory values.

Dizziness

The number of participants who reported dizziness was higher in the zoliflodacin arm (3.4%) than the ceftriaxone-azithromycin arm (1.9%). All cases were reported as mild.

As shown in Table 29, more participants in the zoliflodacin arm had TEAEs grouped as OCMQs in the following system organ classes (SOCs): Blood and lymphatic systems disorders, General disorders and administration site conditions, Hepatobiliary disorders, Infections and infestations, Nervous system disorders and Renal and urinary disorders. In the Blood and lymphatic SOC, there was a higher frequency of leukopenia (4.8%) in the zoliflodacin arm compared to the ceftriaxone-azithromycin arm (2.9%). Dizziness (General disorders and administrative site conditions SOC) and hepatic injury (Hepatobiliary disorders SOC) occurred more frequently in the zoliflodacin arm (3.4% and 2.6%, respectively). In the Infections and infestations SOC, there

were higher frequencies of bacterial infection (2.4%) and viral infection (1.8%) in the zoliflodacin arm. Headache (Nervous system disorders SOC) occurred more frequently in the zoliflodacin arm (9.9%) compared to the ceftriaxone-azithromycin arm (4.9%). TEAEs from the Renal and urinary tract infections (Renal and urinary disorders) SOC were only reported in the zoliflodacin arm (1.3%). See Table 129 in section 17 for a complete list of all TEAEs reported during the trial period.

Table 29. Participants With Adverse Events by System Organ Class and OCMQ (Narrow), Safety Population, Trial STI_Zoli001

System Organ Class OCMQ (Narrow)	Zoliflodacin N=619 n (%)	Ceftriaxone + Azithromycin N=308 n (%)
Blood and lymphatic system disorders (SOC)		
Leukopenia	30 (4.8)	9 (2.9)
Anemia	3 (0.5)	0
Thrombocytopenia	3 (0.5)	2 (0.6)
Cardiac disorders (SOC)		
Palpitations	1 (0.2)	0
Systemic hypertension	1 (0.2)	3 (1.0)
Gastrointestinal disorders (SOC)		
Nausea	16 (2.6)	12 (3.9)
Diarrhea	15 (2.4)	22 (7.1)
Abdominal pain	9 (1.5)	3 (1.0)
Vomiting	3 (0.5)	1 (0.3)
Constipation	1 (0.2)	2 (0.6)
Dyspepsia	1 (0.2)	3 (1.0)
General disorders and administration site conditions (SOC)		
Dizziness	21 (3.4)	6 (1.9)
Fatigue	14 (2.3)	9 (2.9)
Local administration reaction	8 (1.3)	38 (12.3)
Pyrexia	6 (1.0)	4 (1.3)
Decreased appetite	0	1 (0.3)
Hepatobiliary disorders (SOC)		
Hepatic injury	16 (2.6)	5 (1.6)
Infections and infestations (SOC)		
Bacterial infection	15 (2.4)	1 (0.3)
Fungal infection	11 (1.8)	3 (1.0)
Viral infection	11 (1.8)	2 (0.6)
Nasopharyngitis	7 (1.1)	2 (0.6)
Purulent material	2 (0.3)	0
Metabolism and nutrition disorders (SOC)		
Cachexia	0	3 (1.0)
Musculoskeletal and connective tissue disorders (SOC)		
Back pain	3 (0.5)	2 (0.6)
Arthralgia	2 (0.3)	0
Myalgia	2 (0.3)	1 (0.3)
Nervous system disorders (SOC)		
Headache	61 (9.9)	15 (4.9)
Somnolence	3 (0.5)	2 (0.6)
Paresthesia	1 (0.2)	0
Psychiatric disorders (SOC)		

System Organ Class OCMQ (Narrow)	Zoliflodacin N=619 n (%)	Ceftriaxone + Azithromycin N=308 n (%)
Insomnia	1 (0.2)	4 (1.3)
Study agent abuse potential	1 (0.2)	1 (0.3)
Anxiety	0	1 (0.3)
Depression	0	2 (0.6)
Self-harm	0	1 (0.3)
Renal and urinary disorders (SOC)		
Renal & urinary tract infection	8 (1.3)	0
Reproductive system and breast disorders (SOC)		
Abnormal uterine bleeding	3 (0.5)	0
Bacterial vaginosis	1 (0.2)	1 (0.3)
Respiratory, thoracic and mediastinal disorders (SOC)		
Cough	1 (0.2)	3 (1.0)
Skin and subcutaneous tissue disorders (SOC)		
Pruritus	8 (1.3)	3 (1.0)
Rash	2 (0.3)	0
Alopecia	1 (0.2)	0
Vascular disorders (SOC)		
Hemorrhage	5 (0.8)	0
Hypotension	0	1 (0.3)

Source: adae.xpt; Software: R
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Each OCMQ is aligned to a single SOC based on clinical judgment. However, please be aware that some OCMQs may contain PTs from more than one SOC.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants with adverse event; OCMQ, OND custom medical query; OND, Office of New Drugs; PT, preferred term; SOC, system organ class

Bacterial Infection

The incidence of bacterial infections was higher in the zoliflodacin arm, occurring in 2.4% of participants compared to 0.3% in the ceftriaxone-azithromycin arm. In the zoliflodacin treatment arm, all cases of bacterial infection were either mild (1.1%) or moderate (1.3%) in severity. The majority of cases resolved. In total, there were 5 participants with unresolved bacterial infections. Of these participants, 4 of them were newly diagnosed with an infection at the EOS; 2 with chlamydia urethritis, 1 with rectal gonorrhea, and 1 with gonococcal urethritis. One individual had ongoing syphilis which was diagnosed on day 14 and not resolved by EOS.

Viral Infection

The number of participants who reported viral infection was higher in the zoliflodacin arm (1.8%) than the comparator arm (0.6%). All cases were either mild or moderate.

Hypersensitivity

In the phase 3 trial, hypersensitivity reactions were reported in 3 participants (0.3%) and included dermatitis allergic, rash, and swelling of the eyelid; all were reported in one participant each (0.2%) and all were in the zoliflodacin arm. All TEAEs were nonserious, mild, or moderate (Grade 1 or 2) in intensity. One TEAE of rash occurred on Day 1 of the treatment and was considered related to study drug. The participant received treatment and the rash resolved by Day 30 (Participant (b) (6)). The TEAE of swelling of eyelid occurred on Day 1 of the

treatment and resolved on Day 2; the investigator considered it unrelated to the study drug (Participant (b) (6)). The third TEAE of dermatitis allergic occurred on Day 20 of the treatment and was considered not related to study drug. Recovery from this TEAE was not reported.

7.6.1.5.1. Analysis of Treatment-Emergent Adverse Events Known To Be Associated With Fluoroquinolones and Gepotidacin

Zoliflodacin is a novel, first-in-class antibacterial drug that inhibits bacterial DNA replication through inhibition of type II topoisomerase (DNA gyrase and topoisomerase IV). Zoliflodacin is structurally different from fluoroquinolones and gepotidacin. While all three drug classes target type II topoisomerase, zoliflodacin binds to a different target subunit (*GyrB*) than fluoroquinolones or gepotidacin, both of which bind to *GyrA*. However, due to common inhibition of the type II topoisomerase, two exploratory analyses were conducted to evaluate for the occurrence in Trial STI_Zoli001 of specific TEAEs associated with fluoroquinolones and gepotidacin.

In the review of AEs associated with fluoroquinolone use, the PTs analyzed included tendinitis, tendon rupture, peripheral neuropathy, central nervous system effects, aortic aneurysm/dissection, hepatotoxicity, photosensitivity, crystalluria and symptomatic glucose alterations. In total, 13.4% of zoliflodacin participants and 8.8% of ceftriaxone-azithromycin participants had TEAEs that have been associated with fluoroquinolone use. The most common fluoroquinolone-associated TEAEs in the zoliflodacin arm compared to the ceftriaxone-azithromycin arm were headaches (9.9% versus 4.9%, respectively) and dizziness (3.4% versus 1.6%, respectively). The following TEAEs occurred in <0.5% of participants in both arms: depression, myalgia, arthralgia, asthenia, presyncope and hypoasthesia. No cases were reported of tendinitis, tendon rupture, aortic aneurysm/dissection, photosensitivity, crystalluria or symptomatic glucose alterations in trial participants. Based on the available data, zoliflodacin does have mild central nervous system effects, specifically headache and dizziness, but does not appear to share other common TEAEs associated with fluoroquinolone drugs.

For the gepotidacin-related AE analysis, the following PTs were evaluated: diarrhea, nausea, abdominal pain, flatulence, soft feces, dysarthria, presyncope, muscle spasms, vomiting, hypersalivation, and hyperhidrosis. These AEs have been associated with acetylcholinesterase inhibition and were noted in the registrational trials of gepotidacin for treatment of uncomplicated UTI. In total, 6.8% of zoliflodacin participants and 11.4% of ceftriaxone-azithromycin participants had TEAEs that have been associated with gepotidacin use. The most common gepotidacin-related TEAEs in the zoliflodacin arm compared to the ceftriaxone-azithromycin arm were abdominal pain (1.5% versus 1.0%, respectively) and vomiting (0.5% versus 0.3%, respectively). Additional TEAEs associated with gepotidacin use that were reported in both the zoliflodacin and ceftriaxone-azithromycin treatment arms included: diarrhea (2.4% versus 7.1%, respectively), flatulence (0.5% versus 1.0%, respectively), and nausea (2.6% versus 3.9%, respectively). Presyncope occurred in <0.5% of trial participants in either arm. There were no cases of muscle spasms, dysarthria, soft feces, hypersalivation or hyperhidrosis reported in the phase 3 trial for zoliflodacin. Based on the available data, zoliflodacin does not appear to share

common TEAEs associated with gepotidacin administration, specifically, TEAEs that may be due to acetylcholinesterase inhibition.

7.6.1.6. Laboratory Findings, Trial STI_Zoli001

Laboratory values collected in Trial STI_Zoli001 included alanine aminotransferase (ALT), total bilirubin, creatinine, white blood cell count, red blood cell count, hemoglobin, hematocrit, platelets, neutrophil cell count, monocyte cell count, lymphocyte blood cell count, eosinophil cell count, basophil cell count, and mean corpuscular volume. Direct bilirubin was only collected if total bilirubin was elevated. The estimated glomerular filtration rate (eGFR) was calculated using the chronic kidney disease-epidemiology collaboration (CKD-EPI) formula.

In Trial STI_Zoli001, renal and hematology laboratory values between the two treatment arms were similar. As shown in Table 30, the percentage of participants who developed Level 1 elevations in creatinine was the same in both treatment arms (0.3%). No participants developed Level 2 or Level 3 elevations in creatinine. The percentage of participants who developed a Level 1 decrease in estimated glomerular filtration rate was slightly higher in the zoliflodacin arm (1.6%) compared to control (1.3%). No participants developed Level 2 or Level 3 decreases in estimated glomerular filtration rate.

Table 30. Participants With One or More Kidney Function Analyte Values Exceeding Specified Levels, Safety Population, Trial STI_Zoli001

Laboratory Parameter	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Creatinine, high (mg/dL)		
Level 1 (≥1.5× Baseline)	2/610 (0.3)	1/301 (0.3)
Level 2 (≥2× Baseline)	0/610 (0)	0/301 (0)
Level 3 (≥3× Baseline)	0/610 (0)	0/301 (0)
eGFR, low (mL/min/1.73 m^2)		
Level 1 (≥25% decrease)	10/610 (1.6)	4/301 (1.3)
Level 2 (≥50% decrease)	0/610 (0)	0/301 (0)
Level 3 (≥75% decrease)	0/610 (0)	0/301 (0)

Source: adlb.xpt; Software: R
Threshold levels 1, 2, and 3 as defined by the Standard Safety Tables & Figures Integrated Guide.
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
eGFR values are calculated from serum creatinine using chronic kidney disease epidemiology collaboration (CKD-EPI) equation.
Abbreviations: AE, adverse event; eGFR, estimated glomerular filtration rate; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data

Variations in hematological values were similar in both treatment arms. The percentage of participants with Level 1 ($<3.5\times10^3$ cells/µL) low WBC count was higher in the ceftriaxone-azithromycin arm (12.6%) compared to the zoliflodacin arm (10.3%) The comparator arm also had a higher percentage of participants with Level 2 ($<3.0\times10^3$ cells/µL) low WBC count versus zoliflodacin (5.3% and 4.1%, respectively). Almost a third of participants in both the zoliflodacin and ceftriaxone-azithromycin arms had low neutrophil counts. A total of 27.9% of participants in the zoliflodacin arm and 31.2% of participants in the comparator arm had Level 1 ($<2.0\times10^3$ cells/µL) low neutrophil counts. More participants in the ceftriaxone-azithromycin

arm (5.0%) had Level 2 (<1.0×10^3 cells/µL) low neutrophil counts compared to the zoliflodacin arm (3.8%). There was only one participant in the zoliflodacin arm who had a Level 3 (<0.5×10^3 cells/µL) low neutrophil count. There were no participants in the ceftriaxone-azithromycin arm that had a Level 3 low neutrophil counts. As noted in section 7.6.1.5, these laboratory values are different from the investigator reported AEs.

Table 31. Participants With One or More Hematology Analyte Values Exceeding Specified Levels, Safety Population, Trial STI_Zoli001

Laboratory Parameter	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Complete blood count		
WBC, low (10^3 cells/µL)		
Level 1 (<3.5)	63/610 (10.3)	38/301 (12.6)
Level 2 (<3)	25/610 (4.1)	16/301 (5.3)
Level 3 (<1)	0/610 (0)	0/301 (0)
WBC, high (10^3 cells/µL)		
Level 1 (>10.8)	18/610 (3.0)	6/301 (2.0)
Level 2 (>13)	4/610 (0.7)	0/301 (0)
Level 3 (>15)	1/610 (0.2)	0/301 (0)
Hemoglobin, low (g/dL)		
Level 2 (>1.5 g/dL dec. from Baseline)	21/610 (3.4)	9/301 (3.0)
Level 3 (>2 g/dL dec. from Baseline)	4/610 (0.7)	0/301 (0)
Hemoglobin, high (g/dL)		
Level 2 (>2 g/dL inc. from Baseline)	0/610 (0)	3/301 (1.0)
Level 3 (>3 g/dL inc. from Baseline)	0/610 (0)	0/301 (0)
Platelets, low (10^3 cells/µL)		
Level 1 (<140)	10/610 (1.6)	3/301 (1.0)
Level 2 (<125)	6/610 (1.0)	2/301 (0.7)
Level 3 (<100)	0/610 (0)	2/301 (0.7)
WBC differential		
Lymphocytes, low (10^3 cells/µL)		
Level 1 (<1)	16/610 (2.6)	14/301 (4.7)
Level 2 (<0.75)	4/610 (0.7)	3/301 (1.0)
Level 3 (<0.5)	0/610 (0)	0/301 (0)
Lymphocytes, high (10^3 cells/µL)		
Level 1 (>4)	7/610 (1.1)	4/301 (1.3)
Level 2 (>10)	0/610 (0)	0/301 (0)
Level 3 (>20)	0/610 (0)	0/301 (0)
Neutrophils, low (10^3 cells/µL)		
Level 1 (<2)	170/610 (27.9)	94/301 (31.2)
Level 2 (<1)	23/610 (3.8)	15/301 (5.0)
Level 3 (<0.5)	1/610 (0.2)	0/301 (0)

Laboratory Parameter	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Eosinophils, high (10^3 cells/µL)		
Level 1 (>0.65)	27/610 (4.4)	14/301 (4.7)
Level 2 (>1.5)	3/610 (0.5)	0/301 (0)
Level 3 (>5)	0/610 (0)	0/301 (0)

Source: adlb.xpt; Software: R
Threshold levels 1, 2, and 3 as defined by the Standard Safety Tables & Figures Integrated Guide.
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
Abbreviations: AE, adverse event; dec., decrease; inc., increase; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data; WBC, white blood cell

Based on the Applicant's definition of low neutrophil cell count (<2000 cells/µL), at Baseline, there were 44 participants (7.1%) that had a low neutrophil count <2000 to1000 cells/µL in the zoliflodacin arm and 26 participants (8.4%) in the comparator arm. In total, there were 161 participants (26.0%) in the zoliflodacin arm and 90 participants (29.2%) in the ceftriaxone arm who had low neutrophil counts during the safety monitoring period.

A subgroup analysis of participants with low neutrophil counts as defined by the Applicant (<2000 cells/µL) was conducted and is described below. As shown in Table 32, the demographics were similar between the treatment arms.

Table 32. Demographic Information of Participants With Low Neutrophil Count Values (<2000 Cells/µL), Safety Population, Trial STI_Zoli001

Demographics	Zoliflodacin N=161 n (%)	Ceftriaxone + Azithromycin N=90 n (%)
Race		
Black or African American	142 (88.1)	75 (83.3)
Asian	13 (8.1)	9 (10.0)
White	5 (3.1)	6 (6.7)
Other	1 (0.6)	0 (0)
HIV status		
Negative	116 (72.0)	66 (73.3)
Positive	34 (21.1)	20 (22.2)
Missing	11 (6.8)	4 (4.4)
Age		
<18 years	3 (1.9)	1 (1.1)
18-65	158 (98.1)	89 (98.9)
>65	0 (0)	0 (0)
Country		
South Africa	125 (77.6)	67 (74.4)
United States	20 (12.4)	11 (12.2)
Thailand	12 (7.5)	8 (8.9)
European Union	4 (2.5)	4 (4.4)
Sex		
Male	146 (90.7)	84 (93.3)
Female	15 (9.3)	6 (6.7)

Lowest neutrophil count recorded		
<2000-1500 cells/µL	80 (49.7)	43 (47.8)
<1500-1000 cells/µL	60 (37.3)	33 (36.7)
<1000-500 cells/µL	20 (12.4)	14 (15.5)
<500 cells/µL	1 (0.6)	0 (0)

Source: adlb.xpt and admh.xpt; Software: MS Excel
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
Abbreviations: AE, adverse event; HIV, human immunodeficiency virus; N, number of participants in treatment arm; n, number of participants with characteristic

The majority of participants were Black or African American in both the zoliflodacin and ceftriaxone-azithromycin arms (88.1% versus 83.3%, respectively). Most participants were HIV negative and 18 to 65 years of age. In both the zoliflodacin and comparator arms, participants were predominantly from South Africa (77.6% versus 74.4%, respectively). A similar percentage of participants had low neutrophil count values in both arms. In the zoliflodacin arm 49.7% of participants had neutrophil counts <2000 to1500 cells/µL, 37.7% of participants had neutrophil counts <1500 to1000 cells/µL, 12.4% of participants had neutrophil counts to <1000 to 500 cells/µL and 0.6% of participants had neutrophil counts <500 cells/µL. In the ceftriaxone-azithromycin arm, 47.8% of participants had neutrophil counts <2000 to 1500 cells/µL, 36.7% of participants had neutrophil counts <1500 to 1000 cells/µL, 15.5% of participants had neutrophil counts to <1000 to 500 cells/µL and no participants had neutrophil counts <500 cells/µL.

In the zoliflodacin arm, Participant (b) (6) had a neutrophil count <500 cells/µL. This participant was a 25-year-old male who was living with HIV. At his Baseline visit, his neutrophil count was 2660 cells/µL. On Day 7, his neutrophil count dropped to 1090 cells/µL and on Day 36 his neutrophil count decreased further to 380 cells/µL. In addition to a low neutrophil count, this participant also had a low platelet count. The participant was taking tenofovir, lamivudine, and dolutegravir. The healthcare provider thought the drop in neutrophil count could be due to HIV-associated impaired hemopoiesis; however, the participant declined further follow-up care. Given the timing of neutropenia (Day 7), and the $t_{1/2}$ of zoliflodacin (5.5 hours in the fed state), causality is unlikely.

7.6.1.6.1. Additional Analysis of Neutropenia in Participants with Normal Baseline Values

This reviewer conducted a subanalysis utilizing the conventional definition of neutropenia, i.e., neutrophil count <1500 cells/µL and evaluating its occurrence in participants who had normal neutrophil counts at Baseline. Of note, there were a total of 10 participants (1.7%) in the zoliflodacin arm and four participants (1.4%) in the ceftriaxone-azithromycin arm who had mild neutropenia (<1500 to 1000 cells/µL) at Baseline and were excluded from this analysis. No participants had moderate (<1000 to 500 cells/ µL) or severe (<500 cells/µL) neutropenia at baseline in either treatment arm.

Table 33 highlights the change in neutrophil cell counts over the course of the trial in participants with a normal neutrophil count at Baseline.

Table 33. Neutrophil Cell Levels at Baseline, TOC and EOS, Safety Population, Trial STI_Zoli001

Timepoint	Postbaseline Level	Zoliflodacin Baseline Normal	CTX-AZM Baseline Normal
Visit 4 (TOC)	Severe (<500 cells/µL)	0/597 (0.0)	0/296 (0.0)
Visit 4 (TOC)	Moderate (<1000-500 cells/µL)	2/597 (0.3)	5/296 (1.7)
Visit 4 (TOC)	Mild (<1500-1000 cells/µL)	39/597 (6.5)	26/296 (8.8)
Visit 4 (TOC)	Normal (≥1500 cells/µL)	546/597 (91.5)	261/296 (88.2)
Visit 4 (TOC)	Total	587/597 (98.3)	292/296 (98.6)
Visit 5 (EOS)	Severe (<500 cells/µL)	1/563 (0.2)	0/282 (0.0)
Visit 5 (EOS)	Moderate (<1000-500 cells/µL)	12/563 (2.1)	10/282 (3.5)
Visit 5 (EOS)	Mild (<1500-1000 cells/µL)	40/563 (7.1)	18/282 (6.4)
Visit 5 (EOS)	Normal (≥1500 cells/µL)	500/563 (88.8)	250/282 (88.7)
Visit 5 (EOS)	Total	553/563 (98.2)	278/282 (98.6)

Source: adlb.xpt; Source: MS Excel, R
Abbreviations: AZM, azithromycin; CTX, ceftriaxone; EOS, end-of-study; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data (excluding missing data); TOC, test-of-cure

In the zoliflodacin arm at the TOC Visit (Visit 4; Day 6±2), 39 (6.5%) participants had mild neutropenia (<1500 to 1000 cells/µL), and two (0.3%) participants had moderate neutropenia (<1000 to 500 cells/µL). At the EOS Visit (Visit 5; Day 30±3), 40 (7.1%) participants in the zoliflodacin arm had mild neutropenia (<1500 to 1000 cells/µL), 12 (2.1%) participants had moderate neutropenia (<1500 to 1000 cells/µL) and one (0.2%) participant had severe neutropenia (<500 cells/µL).

In the ceftriaxone-azithromycin arm at the TOC Visit, 26 (8.8%) participants had mild neutropenia (<1500 to 1000 cells/µL) and five (1.7%) participants had moderate neutropenia (<1000 to 500 cells/µL). At the EOS Visit, 18 (6.4%) participants in the ceftriaxone-azithromycin arm had mild neutropenia (<1500 to 1000 cells/µL), and 10 participants (3.5%) had moderate neutropenia (<1500 to 1000 cells/µL). No participants in the ceftriaxone-azithromycin arm had severe neutropenia.

Overall, the majority of participants who experienced a reduction in the number of neutrophils from a normal baseline were African or of African American descent. Based on the medical literature, individuals of African, African American descent, or Middle Eastern ancestry are known to have a higher frequency of Duffy-null Associated Neutrophil Count (DANC), formally known as benign ethnic neutropenia. This is a normal occurrence in this population and is characterized by persistent neutropenia. Atallah-Yunes et al. estimates that 25% to 50% of Africans may have DANC.[23] This finding is associated with a homozygous variant in the atypical chemokine receptor 1 (ACKR1) gene, which prevents transcription of ACKR1 in red blood cells,

[23] Atallah-Yunes SA, Ready A, Newburger PE. Benign ethnic neutropenia. Blood Rev. 2019 Sep;37:100586. doi: 10.1016/j.blre.2019.06.003. Epub 2019 Jun 21. PMID: 31255364; PMCID: PMC6702066.

which is the Duffy blood group. This genotype may have arisen due to selective pressure, as the Duffy-null genotype provides some resistance to infection with *Plasmodium vivax*.[24,25] Despite low neutrophil counts, individuals with DANC typically do not have increased rates of bacterial infections.[26]

In Trial STI_Zoli001, the frequency of lower neutrophil counts was similar in both the zoliflodacin arm and the control arm. As stated previously, the majority of participants were Black or African American and were from South Africa. Although the presence of the Duffy-null genotype was not tested in this trial, the predominance of African and African-American participants and the similar distribution of neutropenia between treatment arms suggests that this finding could be due to DANC. The majority of participants with low neutrophil values in both treatment arms were HIV-negative. There was no correlation between HIV status and low neutrophil cell count. Of note, nonclinical data showed variability in these laboratory values, but no consistent pattern emerged to suggest a potential safety signal. Finally, given the short half-life of zoliflodacin (~6 hours), it was considered unlikely that decreases in total white cell or neutrophil counts at TOC or later were related to zoliflodacin administration.

7.6.1.7. Assessment of Drug-Induced Liver Injury, Trial STI_Zoli001

There were no significant differences between the two treatment arms in the proportions of participants with abnormal liver chemistry values (ALT, total bilirubin). Alkaline phosphatase and aspartate aminotransferase values were not evaluated in this trial. One participant (0.2%) in the zoliflodacin arm, discussed below, had an ALT >10× upper limit of normal (ULN) compared to none in the ceftriaxone-azithromycin arm. No participants in the zoliflodacin arm had bilirubin levels >3× ULN compared to one (0.3%) participant in the ceftriaxone-azithromycin arm.

Table 34. Participants With One or More Liver Biochemistry Analyte Values Exceeding Specified Levels, Safety Population, Trial STI_Zoli001

Laboratory Parameter	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Alanine aminotransferase, high (U/L)		
Level 1 (>3× ULN)	10/609 (1.6)	4/300 (1.3)
Level 2 (>5× ULN)	4/609 (0.7)	2/300 (0.7)
Level 3 (>10× ULN)	1/609 (0.2)	0/300 (0)

[24] Daniels G. The molecular genetics of blood group polymorphism. Transpl Immunol. 2005 Aug;14(3-4):143-53. doi: 10.1016/j.trim.2005.03.003. Epub 2005 Apr 26. PMID: 15982556.

[25] Howes, R., Patil, A., Piel, F. et al. The global distribution of the Duffy blood group. Nat Commun 2, 266 (2011). https://doi.org/10.1038/ncomms1265

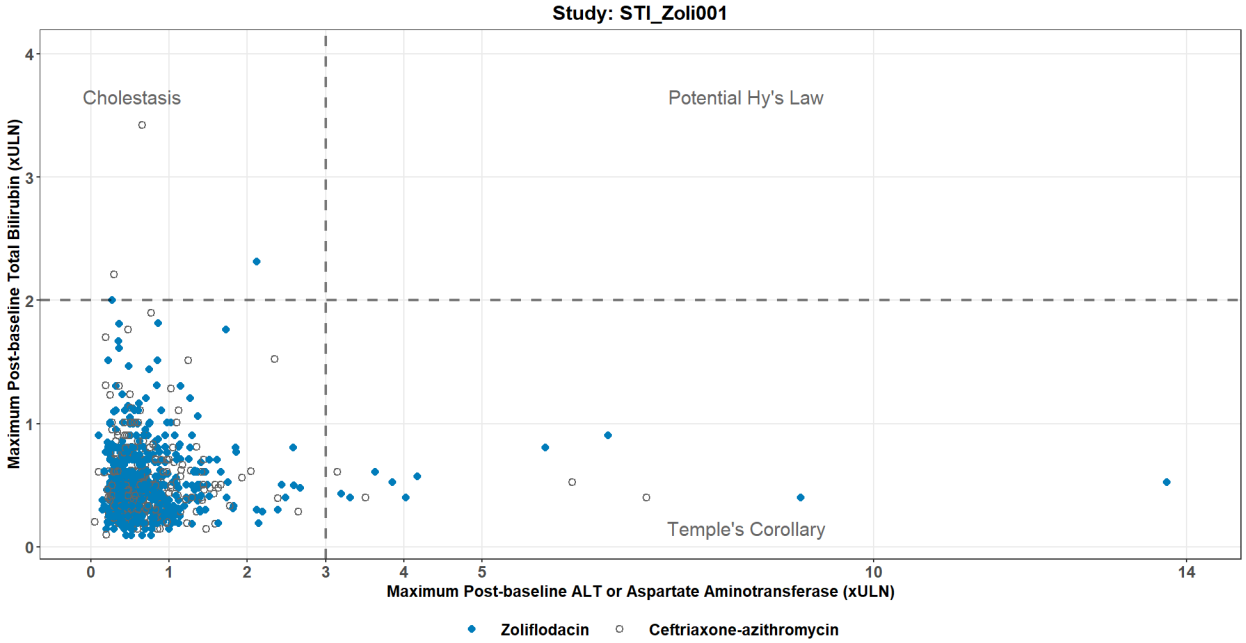
[26] Hysong MR, Shuey MM, Huffman JE and others. 'Characterization of the phenotypic consequences of the Duffy-null genotype'. Blood Advances 2025: volume 9, issue 6, pages 1,452–1462. DOI: 10.1182/bloodadvances.2024014399

Laboratory Parameter	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Bilirubin, total, high (mg/dL)		
Level 1 (>1.5× ULN)	9/609 (1.5)	7/301 (2.3)
Level 2 (>2× ULN)	1/609 (0.2)	2/301 (0.7)
Level 3 (>3× ULN)	0/609 (0)	1/301 (0.3)

Source: adlb.xpt; Software: R
Threshold levels 1, 2, and 3 as defined by the Standard Safety Tables & Figures Integrated Guide.
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data; ULN, upper limit of normal

Figure 3 shows an eDISH plot to identify potential cases of serious drug-induced liver injury. There were no Hy's Law cases in Trial STI_Zoli001 in either arm.

Figure 3. Hepatocellular Drug-Induced Liver Injury Screening Plot, Safety Population, Trial STI_Zoli001



Source: adlb.xpt; Software: R
Each data point represents a participant plotted by their maximum ALT or Aspartate Aminotransferase versus their maximum total bilirubin values in the postbaseline period.
A potential Hy's Law case was defined as having any postbaseline total bilirubin equal to or exceeding 2× ULN after a postbaseline ALT or Aspartate Aminotransferase equal to or exceeding 3× ULN. Those participants who meet total bilirubin equal to or exceeding 2× ULN criteria within 30 days of the ALT or Aspartate Aminotransferase equal to or exceeding 3× ULN criteria are circled in red.
The within 30 days analysis window rule does not apply to cholestasis and temple's corollary cases.
All participants with at least one postbaseline ALT or Aspartate Aminotransferase, bilirubin and ULN are plotted.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
For number of participants in each quadrant, see Table 35.
Abbreviations: ALT, alanine aminotransferase; ULN, upper limit of normal

Table 35. Participants in Each Quadrant for Potential Hepatocellular DILI Screening Plot, Safety Population, Trial STI_Zoli001

Quadrant	Zoliflodacin N=619 n/N_w (%)	Ceftriaxone + Azithromycin N=308 n/N_w (%)
Potential Hy's Law (right upper)	0/609 (0)	0/300 (0)
Cholestasis (left upper)	2/609 (0.3)	2/300 (0.7)
Temple's corollary (right lower)	10/609 (1.6)	4/300 (1.3)
Total	12/609 (2)	6/300 (2)

Source: adlb.xpt; Software: R
A potential Hy's Law case was defined as having any postbaseline total bilirubin equal to or exceeding 2× ULN after a postbaseline ALT or Aspartate Aminotransferase equal to or exceeding 3× ULN.
The within 30 days analysis window rule does not apply to cholestasis and temple's corollary cases.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
Abbreviations: ALT, alanine aminotransferase; DILI, drug-induced liver injury; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data; ULN, upper limit of normal

Although there was no serious DILI in the trial, several participants had post-baseline ALT elevations; relevant details are tabulated in Table 36.

Participants With Elevated Liver Enzymes Postbaseline in Trial STI_Zoli001

Table 36. Clinical Details of Participants With ALT Values >3× ULN and Concomitant Total Bilirubin Values

Fold-Elevation in ALT Above ULN	Participant ID	Age/Sex	HIV Status	Hepatitis Status	Other Med Hx	Baseline ALT (IU/L)	Postbaseline Max ALT (IU/L)	Study Day of Max ALT	Baseline Total Bilirubin (µmol/L)	Total Bilirubin level (µmol/L) at Max ALT	Medications
>3× <5×	(b) (6)	20/M	Neg	Neg	None	13	128	29	7	7	None
		39/F	Pos	Neg	Alcohol-related liver disease, iron deficiency	40	146	15	7	5	Emtricitabine, tenofovir disoproxil fumarate, ferrous sulphate, vitamin C
		37/M	Neg	Neg	None	23	154	56	7	8	None
		27/M	Neg	Neg	None	23	149	8	10.3	8.6	None
		21/M	Neg	Neg	None	122	136	6	6.8	5.1	None
		41/M	Pos	Neg	Hyperlipidemia Hypertension	115	165	4	6.8	6.8	Lamivudine, abacavir, and dolutegravir
>5x <10x		27/M	Neg	Neg	None	45	238	28	5.1	13.7	None
		25/M	Pos	Hep C pos	None	204	271	33	6.8	15.4	Tenofovir, emtricitabine, vitamin C
		28/M	Neg	Hep C pos	None	120	372	29	10.3	6.8	Silymarin marianum
>10x <15x		28/M	Neg	Neg	None	457	550	28	11	11	None

Source: adlb.xpt, admh.xpt, adcm.xpt; Software: MS Excel
Abbreviations: ALT, alanine aminotransferase; F, female; ID, identification; M, male; Max, maximum; Med Hx, medical history; ULN, upper limit of normal

Narratives for Participants in the Zoliflodacin Arm With ALT Values >5× ULN (See Table 36)

1. Participant (b) (6) was a 27-year-old Asian male with a history of thrombocytopenia. ALT was normal at Baseline. At EOS, the participant had ALT elevation >5× ULN and the investigator reported that the participant had consumed a substantial quantity of energy drinks the day preceding the EOS Visit. Additionally, the participant was diagnosed with a coronavirus disease of 2019 (COVID-19) infection 5 days after his EOS Visit. At Day 56, the participant's ALT had normalized. The Applicant attributes the elevation in ALT to a transient fluctuation that could be related either to heavy consumption of energy drinks or early COVID-19 infection. This reviewer agrees that the transient elevation in ALT may be related to the participant's COVID-19 infection, and possibly also to his increased consumption of energy drinks.[27]
2. Participant (b) (6) was a 25-year-old Asian male with a history of HIV and hepatitis C infection. The participant's ALT level was elevated at Baseline and remained elevated during the trial period. The Applicant attributed this finding to the participant's underlying chronic hepatitis C infection. This reviewer agrees with the Applicant's causality assessment for the ALT elevation.
3. Participant (b) (6) was a 28-year-old Asian male who had a new diagnosis of hepatitis C and was taking Samarin (Silybum marianum) daily. The participant's ALT was elevated at Baseline, presumably due to his hepatitis C infection. During the course of the trial, the participant's ALT increased following administration of zoliflodacin and remained above his baseline level. The participant remained asymptomatic. The Applicant attributed the hepatic enzyme fluctuation due to his underlying hepatitis C infection and this reviewer agrees with this assessment.
4. Participant (b) (6) was a 28-year-old Black male who had elevated ALT of unclear etiology at Baseline. He was HIV-negative and had no additional medical history. The participant's ALT increased after receiving zoliflodacin and continued to increase throughout the study period. His baseline ALT was 457 IU/L, which increased to 544 IU/L at TOC (Day 7). At EOS (Day 28), this participant's ALT was 550 IU/L. No additional follow-up visits are documented by the Applicant for this participant. The Applicant attributed these findings to an unknown pre-existing hepatic abnormality. Given the significantly high baseline ALT, this reviewer does not believe that the further increase was related to zoliflodacin. Further, the significant ALT elevation at Baseline should have precluded enrollment of this participant into the trial.

[27] LiverTox: Clinical and Research Information on Drug-Induced Liver Injury [Internet]. Bethesda (MD): National Institute of Diabetes and Digestive and Kidney Diseases; 2012-. Energy Drinks. [Updated 2020 Jun 20]. Available from: https://www.ncbi.nlm.nih.gov/books/NBK559836/

Table 37. Clinical Details of Participants With Total Bilirubin Values >2× ULN and Concomitant ALT Values

Fold-Elevation in Tbili Above ULN	Participant ID	Age/ Sex	HIV status	Med Hx	Baseline Tbili	Postbaseline Max Tbili	Study Day of Max Tbili	Baseline ALT	Total ALT at Max Tbili	Medications
>2×	(b) (6)	21/M	Neg	Conjugated hyperbilirubinemia, persistent dysuria, constipation, and headaches	59	42	1	15	11	Paracetamol, castor oil, metronidazole
		20/M	Neg	Thalassemia	30.8	39.3	8	11	15	None

Source: adlb.xpt, admh.xpt, adcm.xpt; Software: MS Excel
Abbreviations: ALT, alanine aminotransferase; F, female; ID, identification; M, male; Max, maximum; Med Hx, medical history; Neg, negative; Tbili, total bilirubin; ULN, upper limit of normal

Narratives for Participants in the Zoliflodacin Arm With Total Bilirubin Values >2× ULN

1. Participant (b) (6) was a 20-year-old Asian male with a history of thalassemia. He had an elevated total bilirubin at Baseline, which remained elevated throughout the trial. This participant had laboratory findings consistent with thalassemia, including low hemoglobin, low hematocrit and mean cell volume. The Applicant attributed the elevation of his bilirubin to unconjugated hyperbilirubinemia related to his chronic thalassemia and this reviewer agrees with this assessment.
2. Participant (b) (6) was a 21-year-old Black male with a history of conjugated hyperbilirubinemia and total hyperbilirubinemia. At Baseline, the participant had an elevated total bilirubin level, which fluctuated and remained elevated during the trial. The Applicant attributed this finding to the participant's underlying history of hyperbilirubinemia of unknown etiology. There is insufficient information on which to base an assessment of causality, but this reviewer agrees that, given the elevated bilirubin at Baseline, the increase post-baseline is likely not attributable to zoliflodacin.

7.6.1.8. Vital Signs, Trial STI_Zoli001

Weight, height and body mass index were collected on all participants in Trial STI_Zoli001 . Vital signs, including pulse, blood pressure measurements, temperature and respiratory rate were not captured. Review of the values from Baseline to TOC and EOS did not reveal significant differences between the two trial arms in the evaluated parameters.

7.6.1.9. Subgroups, Trial STI_Zoli001

The proportion of participants with at least one TEAE in demographic subgroups is shown in Table 38. Men and women reported similar frequencies of AEs between both treatment arms. AEs occurred with a similar frequency between Black, Asian, and White participants within both arms. The small number of participants of other races and ethnicities precludes further conclusions about race-based or ethnicity-based AE imbalances.

Table 38. Overview Occurrence of Adverse Events by Demographic Subgroup, Safety Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=619 n/N_S (%)	Ceftriaxone + Azithromycin N=308 n/N_S (%)
Sex		
Male	252/542 (46.5)	123/270 (45.6)
Female	35/77 (45.5)	20/38 (52.6)
Age group, years		
<18	2/12 (16.7)	2/2 (100)
18 to 64	284/604 (47.0)	140/305 (45.9)
≥65	1/3 (33.3)	1/1 (100)
Age group ≥75, years		
≥75	0/619 (0)	0/308 (0)

Characteristic	Zoliflodacin N=619 n/N_s (%)	Ceftriaxone + Azithromycin N=308 n/N_s (%)
Race		
American Indian or Alaska Native	6/8 (75.0)	0/1 (0)
Asian	101/193 (52.3)	52/92 (56.5)
Native Hawaiian or Other Pacific Islander	1/2 (50.0)	0/1 (0)
Black or African American	155/348 (44.5)	71/164 (43.3)
White	24/65 (36.9)	18/47 (38.3)
Multiple	0/1 (0)	2/2 (100)
Other	0/2 (0)	0/1 (0)
Ethnicity		
Not Hispanic or Latino	278/601 (46.3)	141/295 (47.8)
Hispanic or Latino	9/18 (50.0)	2/13 (15.4)
Is in United States		
Non-United States	244/512 (47.7)	129/257 (50.2)
United States	43/107 (40.2)	14/51 (27.5)

Source: adae.xpt; Software: R
Abbreviations: N, number of participants in treatment arm; n, number of participants with adverse event; N_s, total number of participants for each specific subgroup and were assigned to that specific arm

7.6.1.10. Exposure-Adjusted Pooled Analyses

Not applicable.

7.6.2. Safety Results, Trial DMID 14-0014

7.6.2.1. Overview of Treatment-Emergent Adverse Events Summary, Trial DMID 14-0014

Trial DMID 14-0014 was a phase 2, multicenter, randomized, open-label trial in which participants were randomized 70:70:40 to receive a single, PO dose of either 2000 mg or 3000 mg of zoliflodacin, or one IM dose of 500 mg of ceftriaxone, respectively. A total of 139 participants received a single dose of zoliflodacin and were included in the safety analysis. The combined percentage of TEAEs in the zoliflodacin arms was 29.5% compared to 45% in the ceftriaxone arm. No TEAEs resulted in discontinuation of zoliflodacin or trial withdrawal. No deaths or SAEs related to zoliflodacin were reported during the trial. One SAE occurred in a participant who suffered a gunshot wound that was life threatening and required hospitalization, but this SAE was determined not to be related to zoliflodacin. See section 7.6.2.3 for more details. There were no AEs that led to discontinuation, modification, or interruption of the study drug.

Table 39. Overview of Adverse Events, Safety Population, Trial DMID 14-0014

	Zoliflodacin			Ceftriaxone
	2 g N=72	3 g N=67	Total N=139	500 mg N=40
Event Category	n (%)	n (%)	n (%)	n (%)
SAE	0	1 (1.5)	1 (0.7)	0
SAEs with fatal outcome	0	0	0	0
Life-threatening SAEs	0	1 (1.5)	1 (0.7)	0
SAEs requiring hospitalization	0	1 (1.5)	1 (0.7)	0
SAEs resulting in substantial disruption of normal life functions	0	1 (1.5)	1 (0.7)	0
Any AE	18 (25.0)	23 (34.3)	41 (29.5)	18 (45.0)
Severe and worse	0	1 (1.5)	1 (0.7)	0
Moderate	4 (5.6)	3 (4.5)	7 (5.0)	4 (10.0)
Mild	14 (19.4)	19 (28.4)	33 (23.7)	14 (35.0)

Source: adae.xpt; Software: R
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Severity as assessed by the investigator.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants with at least one event; SAE, serious adverse event

7.6.2.2. Deaths, Trial DMID 14-0014

No deaths were reported in Trial DMID 14-0014.

7.6.2.3. Serious Treatment-Emergent Adverse Events, Trial DMID 14-0014

One SAE was reported in one participant in this trial, i.e., a nonfatal SAE of gunshot wound to the arm and chest cavity. This event occurred 8 days following administration of the 3 gram dose of zoliflodacin. This participant was hospitalized for 7 days. The investigator and Applicant determined this SAE to be unrelated to zoliflodacin and this reviewer concurs with the causality assessment.

7.6.2.4. Adverse Events and OCMQs Leading to Treatment Discontinuation, Trial DMID 14-0014

No TEAEs resulted in treatment discontinuation in Trial DMID 14-0014.

7.6.2.5. Treatment-Emergent Adverse Events, Trial DMID 14-0014

In the zoliflodacin arm, a total of 41 (29.5%) participants experienced at least one TEAE (25% of participants in the 2 gram dose group and 34.4% of participants in the 3 gram dose group). The most frequently reported AE was headache (4.3%) followed by diarrhea (4.3%) and nausea (2.9%). Headache occurred only in participants receiving the 3 gram dose, suggesting a potential dose-dependent relationship. Most TEAEs were Grade 1 or Grade 2 in severity. Only a single

Grade 4 TEAE was reported as discussed in section 7.6.2.3. No Grade 3 or Grade 5 TEAEs were reported. Most of the TEAEs resolved by the end of the trial.

Table 40. Participants With Common Adverse Events Occurring at ≥1% Frequency, Safety Population, Trial DMID 14-0014

	Zoliflodacin			Ceftriaxone
Preferred Term	2 g N=72 n (%)	3 g N=67 n (%)	Total N=139 n (%)	500 mg N=40 n (%)
Any AE	18 (25.0)	23 (34.3)	41 (29.5)	18 (45.0)
Headache	0	6 (9.0)	6 (4.3)	2 (5.0)
Gonorrhea	1 (1.4)	6 (9.0)	7 (5.0)	4 (10.0)
Diarrhea	4 (5.6)	2 (3.0)	6 (4.3)	3 (7.5)
Flatulence	0	2 (3.0)	2 (1.4)	0
Nausea	2 (2.8)	2 (3.0)	4 (2.9)	0
Abdominal distension	0	1 (1.5)	1 (0.7)	0
Abdominal pain	0	2 (3.0)	2 (1.4)	0
Cervicitis	0	1 (1.5)	1 (0.7)	0
Chlamydial infection	1 (1.4)	4 (6.0)	5 (3.6)	2 (5.0)
Gun shot wound	0	1 (1.5)	1 (0.7)	0
Leukopenia	1 (1.4)	2 (3.0)	3 (2.2)	0
Mean cell hemoglobin concentration decreased	0	1 (1.5)	1 (0.7)	0
Metabolic disorder	0	1 (1.5)	1 (0.7)	0
Oropharyngeal pain	0	1 (1.5)	1 (0.7)	0
Pruritus	0	1 (1.5)	1 (0.7)	0
Urinary tract infection	0	1 (1.5)	1 (0.7)	0
Vomiting	0	1 (1.5)	1 (0.7)	0
Vulvovaginal mycotic infection	0	1 (1.5)	1 (0.7)	0
Balanoposthitis	3 (4.2)	0	3 (2.2)	1 (2.5)
Blood bilirubin abnormal	1 (1.4)	0	1 (0.7)	0
Depressed mood	1 (1.4)	0	1 (0.7)	0
Dermatitis	1 (1.4)	0	1 (0.7)	0
Fatigue	1 (1.4)	0	1 (0.7)	0
Hyperglycemia	2 (2.8)	1 (1.5)	3 (2.2)	0
Neutrophil count decreased	2 (2.8)	0	2 (1.4)	0
Secondary syphilis	1 (1.4)	0	1 (0.7)	0
Skin laceration	1 (1.4)	0	1 (0.7)	0
Upper respiratory tract infection	0	0	0	1 (2.5)
Urethral discharge	1 (1.4)	0	1 (0.7)	0
Urethritis	1 (1.4)	0	1 (0.7)	3 (7.5)

Source: adae.xpt; Software: R, MS Excel
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Coded as MedDRA version 26.0 preferred terms.
Combined terms: Skin laceration includes laceration; Diarrhea includes diarrhea and loose stools; Flatulence includes flatulence and gas; Chlamydial infection includes cervical chlamydia, anal chlamydia infection and chlamydial infection; Hyperglycemia includes hyperglycemia and elevated glucose; Gonorrhea includes pharyngitis exposure to gonorrhea, re-exposure to gonorrhea, gonorrhea of the pharynx and gonorrhea; Leukopenia includes leukocytopenia and worsening of low white blood cell counts; Abdominal pain includes right lower quadrant abdominal pain and abdominal pain.
Abbreviations: AE, adverse event; MedDRA, Medical Dictionary for Regulatory Activities; N, number of participants in treatment arm; n, number of participants with adverse event

In Trial DMID 14-0014, AEs by OCMQ occurring in 2% of the zoliflodacin participants (based on the total percentage from both zoliflodacin dose groups) were in the following SOCs: Blood

and lymphatic system disorders, Endocrine disorders, Gastrointestinal disorders, Infections and infestation disorders, and Nervous system disorders.

Table 41.Participants With Adverse Events by System Organ Class and OCMQ (Narrow), Safety Population, Trial DMID 14-0014

	Zoliflodacin			Ceftriaxone
System Organ Class OCMQ (Narrow)	2 g N=72 n (%)	3 g N=67 n (%)	Total N=139 n (%)	500 mg N=40 n (%)
Blood and lymphatic system disorders (SOC)				
Leukopenia	1 (1.4)	2 (3.0)	3 (2.2)	0
Anemia	0	0	0	1 (2.5)
Endocrine disorders (SOC)				
Hyperglycemia	2 (2.8)	1 (1.5)	3 (2.2)	0
Hypoglycemia	0	0	0	1 (2.5)
Gastrointestinal disorders (SOC)				
Abdominal pain	0	2 (3.0)	2 (1.4)	0
Diarrhea	4 (5.6)	2 (3.0)	6 (4.3)	3 (7.5)
Nausea	2 (2.8)	2 (3.0)	4 (2.9)	0
Vomiting	0	1 (1.5)	1 (0.7)	0
General disorders and administration site conditions (SOC)				
Fatigue	1 (1.4)	0	1 (0.7)	0
Local administration reaction	0	0	0	2 (5.0)
Infections and infestations (SOC)				
Bacterial infection	4 (5.6)	9 (13.4)	13 (9.4)	9 (22.5)
Fungal infection	0	1 (1.5)	1 (0.7)	0
Nasopharyngitis	0	0	0	1 (2.5)
Nervous system disorders (SOC)				
Headache	0	6 (9.0)	6 (4.3)	2 (5.0)
Psychiatric disorders (SOC)				
Depression	1 (1.4)	0	1 (0.7)	0
Renal and urinary disorders (SOC)				
Renal & urinary tract infection	1 (1.4)	1 (1.5)	2 (1.4)	3 (7.5)
Skin and subcutaneous tissue disorders (SOC)				
Pruritus	0	1 (1.5)	1 (0.7)	0
Rash	1 (1.4)	0	1 (0.7)	0

Source: adae.xpt; Software: R
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of study drug until 30 days follow-up period.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Each OCMQ is aligned to a single SOC based on clinical judgment. However, please be aware that some OCMQs may contain PTs from more than one SOC.
Some preferred terms are not included in any OCMQ. Those preferred terms are not shown or counted in this table.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants with adverse event; OCMQ, OND custom medical query; OND, Office of New Drugs; PT, preferred term; SOC, system organ class

Leukopenia

The incidence of leukopenia was higher in the zoliflodacin arm (2.2%) compared to the ceftriaxone arm (0%). The leukopenia was thought to be related to the study drug in only one participant. One participant in the 2 gram dose group and one participant in the 3 gram dose group had moderate leukopenia. One participant in the 3 gram dose group had mild leukopenia. By the end of the trial, leukopenia had resolved or was resolving in two participants.

Hyperglycemia

More participants reported hyperglycemia in the zoliflodacin arm than the ceftriaxone arm (2.2% versus 0%, respectively). Mild hyperglycemia was reported in one participant in the 3 gram dose group. It was determined to be a non-fasting glucose value and thought not to be related to zoliflodacin. In the 2 gram dose group, two participants had mild hyperglycemia which was considered unrelated to the study drug.

Gastrointestinal Disorders

In the 2 gram zoliflodacin dose group, there were four participants with AEs of diarrhea and two participants with AEs of nausea. Of the participants who reported diarrhea, three were reported as related to study treatment as diarrhea occurred on Day 1 of the trial after receiving zoliflodacin. One participant reported diarrhea that was likely not due to zoliflodacin as it occurred on Day 29 of the trial. All episodes of diarrhea were classified as mild and resolved. There were two participants who reported nausea after taking zoliflodacin and this AE was considered due to the study drug. Both cases were considered mild and resolved.

In the 3 gram zoliflodacin dose group, there were two participants who reported abdominal pain, two participants who reported diarrhea, two participants who reported nausea, and one participant who reported vomiting. Of the participants who reported abdominal pain, one participant was thought to be related to the study drug as it occurred on Day 1 of the trial following administration of zoliflodacin. One participant had abdominal pain that was thought to be unrelated to the study drug as it occurred on Day 31 of the trial. Both AEs were classified as mild and resolved. Two participants reported diarrhea and two participants reported nausea. Both AEs were considered related to the study drug. These events were classified as mild and resolved. There was one participant who developed vomiting after receiving zoliflodacin. This AE was considered related to the study drug, was classified as mild, and resolved.

Bacterial Infections

In total there were 13 participants in the zoliflodacin arms who experienced bacterial infections. These infections included gonorrhea, chlamydia, and syphilis; the first infection was reflective of the indication under study. All infections resolved by the end of the trial.

Headache

There were six participants who reported headache in the 3 gram zoliflodacin dose group. All AEs were classified as mild. Only one participant who reported a headache was thought not to be related to the study drug. All events resolved.

Hypersensitivity

A single participant (Participant (b) (6) in the phase 2 trial experienced a hypersensitivity reaction, which was a nonserious TEAE of dermatitis of mild intensity (Grade 1) that occurred on Day 31 in a participant who received zoliflodacin 2 g. The event was considered not related to study drug, and the participant recovered in 8 days.

7.6.2.6. Laboratory Findings, Trial DMID 14-0014

In Trial DMID 14-0014, no participants in the zoliflodacin arm developed elevations in creatinine.

Hematology laboratory values, including WBC count, hemoglobin, and platelets were monitored during the trial. In the zoliflodacin arm, 6.5% of participants had a Level 1 (<3.5×10^3 cell/µL) low WBC count and 4.3% of participants had a Level 2 (<3.0×10^3 cell/µL) low WBC count. A higher percentage of participants in the zoliflodacin arm (3.6%) had a Level 2 (>1.5 g/dL decrease from Baseline) low hemoglobin level compared to ceftriaxone (2.5%); however, a higher percentage of participants had a Level 3 (>2 g/dL decrease from Baseline) low hemoglobin in the ceftriaxone arm compared to the zoliflodacin arm (2.5% versus 0.7%, respectively). In the zoliflodacin arm, 3.6% of participants had a Level 1 low platelet count (<140×10^3 cell/µL), 2.2% had Level 2 (<125×10^3 cell/µL) low platelet count and 0.7% had Level 3 (<100×10^3 cell/µL). No participants in the ceftriaxone arm had low platelet counts.

Table 42. Participants With One or More Hematology Value Exceeding Specified Levels, Safety Population, Trial DMID 14-0014

	Zoliflodacin			Ceftriaxone
	2 g N=72	3 g N=67	Total N=139	500 mg N=40
Laboratory Parameter	n/N_w (%)	n/N_w (%)	n/N_w (%)	n/N_w (%)
Complete blood count				
WBC, low (10^3 cells/µL)				
Level 1 (<3.5)	5/72 (6.9)	4/67 (6.0)	9/139 (6.5)	2/40 (5.0)
Level 2 (<3)	3/72 (4.2)	3/67 (4.5)	6/139 (4.3)	0/40 (0)
Level 3 (<1)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
WBC, high (10^3 cells/µL)				
Level 1 (>10.8)	1/72 (1.4)	3/67 (4.5)	4/139 (2.9)	3/40 (7.5)
Level 2 (>13)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
Level 3 (>15)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
Hemoglobin, low (g/dL)				
Level 2 (>1.5 g/dL dec. from Baseline)	2/72 (2.8)	3/67 (4.5)	5/139 (3.6)	1/40 (2.5)
Level 3 (>2 g/dL dec. from Baseline)	0/72 (0)	1/67 (1.5)	1/139 (0.7)	1/40 (2.5)
Hemoglobin, high (g/dL)				
Level 2 (>2 g/dL inc. from Baseline)	0/72 (0)	1/67 (1.5)	1/139 (0.7)	0/40 (0)
Level 3 (>3 g/dL inc. from Baseline)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
Platelets, low (10^3 cells/µL)				
Level 1 (<140)	4/72 (5.6)	1/67 (1.5)	5/139 (3.6)	0/40 (0)
Level 2 (<125)	2/72 (2.8)	1/67 (1.5)	3/139 (2.2)	0/40 (0)
Level 3 (<100)	0/72 (0)	1/67 (1.5)	1/139 (0.7)	0/40 (0)

Source: adlb.xpt; Software: R
Threshold levels 1, 2, and 3 as defined by the Standard Safety Tables & Figures Integrated Guide.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
Abbreviations: dec., decrease; inc., increase; N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data; WBC, white blood cell

7.6.2.7. Assessment of Drug-Induced Liver Injury, Trial DMID 14-0014

There were no significant differences among the treatment arms in the proportion of participants with abnormal liver chemistry values (ALT, total bilirubin). Alkaline phosphatase and aspartate

aminotransferase values were not evaluated during this trial. There was one participant in the zoliflodacin arm who had ALT>3× ULN (0.7%) at Baseline and ALT >5× ULN (0.7%) at EOS. No participants in any treatment arm had a total bilirubin level >2× ULN. There were no Hy's Law cases in any of the three arms.

Table 43. Participants With One or More Liver Biochemistry Analyte Values Exceeding Specified Levels, Safety Population, Trial DMID 14-0014

	Zoliflodacin			Ceftriaxone
	2 g N=72	3 g N=67	Total N=139	500 mg N=40
Laboratory Parameter	n/N_w (%)	n/N_w (%)	n/N_w (%)	n/N_w (%)
Alanine aminotransferase, high (U/L)				
Level 1 (>3× ULN)	0/72 (0)	1/67 (1.5)	1/139 (0.7)	0/40 (0)
Level 2 (>5× ULN)	0/72 (0)	1/67 (1.5)	1/139 (0.7)	0/40 (0)
Level 3 (>10× ULN)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
Bilirubin, total, high (mg/dL)				
Level 1 (>1.5× ULN)	2/72 (2.8)	2/67 (3.0)	4/139 (2.9)	1/40 (2.5)
Level 2 (>2× ULN)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)
Level 3 (>3× ULN)	0/72 (0)	0/67 (0)	0/139 (0)	0/40 (0)

Source: adlb.xpt; Software: R
Threshold levels 1, 2, and 3 as defined by the Standard Safety Tables & Figures Integrated Guide.
Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
For specific evaluation of drug-induced liver injury (DILI), see the figure "Hepatocellular Drug-Induced Liver Injury Screening Plot.." and the table "Patients in Each Quadrant for Potential Hepatocellular DILI Screening Plot.."
In addition to central laboratory data, local laboratory data may be included in the analysis, if applicable.
Abbreviations: N, number of participants in treatment arm; n, number of participants meeting criteria; N_w, number of participants with data; ULN, upper limit of normal

Participants With Elevated Liver Enzymes Postbaseline

A narrative for the participant in the zoliflodacin arm with ALT values >5× ULN is provided below:

Participant (b) (6) was a 20-year-old male with a medical history of hepatitis C. At Baseline, his ALT level was 219 IU/L. On Day 6, his ALT level was 176 IU/L and on Day 42 (EOS) his ALT level was 466 IU/L. He received treatment with azithromycin on Day 6 for a *Chlamydia* infection, but no additional medications were reported. Given that the ALT was elevated at Baseline and there is history of hepatitis C and treatment with azithromycin, this reviewer does not attribute the ALT elevation to administration of zoliflodacin.

7.6.2.8. Vital-Sign Analyses, Trial DMID 14-0014

Oral temperature was collected in Trial DMID 14-0014 along with weight, and height. Pulse, blood pressure measurements, and respiratory rate were not captured. No clinically significant changes in temperature, height, or weight were observed in participants who received zoliflodacin during Trial DMID 14-0014.

7.6.2.9. Subgroup Analyses, Trial DMID 14-0014

Not applicable.

7.6.2.10. Exposure-Adjusted Analyses, Trial DMID 14-0014

Not applicable.

7.7. Key Safety Review Issues

7.7.1. Male Reproductive Toxicity

Issue

Male reproductive organs in rats and dogs and male rat fertility were identified as targets of toxicity of zoliflodacin with 2 days to 5 weeks of oral zoliflodacin administration. In these studies, the reduction in male rat fertility was fully reversible following a recovery period, but the histopathology in the male reproductive organs in rats and dogs was incompletely reversed in that time. The review team evaluated the available nonclinical safety data to propose edits to the Applicant's draft labeling to communicate a potential male fertility and reproductive organ toxicity risk in the zoliflodacin PI.

Background and Assessment

Decreased male rat fertility and male reproductive organ toxicity in rats and dogs were observed in oral repeat-dose studies of zoliflodacin. Summaries of these studies are described in Section 7.1 and Section 13.

The impact of zoliflodacin on human male reproductive toxicity is uncertain as no human data are available. While the fertility in the rats recovered after a 4-week recovery period, there was incomplete reversibility of the histopathology in the epididymides in rats and dogs. Analyses of sperm count and viability have not been conducted in animals or humans. Because zoliflodacin will be administered as a single dose to patients and has an approximately 6.5-hour human plasma $t_{1/2}$, there are uncertainties in determining the risk to patients based on the findings from repeat-dose animal studies, and the higher exposures seen in the animal studies compared with those expected in humans. However, the potential for male reproductive organ toxicity (decreased spermatogenesis and degeneration), and decreased fertility in human males after treatment with zoliflodacin is relevant to clinical use and the review team determined that it should be addressed by inclusion of a Warning in labeling (see Section 23).

Conclusion

Since decreased male fertility and toxicity to male reproductive organs was observed following zoliflodacin dosing in two animal species across several but not all studies, and clinical data on the risk to human male reproductive organs are not available, the review team recommends that the risk to patients be managed through the addition of a Warning in the PI, describing the observed toxicity observed in animal studies. Inclusion of this Warning will underscore the need to administer zoliflodacin in patients for whom the benefits outweigh the risks. The review team has also recommended a postmarketing sperm study in humans to evaluate sperm count and viability following treatment with zoliflodacin.

7.7.2. Embryo-Fetal Toxicity

Issue

In a mouse embryo-fetal development study after zoliflodacin administration, an increase in exencephaly, a neural tube defect, was detected above concurrent and historical control levels at exposures of 1.6-fold the clinical AUC and above. Additionally, an increase in intrauterine pre- and post-implantation loss was found in female rats dosed from two weeks prior to mating through organogenesis at and above the approximate equivalent of 6-fold the clinical AUC. Reductions in total implants and live implants were found in gravid mice dosed during organogenesis at doses equivalent to and above 1.6-fold the clinical AUC.

Background and Assessment

Embryo-fetal toxicity was found in studies of gravid rats and mice. In mice, exencephaly, a rare neural tube defect was reported, and in both species, an increase in embryonic loss or decrease in live fetuses was reported. Summaries of these studies are described in Section 7.1 and Section 13.

The Applicant's mouse embryo-fetal development study report had attributed the exencephaly to background findings. This finding is generally rare but upon review, the rate of occurrence in the study exceeded that in concurrent controls (where there were none reported) and in historical control data from the same lab during the years around when the study was conducted. The Division therefore concluded that exencephaly was related to zoliflodacin administration in the study. Additionally, a reduction in mean numbers of implants and mean live implants were found at doses leading to exposure at and above approximately 1.6-fold the clinical AUC and an increase in post-implantation loss at about 3-fold the clinical AUC.

In a study of female rats dosed from 2 weeks prior to mating with untreated males and through organogenesis, early intrauterine deaths (dead implants and early embryonic deaths) were increased 3-fold at doses leading to extrapolated exposures of 6-fold the clinical AUC and above.

Because of the potential risk of exencephaly, a Warning was included in the PI regarding the embryo-fetal toxicity seen in mice and rats. Risk mitigation measures include alerting healthcare providers to avoid administering zoliflodacin to pregnant women and to test for pregnancy in women of reproductive potential before prescribing zoliflodacin.

Conclusion

Since embryo-fetal toxicity from zoliflodacin administration during pregnancy was reported in mice and rats, the review team recommends that the potential risk to female patients be managed through the addition of a Warning in the PI (see Section 23), pregnancy testing in women of reproductive potential, and avoiding administration to pregnant women.

7.7.3. Potential Risk Related to Males with Female Partners of Reproductive Potential

Issue

Increase in fetal loss was reported in rat studies where untreated females were mated with males treated with zoliflodacin for 4 weeks prior to mating. This effect was no longer present after a 4-week recovery period. The review team evaluated the available nonclinical safety data to propose edits to the Applicant's draft labeling to communicate a potential developmental toxicity risk for Males with Female Partners of Reproductive Potential in the zoliflodacin PI and recommend contraceptive use for 90 days following treatment.

Background and Assessment

In the male rat fertility study, reduced number of live embryos and increased number of embryonic losses were observed in untreated female rats mated with male rats administered zoliflodacin at exposures approximately 4-fold the clinical exposure at the MRHD for 4 weeks. This indicates a potential risk of developmental toxicity in offspring of a partner of a male taking zoliflodacin. The risk may be related to the testicular toxicity or a direct effect causing decreased sperm quality, however, no direct data on sperm from clinical or nonclinical studies is available. The potential for developmental risk in offspring of female partners of reproductive potential after treatment of males with zoliflodacin is relevant to clinical use and the review team recommends use of contraception by males taking zoliflodacin who have female partners of reproductive potential for a full male sperm cycle to mitigate the risk (see Section 8.3 of the PI). The review team also determined that this risk should be addressed by inclusion of a Warning in labeling (see Section 23).

Conclusions

A clinical risk of increased early pregnancy loss in female partners of reproductive potential of males administered zoliflodacin was derived from this finding in a male rat fertility study. This risk can be mitigated with the use of contraception for the duration of the cycle time for the sperm exposed during treatment and is highlighted with the inclusion of a Warning in Section 5.2 of the PI and Section 8.3 of the PI.

7.7.4. Additional Analysis of Neutropenia

Low neutrophil cell counts were reported as an adverse event in more than 10% of trial participants in both treatment arms. Further analysis of objective laboratory data confirmed this finding of neutropenia; however, both treatment arms had similar rates of neutropenia. Please refer to section 7.6.1.6.1 for further details.

8. Therapeutic Individualization

8.1. Intrinsic Factors

Renal Impairment

Zoliflodacin is eliminated primarily by a nonrenal route. Less than 25% of the parent drug and metabolites are detected in urine in a mass balance study of radiolabeled zoliflodacin. Additionally, zoliflodacin is a single-dose drug product. Overall, the available data suggest no dose adjustment for zoliflodacin is warranted for patients with renal impairment.

Hepatic Impairment

Zoliflodacin clearance (CL) is mainly via metabolism by cytochrome P450 (CYP)- and non-CYP-mediated pathways. The Applicant proposed that zoliflodacin is a single-dose drug product, therefore no dosage adjustment is needed for patients with hepatic impairment. The Applicant also noted that because no clinical data are available in patients with hepatic impairment, caution should be used in patients with severe hepatic impairment, especially when other factors that could increase exposures are considered (e.g., low body weight, fasted status, and/or use of CYP3A4 inhibitors). However, the Applicant did not propose any language related to zoliflodacin's use in patients with severe hepatic impairment in Section 8 of the proposed label.

The potential risk for patients with hepatic impairment after administration of zoliflodacin would be primarily related to the risk of QT prolongation at increased C_{max} (compared to patients without hepatic impairment). Specifically, patients with severe hepatic impairment have the potential for a higher unbound fraction of zoliflodacin and a higher C_{max} and $AUC_{0-\infty}$ due to the typically lower abundance of plasma proteins and metabolizing enzymes in this patient population. Based on the review of the following safety findings, we agree that no dose adjustment is recommended for patients with hepatic impairment:

No clinically significant QTc prolongation was observed in the food effect study (STI_Zoli002), which included 24-hr Holter recordings and evaluated doses up to 4 g under fed condition (1.3-fold the proposed dosage).

No clinically concerning cardiovascular adverse events were reported in the completed phase 3 trial for the zoliflodacin arm. STI_Zoli001 had exclusion criteria to exclude participants with known chronic renal, hepatic, or hematologic impairment or other conditions interfering with the absorption, distribution or elimination of the drug based on medical history and physical examination. However, examination of the medical history dataset shows that participants with the following conditions, which may be indicative of potential liver dysfunction, were enrolled in the phase 3 trial: jaundice, alcoholic liver disease, acute Hepatitis A, acute Hepatitis B, chronic Hepatitis B, chronic Hepatitis C, Hepatitis B carrier, Hepatitis C, viral Hepatitis B, viral Hepatitis C, transaminases increased, direct hyperbilirubinemia, hyperbilirubinemia, conjugated bilirubin increased, total bilirubin increased, and blood bilirubin increased. Additionally, examination of the baseline serum chemistry dataset of the phase 3 trial participants demonstrated that participants with up to approximately 2 x ULN of total bilirubin (range of 0.2 to 3.3 mg/dL) and approximately 8 x ULN ALT (range 1 to 457 IU/L) were enrolled in the phase 3 trial. The enrollment of participants with a medical history

or baseline laboratory values that could be indicative of hepatic dysfunction in the phase 3 trial, and the lack of clinically concerning cardiovascular adverse events in this trial provide additional data to mitigate the risk of zoliflodacin in patients with hepatic impairment.

The review team also evaluated the impact of other patient factors (e.g., low body weight, fasted status, and/or use of CYP3A4 inhibitors) and findings are summarized in various sections of this review. Specifically, the body weight and food effect on zoliflodacin exposure is summarized in Section 8.1 and Section 8.2, respectively. The impact of CYP3A4 inhibitors on zoliflodacin exposure is summarized in Section 8.2. Collectively, the impact of these patient factors is either properly mitigated (body weight and food effect) or considered insignificant (CYP3A4 inhibitors) and does not warrant cautionary language for zoliflodacin's use in patients with severe hepatic impairment.

Overall, based on the totality of data and considering zoliflodacin is a single-dose drug product, we do not recommend any further dedicated PK studies to address the risks associated with hepatic impairment, beyond normal postmarketing safety data collection.

Other Intrinsic Factors

In a population PK analysis of zoliflodacin, body weight was identified to have a clinically meaningful impact on zoliflodacin pharmacokinetics. Specifically, an inverse relationship between body weight and zoliflodacin exposure was found, for both $AUC_{0-\infty}$ and C_{max}. These findings informed the following varying dosage administration instructions proposed by the Applicant with respect to food and weight:

Patients weighing $\geq$35 kg to <50 kg are instructed to take zoliflodacin under fasted conditions.

Patients weighing >50 kg are instructed to take zoliflodacin under fed conditions.

The population PK analysis indicated that sex (72.8% male) did have an impact on zoliflodacin clearance, with female participants having 13.9% slower clearance, which is not anticipated to be clinically significant. Additionally, zoliflodacin is a single-dose drug product, therefore there are no concerns regarding accumulation, and no dosage adjustment is needed to address the observed sex-based differences in clearance.

The population PK analysis also indicated that race (60.9% Caucasian, 28.0% Black/African American, 7.66% Asian, 1.92% American Indian/Alaska Native and 1.53% other or unknown), or age (19 to 55 years), do not have a clinically meaningful impact on zoliflodacin pharmacokinetics (see Section 14.5.1).

8.2. Extrinsic Factors

Food Effect

Zoliflodacin bioavailability is significantly increased when administered with food, as summarized in Table 44 below. The to-be-marketed formulation, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd. (ZoliDr GFOS), and the formulation used in the phase 3 clinical trial, zoliflodacin GFOS manufactured by Patheon (ZoliPa GFOS), demonstrated comparable zoliflodacin bioavailability under fasting and fed conditions in a dedicated relative bioavailability study (See Section 14.2.2). The only differences between the

ZoliPa GFOS formulation and the ZoliDr GFOS formulation are that ZoliDr GFOS is manufactured at a different site with a different process than that used for the manufacturing of ZoliPa GFOS.

Zoliflodacin GFOS was evaluated under both fed and fasted conditions in food effect studies. Formulation ZoliDr GFOS was evaluated under moderate-fat, moderate-calorie standardized meal fed and fasted conditions (see Section 14.2.2). Formulation ZoliPa GFOS was evaluated under high-calorie, high-fat meal fed and fasted conditions (see Section 14.2.2).

When a single 3 g dose of zoliflodacin GFOS is administered within 30 minutes of consuming a moderate-to-high-fat meal, $AUC_{0-\infty}$ estimate was approximately 1.5- to 2-fold, and C_{max} was approximately 1.5-fold when compared to respective estimates under fasted condition.

The proposed dosage administration instructions with respect to food vary based on patient weight.

Table 44. Summary of Statistical Analysis To Assess the Effect of Food on Plasma PK Parameters of Zoliflodacin

			Ratio (%) (Fed/Fasted)	
Study	Formulation and Dose	Meal Composition	C_{max} GM Ratio (90% CI)	$AUC_{0-\infty}$ GM Ratio (90% CI)
STI_Zoli003	3 g ZoliDr GFOS	Moderate-fat/moderate-calorie standardized meal	149.182 (138.48, 160.71)	152.901 (145.20, 161.01)
STI_Zoli002	3 g ZoliPa GFOS	High-calorie, high-fat meal	152.39 (139.30, 166.71)	200.87 (185.10, 217.99)

Source: Reviewer compiled from Table 15 STI_Zoli003 CSR (page 69), and Table 9 of STI_Zoli002 CSR (page 41)
Abbreviations: AUC, under the time-concentration curve; CI, Confidence Interval; C_{max}: maximum plasma concentration; GM, Geometric Mean; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Drug-Drug Interactions

In Vitro Studies

Zoliflodacin is a substrate of CYPs 3A4/5, 1A2, 2C9, 2C8, and 2C19, and the drug transporters P-glycoprotein (P-gp) and breast cancer resistance protein (BCRP). Zoliflodacin demonstrated inhibition potential of CYPs 2C8, 2C9, and 2C19 in vitro, however, based on mechanistic static model predictions, zoliflodacin has a low risk of a clinically relevant interaction as a direct inhibitor of CYPs 2C8, 2C9, and 2C19 in vivo. Zoliflodacin demonstrated induction of CYP1A2 in vitro. Zoliflodacin has inhibition potential for P-gp, BCRP, organic anion transporting polypeptide (OATP) 1B1/1B3, organic anion transporter (OAT) 1/3, multidrug and toxin extrusion transporter 1 (MATE1), and MATE2K drug transporters. For more details see Sections 14.1.2.2 through 14.1.2.4.

No reversible or time-dependent inhibition of CYP3A4/5 was observed for zoliflodacin at <500µM (50× maximum clinical concentration) in vitro. Zoliflodacin's potential to inhibit CYP3A4/5 has not been evaluated at concentrations >500µM. Of note, the anticipated maximum zoliflodacin concentration in the gastrointestinal (GI) tract is estimated to be 1538µM and 24620µM after considering maximum reported solubility and without regard to solubility, respectively. Given that zoliflodacin 1) has been shown to be rapidly absorbed with a median time to maximum plasma concentration (T_{max}) of 2.5 and 4 hours under fasted and fed

conditions, respectively; 2) has a $t_{1/2}$ of approximately 6 hours (see Section 14.2.2); and 3) is being recommended for single dose, the risk of zoliflodacin inhibition of intestinal CYP3A4/5 would be transient, if any. Therefore, further studies are not being recommended to evaluate the drug-drug interactions (DDIs) of zoliflodacin as an inhibitor of intestinal CYP3A4/5.

Clinical Studies

A drug-drug interaction study in healthy participants showed <40% increase in zoliflodacin exposure when a single 3 g zoliflodacin dose was administered with the strong CYP3A4 inhibitor itraconazole compared to when administered alone. Based on the available safety data for up to a 4 g single dose under fed condition, no dose adjustment is recommended for zoliflodacin when coadministered with CYP3A4 inhibitors. For more details see Section 14.2.2.

Physiologically Based Pharmacokinetic Modeling Studies

Physiologically based pharmacokinetic (PBPK) modeling and simulations were conducted by the Applicant to predict the impact of the following scenarios on zoliflodacin pharmacokinetics following administration of a single 3 g dose: 1) CYP3A inhibition with or without food effect; 2) CYP3A induction; and 3) mild, moderate, and severe hepatic impairment. The Applicant also used PBPK analysis to predict the impact of zoliflodacin as a precipitant on the pharmacokinetics of substrates for transporters (i.e., P-gp, BCRP, OAT1, OAT3, OATP1B1, OATP1B3, MATEs).

The PBPK model was adequate to simulate the effect of the CYP3A inhibitors on a single oral dose of zoliflodacin. A strong CYP3A inhibitor (e.g., itraconazole) is predicted to increase zoliflodacin AUC by ~1.4-fold under fasted conditions. Food is not expected to impact the magnitude of predicted DDI. Coadministration of zoliflodacin with the CYP3A4 inducers, rifampin and efavirenz, is predicted to reduce the geometric mean zoliflodacin exposure ($AUC_{0-\infty}$) by approximately 56% and 41%, respectively. The review team agrees that moderate to strong CYP3A4 inducers should not be coadministered with zoliflodacin and are included as a contraindication in product labeling. The likelihood of interaction with substrates of P-gp, BCRP, MATEs, OAT1, OAT3, OATP1B1, and OATP1B3 cannot be ruled out. However, given zoliflodacin's single dose administration and short $t_{1/2}$, the DDI risk is low and no dose adjustment is needed. The hepatic impairment simulations were not reviewed since hepatic impairment is not considered a significant clinical risk for this drug (see Section 8.1). See Section 14.5.2 for details on PBPK modeling review.

8.3. Plans for Pediatric Drug Development

There are currently no clinical data available on the safety and efficacy of zoliflodacin in pediatric patients <12 years old. An agreed initial Pediatric Study Plan (iPSP) was finalized on December 17, 2019, and included plans to enroll pediatric participants 12 years and older with uncomplicated urogenital gonorrhea in the phase 3 clinical trial, STI_Zoli001. A partial waiver for pediatric participants <12 years of age was requested. The rationale for this partial waiver was based on 1) limited therapeutic benefit compared to established therapies in pediatric participants, particularly in neonates with disseminated gonococcal infections, and 2) the impracticability of clinical trials in children aged 11 years and younger with uncomplicated gonorrhea, as occurrence in this age group is rare and usually associated with sexual assault or abuse.

Trial STI_Zoli001 began enrollment of adult participants in 2019 and following the iPSP agreement, enrollment of adolescent pediatric participants began in 2020. Due to COVID-19 prevention measures, trial enrollment was paused on March 23, 2020, but reopened on June 8, 2020.

The dose of zoliflodacin for pediatric participants aged 12 years and older, weighing at least 35 kg, was extrapolated to be the same as in adults. There were no PK data in adolescent participants (12 to 18 years of age) and limited PK data in participants weighing <50 kg. The Applicant conducted a PopPK analysis of zoliflodacin using data from six phase 1 studies and the phase 3 trial, along with PK-PD target attainment analyses to support the proposed dosing regimen and susceptibility testing criteria for *N. gonorrhoeae*. The PopPK model adequately described observed pharmacokinetic data in healthy participants and participants with uncomplicated urogenital gonorrhea ≥50 kg, reasonably captured food effects, and used key assumptions for unstudied populations including that body weight (not age) is the primary PK determinant and that food effects and PK-PD relationships are similar between adults and adolescents. Simulations demonstrated that adolescent and adult patients weighing 35 to 50 kg receiving 3 g in a fasted state achieve comparable exposures to those ≥50 kg receiving 3 g in a fed state, with no additional QT prolongation risk expected for the lighter weight group based on concentration-QTc relationships. The PK-PD target attainment analyses showed >90% probability of achieving the efficacy target across both weight groups. Overall, the proposed weight-based dosing regimen is supported by the integrated PopPK analysis, PTA analysis, and phase 3 efficacy and safety data.

No further pediatric pharmacokinetic or clinical studies are planned.

8.4. Pregnancy, Lactation, and Females/Males of Reproductive Potential

The following nonclinical information was used in support of the drug's labeling. Additional details are available in Section 13.

Table 45. Nonclinical Data in Sections 8 and 13 of the Zoliflodacin Prescribing Information

Labeling Section	Nonclinical Data
8.1 Pregnancy	In a fertility and embryo-fetal development study in female rats, zoliflodacin was administered by oral gavage at doses of 0, 200, 500, or 1000 mg/kg/day for 2 weeks prior to mating until gestation day (GD) 16. No significant differences in estrus cycles, mating, or precoital index were reported. In the 500 and 1000 mg/kg/day dose groups, the number of early intrauterine deaths increased. Mean gravid uterine weights were 24% lower in the 1000 mg/kg dose group compared to controls. Mean litter weights were significantly lower in the 200 and 1000 mg/kg dose groups (18% and 29%, respectively). No test article-related external or visceral fetal abnormalities were reported. In all dose groups, an increase in pups per litter with skeletal variations and a decrease in skeletal ossification were reported. There was no significant evidence of maternal toxicity in this study. A no-adverse-effect-level (NOAEL) for fetal development could not be determined in this study based on the change in litter weights. In an embryo-fetal development study in mice, zoliflodacin was administered to gravid mice by oral gavage from GD 5 to GD 16, twice daily at 0, 125, 250, or 500 mg/kg/dose (250, 500, or 1000 mg/kg/day). One 1000 mg/kg female was euthanized early due to adverse clinical signs. A reduction in mean numbers of implants and mean live implants was found in the 500 and 1000 mg/kg/day dose groups, and an increase in post-implantation losses was found in the 1000 mg/kg/day group. Mean gravid uterine weights were lower in all test-article treated groups compared to controls. Mean litter male fetus weights, female fetus weights, and combined fetus weights were significantly lower in the 1000 mg/kg/day group compared to controls. In the 500 and 1000 mg/kg/day dose groups, exencephaly, a rare malformation, was present at rates above that in concurrent and historical controls. In the 500 and 1000 mg/kg/day dose groups, an increase in delayed ossification was reported. There was no evidence of test article-related maternal toxicity up to the highest dose tested. The NOAEL was 250 mg/kg/day (125 mg/kg/dose) in this study for developmental endpoints. In a pre- and postnatal development study, gravid female rats were administered 0, 50, 100, or 200 mg/kg/day zoliflodacin by oral gavage from GD 6 until weaning on lactation day 20. No differences in mating, fertility, or pregnancy parameters were reported in the F1 groups. In a motor task assessment, ambulatory counts and distance traveled were higher in high-dose F1 males and mid- and high-dose females compared to controls. No test article-related abnormalities were reported in the F2 pups. Based on the changes in motor tasks in the F1 animals at and above 100 mg/kg/day, the NOAEL was 50 mg/kg/day. This determination does not consider any measurement of learning and memory in F1 animals, which were not assessed with an adequately designed test in this study.
8.2 Lactation	No data were available.
8.3 Females and males of reproductive potential	As described in labeling section 8.1, in an embryo-fetal development study in mice, administration of 500 mg/kg/day zoliflodacin was associated with an increase in occurrence of exencephaly. Based on this increase, avoidance of zoliflodacin administration is recommended during pregnancy. Administration of zoliflodacin to male rats for 4 weeks caused a reversible loss of fertility. Pre- and post-implantation loss increases were observed when treated male rats were mated with untreated female rats. These findings will be described in Section 13.1 Carcinogenesis, mutagenesis, impairment of fertility. A contraceptive recommendation is therefore warranted for one human male sperm production cycle or approximately 90 days after a single dose administration.

Labeling Section	Nonclinical Data
13.1 Carcinogenesis, mutagenesis, impairment of fertility	In a fertility and early embryonic rat development study, 0, 200, 500, or 1000 mg/kg/day zoliflodacin was administered by oral gavage to male rats for 4 weeks prior to mating and through the mating period. The male rats were then remated with a second set of untreated female rats after a 4-week recovery period. During the first mating period, the number of matings and time to mating were similar across groups. The male fertility index was reduced to 68.8% in the 500 mg/kg dose group and 0% in the 1000 mg/kg dose group. There was an increase in pre-implantation loss (24.9% versus 3.2%) and post-implantation loss (12.2% versus 2.3%) compared to controls in the 500 mg/kg/day dose group for the first mating period (during dosing). During the second mating period, mating and pregnancy parameters were similar across groups. At necropsy at the end of the recovery period (around Day 91), histopathological changes in the male rats were noted in the 500 and 1000 mg/kg/day dose groups in the epididymides (minimal cellular debris associated with testicular degeneration) and testes (minimal to marked tubular degeneration, minimal tubular vacuolation, and minimal cellular debris). The NOAEL for the study was 200 mg/kg.

Source: Reviewer table
Abbreviations: F1, first generation; F2, second generation; NOAEL, no-observed-adverse-effect level

The information detailed above was derived from fertility and reproductive toxicology studies listed below (Table 46). All safety factors shown are based on systemic exposures compared between animals and humans. Further details are available in Section 13.1.

Table 46. Safety Margins From Reproductive and Developmental Toxicity Studies

Study	NOAEL (mg/kg/day)	Nonclinical Systemic Exposure (µg*hr/mL)	Safety Margin Multiples Based on Exposure[a] or [Multiples Based on HED]
Male rat FEED	200	495	2 [0.6]
Female rat fertility and embryo-fetal development	(200[b])	(1082[c])	3 [0.6]
Mouse EFD	250	204	0.6 [0.4]
Rat PPND	50[d]	42[e]	0.1 [0.2]

Source: Reviewer table
[a] Compared to clinical $AUC_{0\text{-}24h}$ =353 µg*h/mL
[b] LOAEL
[c] No direct measure was available. AUC extrapolated from same dose in the 4-week repeat dose study female AUC values.
[d] Not considering learning and behavior testing for which an adequately conducted test was not conducted.
[e] Exposure as measured on lactation day 21.
Abbreviations: EFD, embryo-fetal development; FEED, fertility and early embryonic development; HED, human equivalent dose; LOAEL, lowest-observed adverse effect level; NOAEL, no-observed-adverse-effect level; PPND, pre- and postnatal development

9. Product Quality

Approval

The proposed commercial drug product is formulated as immediate-release granules for oral suspension, packaged in a laminated aluminum foil sachet as a single dose (3 grams). It will be supplied in a convenience kit with a carton containing one sachet and a mixing container to prepare the suspension in water. The NDA, as amended, has provided sufficient chemistry, manufacturing, and controls (CMC) information to assure the identity, strength, purity, and quality of the proposed drug product, zoliflodacin for oral suspension. That includes stability information to support the proposed 36-month expiry dating for the drug product, to be stored

under controlled room temperature conditions. The manufacturing and testing facilities have been found acceptable and the Overall Manufacturing Inspection Recommendation of "Approve" was entered into Panorama by the Office of Pharmaceutical Manufacturing Assessment on October 3, 2025.

The Office of Pharmaceutical Quality (OPQ) review team has assessed NDA 219491 with respect to CMC and has determined that it meets all applicable standards to support the identity, strength, quality, and purity that it purports. As such, OPQ recommends approval of this NDA from a quality perspective (refer to the OPQ Integrated Quality Review dated October 14, 2025).

9.1. Device or Combination Product Considerations

Not Applicable.

10. Human Subjects Protections/Clinical Site and Other Good Clinical Practice Inspections/Financial Disclosure Review

The Office of Scientific Investigations (OSI) performed inspections at four study sites that participated in Trial STI_Zoli001 and concluded that the trial was conducted in compliance with GCP guidelines and that all local and regulatory requirements were followed. The data generated by these sites appear acceptable in support of the proposed indication.

In the NDA submission, the Applicant provided attestation that Trial STI_Zoli001 was conducted in compliance with GCP guidelines and that all local regulatory requirements were followed. Of note, the Applicant disclosed that there was a serious breach of GCP at a site in South Africa (710-005). Concerns regarding data quality and monitoring at this site were raised during an Applicant site visit in August 2022. An internal audit was conducted and identified irregularities and nonconformance with GCP and standard operating procedures. Additionally, the site principal investigator left the site in September 2022. The Applicant placed recruitment on hold at the site and further site monitoring was led by the Applicant's partnering organization, the GARDP Foundation. In October 2022, the study site was discontinued. In January 2023, a for-cause audit was conducted by the GARDP Foundation and concluded that a serious breach had occurred at the site due to lack of a principal investigator and medical oversight, including failure to appropriately obtain informed consent and failure to provide ongoing medical care. Due to this breach, data obtained from the 31 participants enrolled at this site were excluded from analyses of efficacy and safety.

OSI conducted a review of study sites for Trial STI_Zoli001. Three clinical sites (site 840-102 in Seattle, Washington, site 710-003 in Johannesburg, South Africa, and site 764-001 in Bangkok, Thailand) were inspected based on high enrollment and high treatment response. Additionally, the Applicant's office in Waltham, Massachusetts was inspected. Per the Clinical Inspection Summary by Dr. John Lee, data from the inspected sites appear to be acceptable in support of the proposed indication for zoliflodacin. The primary and secondary endpoints were verifiable and

no significant underreporting of AEs was observed. Inspection of the Applicant site noted no significant regulatory violations and the Applicant's oversight of the trial, including clinical site monitoring, was adequate to ensure participant safety and data quality. The trial appeared to be conducted in compliance with GCP principles and regulations.

At site 710-003, no GCP deficiencies or regulatory violations were noted. At site 764-001, a GCP deficiency was noted. A total of seven participants under the age of 18 years were enrolled without either signed parental consent or a documented institutional review board (IRB) waiver. Verbal IRB approvals via phone calls were documented in the study records but were not followed by written IRB approvals. No other significant GCP deficiencies or regulatory violations were observed. At site 840-102, a GCP deficiency was noted for not excluding four participants from the trial for protocol violations. One participant had limited use of a nonstudy antibacterial CYP3A4 inhibitor drug, two participants self-reported current substance abuse, and one participant reported inadequate contraception. Based on OSI review, these findings appeared isolated and unlikely to significantly impact the study outcome and efficacy results. For additional details, please refer to Section 22 and the clinical inspection summary by Dr. John Lee.

11. Advisory Committee Summary

An advisory committee meeting was not held for this application.

III. Additional Analyses and Information

12. Summary of Regulatory History

Zoliflodacin for oral suspension, for the treatment of uncomplicated gonorrhea, was developed by the following entities under their respective IND applications: IND 118958, established by AstraZeneca Pharmaceuticals LP (AstraZeneca) in 2013, and transferred to Entasis Therapeutics, Inc. in 2015; NIAID/DMID under IND (b) (4) and the GARDP Foundation under IND 139105. Entasis Therapeutics, Inc. (Entasis) is the Applicant of NDA 219491.

AstraZeneca submitted IND 118958 for zoliflodacin on August 8, 2013, with a proposed indication for treatment of uncomplicated *N. gonorrhoeae* infection. The IND opening study, Study D4930C00001, was titled **"***A Phase I, Randomized, Placebo-controlled, Single-center Study to Assess the Safety, Tolerability, and Pharmacokinetics of AZD0914 After Oral Administration of Single Doses or Multiple Doses over 24 Hours and to Assess the Effect of Food in Healthy Adult Volunteers.*"

On January 24, 2014, a Type C guidance meeting was held between AstraZeneca and the Division to discuss the proposed design of clinical studies for uncomplicated gonorrhea and a NI margin of 10%. The Division explained that their literature review supported the justification for a 10% NI margin. The Division recommended AstraZeneca consider a comparator arm in their proposed phase 2 trial. The Division also stated that participants from a phase 2 trial could be included to generate safety data if the dose and duration were the same in the phase 3 trial. The Division noted they understood a significant dropout rate was likely in the study population, but AstraZeneca should strive to minimize the number of participants lost to follow-up. The Division requested that AstraZeneca propose an approach for handling participants lost to follow up for the Division's review along with additional sensitivity analyses.

IND (b) (4) for zoliflodacin was submitted on August 28, 2014, by DMID in partnership with AstraZeneca. DMID proposed to conduct a phase 2 clinical trial (DMID 14-0014), titled "*A Randomized, Open-label Phase 2 Study to Evaluate the Efficacy and Safety of a Single Dose of Oral AZD0914 Compared to Intramuscular Ceftriaxone in the Treatment of Male and Female Subjects with Uncomplicated Gonorrhea."*

Ownership of IND 118958, for zoliflodacin (AZD0914) powder for oral suspension, was transferred from AstraZeneca to Entasis on May 20, 2015.

On January 20, 2016, a Type B, End-of-Phase 2 meeting was held between Entasis (with representatives from NIAID) and the Division to discuss the planned phase 3 trial for zoliflodacin. The Division and Entasis discussed PK extrapolation from adults to adolescents and the Division requested information to support the extrapolation. Additionally, the Division commented that they had concerns with a potential increase in bacterial resistance in the future and that the proposed dosage of (b) (4) may not be sufficient. Entasis stated it would not finalize the dosage until efficacy and microbiology data from the phase 2 trial were thoroughly reviewed. The Division advised Entasis that conducting an open-label study for its planned phase 3 trial could potentially create bias that may impact interpretation of the trial results.

On March 11, 2016, Entasis submitted their iPSP. The iPSP included a waiver request for evaluation of zoliflodacin in pediatric patients from birth to (b) (4) of age. Entasis stated in the iPSP that participants aged (b) (4) years of age would be enrolled in the phase 3 trial. On August 19, 2016, the Division communicated agreement with Entasis regarding the iPSP.

On April 11, 2018, the Drugs for Neglected Diseases initiative (DNDi), acting through the GARDP Foundation, submitted a Type B pre-IND meeting with the Division to discuss the development of zoliflodacin under PIND 139105.This submission also contained information that DNDi-GARDP Foundation had entered into an agreement with Entasis Therapeutics to collaborate on the clinical development of zoliflodacin. During the meeting, the Division reiterated their recommendation regarding dual therapy as the comparator in the proposed phase 3 noninferiority trial with zoliflodacin. The Division referenced Centers for Disease Control and Prevention treatment guidelines and noted that dual therapy represents the established standard of care, with approximately 80% of U.S. healthcare providers prescribing dual therapy for gonococcal infections in the United States. The Division indicated that the DNDi-GARDP Foundation partnership would need to provide justification showing that antimicrobial resistance patterns among *N. gonorrhoeae* isolates in the United States are similar to those observed in the proposed global regions where the clinical trials would take place. IND 139105 for zoliflodacin was submitted on August 31, 2018 by the DNDi-GARDP Foundation partnership to conduct the phase 1 protocol (STI_Zoli002), titled "*A Phase I, Open-Label Study to Investigate Pharmacokinetics, Effect of Food and Safety of a New Immediate-Release Formulation of Zoliflodacin in Healthy Subjects*."

On January 9, 2019, a Type B, End-of-Phase 2 CMC meeting was held between the DNDi-GARDP Foundation partnership and the Office of Pharmaceutical Quality to discuss aspects of the drug substance and granules for the oral suspension formulation of zoliflodacin.

On February 19, 2019, Entasis submitted requests for QIDP and Fast Track designations to IND 118958 for zoliflodacin granules for oral suspension for the indication of treatment of uncomplicated gonorrhea. The Fast Track designation was granted on April 11, 2019, and the QIDP designation was granted on April 16, 2019.

On March 8, 2019, the Division was notified that ownership of IND 139105 was transferred from DNDi to the GARDP Foundation.

On May 7, 2019, the GARDP Foundation submitted a phase 3 protocol (STI_Zoli001), titled "*A Multi-Center, Randomized, Open-label, Noninferiority Trial to Evaluate the Efficacy and Safety of a Single, Oral Dose of Zoliflodacin Compared to a Combination of a Single Intramuscular Dose of Ceftriaxone and a Single Oral Dose of Azithromycin in the Treatment of Patients with Uncomplicated Gonorrhea."*

On June 19, 2019, the Division provided comments to the GARDP Foundation regarding protocol STI_Zoli001. Additionally, the Division recommended lowering the enrollment to 12 years of age. The Sponsor was reminded that an agreed iPSP must be in place before submitting the NDA.

On July 26, 2019, the GARDP Foundation submitted their iPSP under IND 139105. The GARDP Foundation noted that its plan was consistent with the agreed upon iPSP submitted by Entasis under IND 118958, which included enrollment of participants aged (b) (4) years of age in the phase 3 trial. The Division held a teleconference call with the GARDP Foundation on

October 29, 2019, to discuss the lower age limit for enrollment. Subsequently, on November 27, 2019, the GARDP Foundation submitted a revised iPSP to include enrollment of pediatric participants down to 12 years of age in the phase 3 trial. On December 17, 2019, the Division communicated their initial agreement with the iPSP.

On December 21, 2020, the Division provided comments to a Type C Written Responses Only meeting request by the GARDP Foundation to discuss the design of a phase 1 bioequivalence study, STI_Zoli003, titled, "*A Phase 1, Single-Dose, Open-Label, Randomized, 4-way Crossover Zoliflodacin Bioequivalence Study of the Reference Product (ZoliPa) with the Test Product (ZoliDr) in Healthy Adult Volunteers Under Fasted and [Specific] Fed Conditions Paired with an Investigation of Effect of Cytochrome P450 3A4 Inhibition by Itraconazole on 3 g Zoliflodacin (ZoliPa) Single-Dose Pharmacokinetics."* The Division's comments included aspects of zoliflodacin's drug-food interactions and drug-drug interactions.

On April 16, 2021, the GARDP Foundation submitted a request for a Type C guidance meeting to discuss changes in the statistical analysis plan for the phase 3 trial, STI_Zoli001. The Division sent preliminary meeting comments on June 28, 2021. The Division stated that they did not agree with GARDP Foundation's proposal to revise the NI margin to (b) (4). Additionally, the Division noted that while this margin has been used in other indications, it would not be clinically acceptable to approve a therapy for uncomplicated gonorrhea that may be substantially less effective than existing treatment options. After reviewing the preliminary meeting comments and determining no further discussion was warranted, the GARDP Foundation cancelled the meeting on July 1, 2021.

On October 11, 2022, a Type C guidance meeting was held between the GARDP Foundation (with representatives from Entasis and NIAID) and the Division to discuss the margin to demonstrate NI of zoliflodacin to the comparator regimen. During the meeting, the GARDP Foundation discussed their proposal to use a NI margin of (b) (4). The Division stated that they were still reluctant to approve a therapy that could potentially be less effective than therapies currently available for uncomplicated gonorrhea given the public health implications of treatment failure. The Division advised the GARDP Foundation that if they were considering a limited use indication, they would need to consider how best to define the participant population with limited treatment options.

On May 2, 2023, the GARDP Foundation submitted a request for clarification to the Division's meeting minutes dated November 10, 2022, for the Type C meeting held on October 11, 2022. On May 24, 2023, the Division clarified that if a 12% NI margin is prespecified, but a 10% NI margin is met, it would not preclude consideration for a full indication in labeling. However, if a 12% NI margin is prespecified and the trial only meets this margin, the Division would consider an indication for a limited population as a review issue. Additionally, the Division noted if a 10% NI margin is prespecified and not met, it could be a significant review issue.

On July 12, 2024, Entasis submitted an amended iPSP to IND 118958 to align with its collaborative partner, the GARDP Foundation's agreed iPSP under IND 139105. The iPSP amendment lowered the enrollment age from (b) (4) to 12 years of age. On October 10, 2024, the Division issued an Amended Agreed iPSP – Agreement letter to Entasis.

On September 10, 2024, a pre-NDA meeting was held between Entasis and the Division to discuss the submission of an NDA for zoliflodacin for the treatment of uncomplicated gonorrhea caused by *N. gonorrhoeae* in adults and adolescents. During the meeting, the Division stated

they agreed with Entasis' plan to submit data from the phase 2 and 3 trials with the proposed nonclinical package. However, the Division indicated that the determination of whether one adequate and well-controlled phase 3 trial combined with the proposed confirmatory evidence from the phase 2 trial would constitute substantial evidence of effectiveness would be a review issue. Entasis stated that their pivotal trial did not focus on a limited population and they planned to submit the NDA with an indication for the treatment of uncomplicated gonorrhea caused by *N. gonorrhoeae* in adults and adolescents. The Division acknowledged that based on the topline data provided in the meeting package it appeared that the phase 3 trial met the prespecified NI margin but emphasized that zoliflodacin was nominally statistical inferior to the comparator arm of ceftriaxone plus azithromycin, which would be a review issue. Additionally, Entasis confirmed that a risk evaluation and mitigation strategy was not being proposed. There were no agreements for late submissions and a complete application was expected at the time of NDA submission.

Entasis submitted a Proprietary Name Review request for "NUZOLVENCE" on September 30, 2024, and received a Proprietary Name Request Conditionally Acceptable letter from the Division of Medication Errors and Prevention Analysis on March 13, 2025.

On November 18, 2024, a dedicated CMC, Type B, Pre-NDA meeting was held between Entasis and OPQ to discuss the CMC section of the planned NDA for zoliflodacin. OPQ acknowledged Entasis' planned submission of a use-related risk analysis for zoliflodacin granules co-packaged with a mixing container ((b) (4) Convenience Kit).

On April 15, 2025, Entasis Therapeutics Inc. (Applicant) submitted NDA 219491 for zoliflodacin, granules for oral suspension, for the treatment of uncomplicated gonorrhea due to *N. gonorrhoeae* in adult and pediatric patients 12 years and older, weighing at least 35 kg. The Applicant requested the tradename "NUZOLVENCE." The NDA, submitted under 505(b)(1), is a New Molecular Entity being reviewed under The Program. The application has both Qualified Infectious Disease Product and Fast Track designations. The application was granted a priority review and filed on June 14, 2025. The NDA has a Prescription Drug User Fee Act goal date of December 15, 2025.

13. Pharmacology Toxicology

13.1. Summary Review of Studies Submitted With the Investigational New Drug Application

13.1.1. Pharmacology

Zoliflodacin (also referred to as AZD0914, AZ13420914, ETX0914, or CZOF11) is a spiropyrimidinetrione bacterial Type II topoisomerase inhibitor that selectively inhibits Type II bacterial topoisomerases, namely deoxyribonucleic acid (DNA) gyrase and topoisomerase IV.

In an in vitro assay comparing ATP-dependent supercoiled DNA relaxation, the inhibitory activity of zoliflodacin against human topoisomerase IIα was compared to etoposide (a drug

targeting human topoisomerase indicated for cancer treatment) and fluoroquinolones. Zoliflodacin had an IC_{50} >30-fold higher than etoposide.

Further details regarding mechanism of action, primary pharmacology, and nonclinical efficacy of zoliflodacin can be found in Section 19 Clinical Microbiology and Section 20 Mechanism of Action/Drug Resistance.

13.1.2. Secondary Pharmacology

AZD0914 was tested using in vitro electrophysiology assays for 7 different human recombinant voltage-gated cardiac ion channels: hCav1.2/β2/α2δ (ICa_L), hCav3.2 (I_{CaT}), hHCN4 (I_f), hKv1.5 (I_{Kur}), hKv4.3/hKChIP2.2 (I_{to}), hKv7.1/hKCNE1 (I_{Ks}) and $hNa_v1.5$ (IN_a). AZD0914 had a mean inhibitory effect >50% in $hK_v4.3$/hKChIP2.2 (I_{to}) of 156.4µM and $hCa_v1.2$/β2/α2δ (ICa_L) of 325.5µM (Study 1168SY). In a second screen, the mean IC_{50} was determined to be 157.1µM for hKv4.3/hKChIP2.2 (hI_{to}), 325.5µM for hCav1.2/β2/α2δ ($hICa_L$), and 171.9µM for $hNA_v1.2$ (Study 2991SV).

13.1.3. Safety Pharmacology

Table 47. Safety Pharmacology Studies

Study/Study No.	Key Methods Highlights and Findings
Study VKS0797 Study Title: AZD0914 Effects on Human Ether-a-go-go-related Gene Encoded Potassium Channel in vitro	CHO cells expressing hERG had current measured and recorded using whole cell patch clamp techniques and the response to incubation with ascending target concentrations of AZD0914 (10, 30, 100, and 300μM) was determined. AZD0914 inhibited the hERG current with an estimated IC_{50} of 449μM (95% CI: 297%, 678%, n=4)
Study 0263SG Study Title: AZ13420914, (b) (4) and (b) (4) Cardiovascular Effects in Anaesthetised Guinea Pigs Following Intravenous Infusion	Male Harlan guinea pigs were administered 90 or 180 mg/kg AZD0914 by 2 consecutive 15 min IV infusions, in a 10% HP-β-CD and 5% dextrose pH 9 vehicle, and cardiovascular parameters of carotid arterial blood pressure, left ventricular pressure, Lead II ECG, PR, RR < QT, QTc, QRS duration, and QA interval were measured. In addition, rectal temperature and plasma concentrations were measured (N=6/group). AZ13420914 decreased blood pressure parameters (systolic at lower dose and diastolic at both the low and high doses), increased heart rate, and decreased QT (uncorrected and corrected). (See Table 48 below).

Table 48. Cardiovascular Parameters, Study 0263SG

	Dose	
	90 mg/kg	180 mg/kg
Parameter	Percent Change	Percent Change
Systolic blood pressure		−27%
Diastolic blood pressure	−26%	−50%
Heart rate	11%	10%
dP/dT_{min}	−20%	−55%
QT interval	−9%	−11%
QTcB interval	−5%	−7%

Source: Reviewer created table
Abbreviation: dP/dTmin, index of the rate of cardiac relaxation

Recovery from these changes did not occur during the 30-minute washout phase.
Peak total plasma concentrations were 447.27 and 1065.18 μmol/L following administration of 90 and 180 mg/kg AZ13420914, respectively.

Study/Study No.	Key Methods Highlights and Findings
Study No. 05622 Study Title: AZ13420914-004 on Cardiovascular Parameters of Male Han Wistar Rats with Simultaneous Pharmacokinetic Sample Collection	Male Han Wistar rats were administered 0 (vehicle control), AZ13420914-004 (intended doses of 100 (low dose) and 200 mg/kg (high dose), and a second 200 mg/kg dose) by 50 min IV infusion in a crossover design (N=4/group). Group 1 received vehicle, vehicle, vehicle (set 1), low dose, high dose, high dose (set 2). Group 2 received low dose, high dose, high dose (set 1), vehicle, vehicle, vehicle (set 2). Actual doses were 85.5 and 119 mg/kg for the low and high doses, respectively. Treatments were 4 hours apart and treatment sets were 12 hours apart. Observations for appearance, behavior, morbidity, and mortality were made and measurements of cardiovascular telemetry data and toxicokinetic parameters were reported. No morbidity, mortality, or clinical signs of toxicity were observed during the study. Mean C_{max} was 97.2μM after the 85.5 mg/kg dose, 173.0μM after the first 119 mg/kg dose, and 217.0μM after the second 119 mg/kg dose. No consistent changes in body temperature or cardiovascular parameters were reported.
Study No. 1348ZD Study Title: AZD0914 Cardiovascular Effects in Conscious, Telemetered Beagle Dogs following Single Intravenous Administration	In an escalating dose design, male Beagle dogs were administered 0 (vehicle control), 20, 50, or 100 mg/kg AZD0914 by 1 hour IV infusion and systolic, diastolic, and mean arterial blood pressures, heart rate, left ventricular systolic and end-diastolic pressures, left ventricular dP/dt+ (index of cardiac contractility) and dP/dt− (index of cardiac relaxation), body temperature, and lead II ECG parameters: PR, QA, QT interval, and QRS duration were measured (N=4). No cardiac effects were reported at 20 or 50 mg/kg 100 mg/kg (group means): Systolic and diastolic blood pressures were decreased 22% to 25%, and heart rates increased up to 40%, beginning at 30 minutes, with peak effect at 2 hours. Blood pressures recovered to Baseline at 12 hours and heart rates at 4 hours. The increases in heart rates were associated with decreased PR interval (1.5 to 3 hours, up to 11%), decreased QA interval (0.5 to 3 hours, up to 8%) and decreased QT interval (1.5 hours, 8%); however, corrected QT was increased by about 6% at the 2-hour timepoint. Cardiac contractility increased by up to 24% at 1.5 to 2 hours, peaking at the latter timepoint. Cardiac relaxation decreased by up to 18%, peaking at 2.5 hours, but the effect was observed by the 45-minute timepoint. The contractility and relaxation indices had both returned to Baseline at the 4-hour timepoint. Transient decreases in body temperature (not exceeding a 1.8% change) were observed beginning at 1.5 hours through 12 hours after dosing. The postural challenge showed that the high dose of AZ13420914 caused orthostatic hypotension, with marked reduction of tilt-evoked increases in systolic and diastolic blood pressures seen at the 1-hour timepoint, with recovery by 20 hours. Toxicokinetics: T_{max} for all three groups was 0.917 hours. C_{max} was 43.2, 130, and 319μM, for 20, 50, and 100 mg/kg dose groups, respectively.

Study/Study No.	Key Methods Highlights and Findings
Study No. 611610 Study Title: AZD0914 Respiratory Effects in the Han Wistar Rat following Single Intravenous (1h) Infusion Administration	Male Han Wistar rats were administered 0 (vehicle control), 25, 100, or 250 mg/kg AZD0914 or 5 mg/kg baclofen by 1 hour IV infusion and respiratory effects were measured using whole body plethysmography before and after dosing and plasma concentrations were measured at 4 hours (N=8/group). AZD0914 did not appear to have any effects on respiratory parameters. Baclofen (positive control) significantly reduced respiratory rate, minute volume, and peak inspiratory flow, and increased inspiration and expiration times. Piloerection and hunched posture were observed in 5/8 rats treated with 250 mg/kg of AZD0914 beginning about 1 hour 45 minutes after dosing and lasting through the end of the experiment. Mean plasma concentrations at 4 hours were 0, 3.51, 70.7, and 255 µMol/L for 0, 25, 100, and 250 mg/kg dosing, respectively.
Study No. 2945SV Study Title: AZ13420914 Effects on Synaptic Transmission in the Mouse Hippocampal Brain Slice in vitro	The effects of AZ13420914 on evoked population spike responses in mouse hippocampal brain slices (C57BL6 mouse strain, 6 slices from 2 brains were tested) was tested up to a concentration of 100µM. AZ13420914 did not change synaptic activity or cell excitability, reducing concern that the substance has a pro-convulsant effect.
Study No. 611605 Study Title: AZD0914 Effects in the Irwin Screen after Single Intravenous (1h) Infusion Administration in Male Han Wistar Rats	Male Han Wistar rats were administered 0 (vehicle control), 25, 100, or 250 mg/kg AZD0914 by 1 hour IV infusion and Irwin screen was conducted, recording behavioral observations, changes in bodily functions, body temperatures, and body weights before and 24h after administration; plasma concentration was measured at 2 hours (N=6/group). At 25 mg/kg: no significant observations. At ≥100 mg/kg, a reduction in body weight was observed. At 250 mg/kg, statistically significant decreases were seen in pupil size (all time points, $p<0.01$, except for $p<0.001$ at 24 hours) and body temperature (15 minutes, $p<0.01$). Other Irwin parameters at 250 mg/kg that were considered of biological significance were rolling gait (15 to 120 minutes), abnormal urination (120 minutes), hunched posture (30 to 120 minutes), decreased grip strength (60 to 120 minutes), decreased spontaneous activity (30 minutes), and decreased traction response (15-, 60-, and 120-minute timepoints).

Study/Study No.	Key Methods Highlights and Findings
Study No. 611626 Study Title: AZD0914 Renal Function-Diuresis and Electrolyte Excretion in the Han Wistar Rat following Single Intravenous (1h) Infusion Administration	Male Han Wistar rats were administered 0 (vehicle control), 25, 100, or 250 mg/kg AZD0914 by 1 hour IV infusion or 30 mg/kg furosemide by oral gavage (positive control) and renal function parameters were measured in urine samples for 0 to 4 hours and 4 to 24 hours, water consumption was measured, and blood samples were collected 2 days before dosing, and 4 and 24 hours postdosing (N=8/group). • 0 to 4 hours: 250 mg/kg: urinary pH, plasma glucose, and rate of water consumption increased. Decreased urinary calcium excretion, and decreased plasma calcium and total protein were observed. 100 mg/kg and above: decreased urinary potassium excretion, decreased renal potassium clearance and decreased plasma sodium All treated groups: decreased urinary potassium expressed as a ratio with creatinine • 4 to 24 hours: 250 mg/kg: decreased urinary calcium excretion and decreased renal clearance, increased plasma creatinine, decreased plasma sodium. 100 mg/kg and above: increased water consumption and free water clearance. All treated groups: increased urinary rate and decreased urine osmolality, decreased urine creatinine. Furosemide at both time points showed a diuretic effect, as expected. Conclusion: AZD0914 induced diuresis at all 3 doses tested in this study, especially during the 4- to 24-hour period. The diuretic effect was dose related.

Source: Reviewer table from Applicant's study report data
Abbreviations: AZ13420914, zoliflodacin; (b) (4), zoliflodacin; AZD0914, zoliflodacin; CHO, Chinese hamster ovary; C_{max}, maximum plasma concentration; ECG, electrocardiogram; hERG, human ether-à-go-go-related gene; HP-β-CD, 2-Hydroxypropyl-β-cyclodextrin; IV, intravenous; QTc, corrected QT interval; T_{max}, time to maximum concentration

13.1.4. Absorption, Distribution, Metabolism, Excretion/Pharmacokinetics

Absorption

Bioavailability of orally administered zoliflodacin was calculated to be about 46% in CD-1 mice, 58% in cynomolgus monkeys, 28% in Han Wistar rats, and 42% to 95% in Beagle dogs (depending on the formulation).

In vitro apparent permeability of zoliflodacin across human jejunum and Caco-2 confluent cell layers were tested in vitro with tissues from two donors. There was variability in the permeability by donor across the jejunum tissue with apical to basal transport (i.e., gut to bloodstream absorption) of zoliflodacin characterized as moderate in tissue from one donor and high in the other. The permeability of zoliflodacin in basal to apical transport (i.e., active efflux back into the gut lumen) was generally higher than apical to basal in both test systems and was dose dependent.

Distribution

Transporters and Protein Binding

Plasma protein binding of zoliflodacin in vitro using human, dog, rat, mouse and guinea pig proteins was approximately 83%, 80%, 78%, 92%, and 74%, respectively, and was similar across concentration tested from 1 to 50µM.

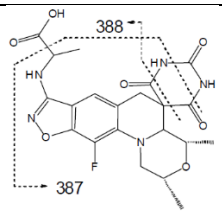
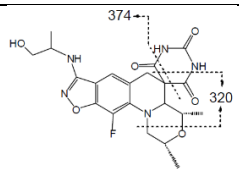
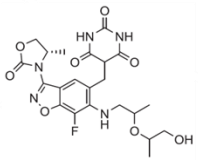
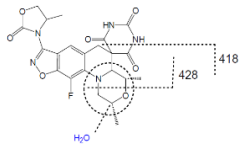
Zoliflodacin is a substrate of p-glycoprotein and an inhibitor with an IC_{50} ranging from 30 to 300µM in different in vitro studies with different substrates.

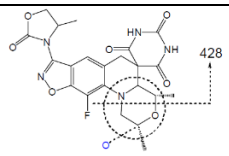
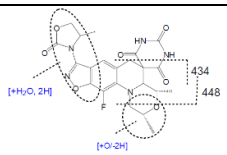
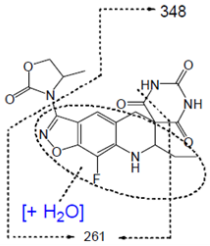
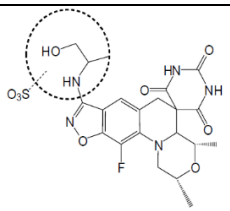
Increased influx of zoliflodacin was found in HEK293 cells expressing OATP1B1 or OATP1B3 indicating that zoliflodacin is a substrate of these intake transporters. Zoliflodacin also inhibited OATP1B1 with an IC_{50} of 7.7µM.

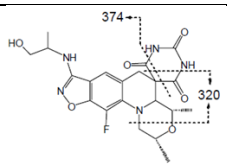
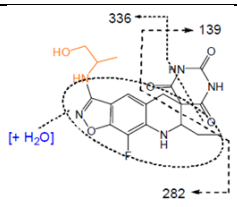
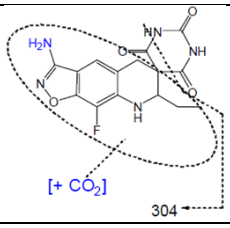
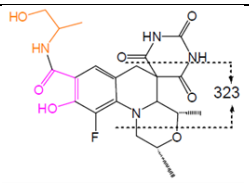
Metabolism

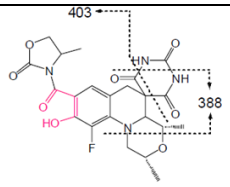
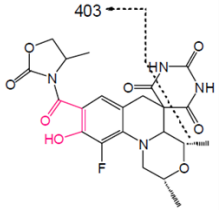
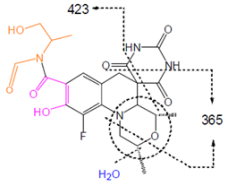
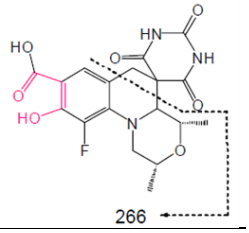
Metabolites are denoted with the notation "M" followed by a number. Structures of the metabolites can be found in Table 49 below.

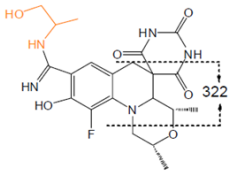
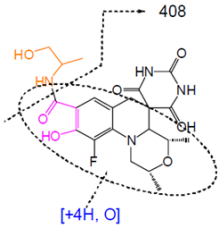
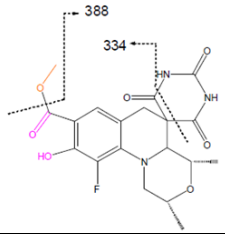
Table 49. Metabolite Structures

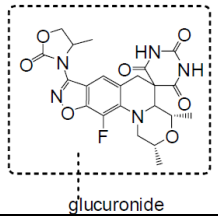
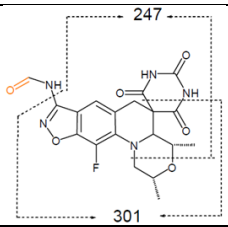
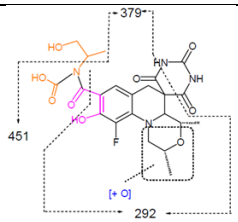
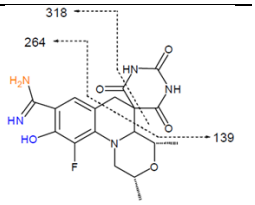
Metabolite Designation	M1	M2	M3	M4
Structure				

Metabolite Designation	M6	M7	M8	M9
Structure				

Metabolite Designation	M10	M11	M12	M13
Structure				

Metabolite Designation	M14	M15	M16	M17
Structure				

Metabolite Designation	M18	M19	M20	M21
Structure				Unknown

Metabolite Designation	M22	M23	M24	M25
Structure				

Source: Reviewer generated table, copied images from submitted documents
Abbreviation: M, metabolite

In Vitro

Metabolism in vitro using human hepatocytes was predominantly by CYP3A4 (68%) with metabolism also contributed by CYP1A2 (14%), CYP2C9 (10%), CYP2C8 (5%), and CYP2C19 (3%).

Zoliflodacin in vitro inhibited CYP2C8 with an IC_{50} of 137µM, CYP2C9 with an IC_{50} in the range of 217 to 274µM, and CYP2C19 in the range of 251 to 331µM.

Metabolism of ^{14}C-radiolabeled zoliflodacin with recombinant human enzymes was with only CYP3A4 and CYP3A5 and formed M2 and M6.

Metabolism of ^{14}C-radiolabeled zoliflodacin by hepatocytes from mouse, rat, dog, or human livers was tested. The major metabolites from mouse and rat hepatocytes were identified as M1, M2, M4, M6, and M9. With dog hepatocytes, metabolites observed were M1, M2, M4, and M22. In human hepatocytes only trace metabolites M1, M2, and M6 were observed.

In Vivo

In mice administered IV zoliflodacin, plasma clearance of the parent was reduced 5-fold, and the $t_{1/2}$ was increased 5-fold with pretreatment of 1-aminobenzotiazole (ABT, a CYP450 inhibitor), indicating that CYP450 enzymes have a role in clearance in the mouse.

In male CD-1 mice administered single 30-minute intravenous infusions of 25 mg/kg ^{14}C-radiolabeled zoliflodacin, concentrations of zoliflodacin and radiolabeled metabolites were measured in plasma, bile urine, and feces, as shown in Table 50 below. Metabolites were also measured in animals that had been pretreated with ABT.

Table 50. Mouse Metabolites

Sample (% of Total)	ABT Status[a]	Metabolite (% of Dose)
Plasma (0.5 hours post administration)	No	Parent (88.9%), M2 (3.2%), M3 (2.1%)
	Yes	Parent (89.1%), M3 (1.3%)
Liver (2 hours post administration)	No	Parent (61%), M3 (2.2%), M7 (1.8%), M12 (2.2%), M14 (13.8%), M15 (4.0%), M18 (2.1%), M25 (3.4%)
	Yes	Parent (81.2%), M3 (1.2%), M14 (10.9%), M15 (1.7%%)
Urine	No	Parent (2.9%), M1 (0.3%), M2 (0.4%), M3 (1.2%), M4 (0.3%), M6 (0.5%), M7 (1.0%), M12 (0.4%)
	Yes	Parent (3.7%), M3 (2.2%), M7 (1.4%)
Feces	No	Parent (0.5%), M3 (1.6%), M13 (3.1%), M14 (3.3%), M15 (9.3%), M16 (3.1%), M18 (3.3%), M23 (8.7%), M24 (11%), M25 (22.1%)
	Yes	Parent (4.1%), M3 (2%), M13 (8.5%), M14 (5.9%), M15 (21.4%), M16 (6.6%), M18 (3%), M24 (16.1%), M25 (2.9%)

Source: Reviewer generated table from submitted data
[a] ABT 1-aminobenzotiazole, a CYP450 inhibitor
Abbreviation: M, metabolite

In male Han Wistar rats administered single 30-minute intravenous infusions of 50 mg/kg ^{14}C-radiolabeled zoliflodacin, concentrations of zoliflodacin and radiolabeled metabolites were measured in plasma, bile urine, and feces, as shown in Table 51 below.

Table 51. Rat Metabolites

Sample (% of Total)	Metabolite (% of Dose)
Plasma (100%)	Parent (89.7%), M3 (10.3%)
Urine (15%)	M3 (4.1%), M2 (2.7%), M4 (2.6%), Parent (2%), M6 (0.9%), M10 (0.8%), M1 (0.6%), M7 (0.6%), M8 (0.6%),
Bile (47.9%)	M3 (11.8%), M9 (8.4%), M2 (8.0%), M4 (4.8%), M10 (4.4%), M1 (4.2%), M13 (2.1%), Parent (1.3%), M12 (1.2%), M8 (1.1%), M11 (0.8%),
Feces (22.1%)	M17 (6.4%), M15 (7.3%), M14 (2.9%), M13 (2.3%), M16 (1.0%), M18 (1.0%), M20 (0.6%), M19 (0.5%),

Source: Reviewer generated table from submitted data
Abbreviation: M, metabolite

Metabolites M13, M14, M15, and M17 could be detected when the parent was incubated with blank rat feces for 20 hours, which would suggest that these products are microbially derived in the gut.

Excretion

Mean total recovery of radioactivity from bile-duct cannulated male and female Han Wistar rats orally administered ^{14}C-radiolabeled zoliflodacin, was monitored for 48 hours after dosing. Radioactivity was recovered with 52% in the bile, 38% in the feces, 15% in the urine, and 3% in the cage wash.

Mean total recovery of radioactivity from male Han Wistar rats administered ^{14}C-radiolabeled zoliflodacin IV was monitored for 48 hours after dosing to bile-duct cannulated rats. The major route of excretion was biliary elimination (55%), followed by feces (24%), and urine (15%).

Mean total recovery of radioactivity from male CD-1 mice administered ^{14}C-radiolabeled zoliflodacin IV was measured at time points up to 6 hours after dosing. Recovery of radioactivity was mostly in feces (83% to 85%), followed by urine (9%), and cage wash (2% to 4%).

Pharmacokinetics

Pharmacokinetics were evaluated in general toxicology studies and are included with the study reviews in Section 13.1.5.

13.1.5. Toxicology

13.1.5.1. General Toxicology

Repeat Dose Toxicology/Toxicokinetic Studies

Study 523980/AZ13420914: One Month Oral (Gavage) Toxicity Study With a Three Month Assessment of Recovery in the Rats (Good Laboratory Practice)

Key Study Findings

Dose-related microscopic findings were present in the male reproductive tissues. Findings included minimal to mild tubular degeneration observed in the testes of 8/10 male rats administered 1000 mg/kg/day (high dose), and minimum to mild cellular debris in the epididymides of 5/10 male rats administered 500 mg/kg/day (mid dose) and 10/10 rats administered the high dose. Mild to moderate secretory depletion in the seminal vesicles was observed in 10/10 males administered the high dose, 5/10 males in the mid-dose group, and 1/10 in the 200 mg/kg/day (low dose) group. Persistence of these findings in one of four males from the recovery group (minimal to mild) suggested incomplete reversibility. The mild to moderate secretory depletion observed in the seminal vesicles at end of dosing was no longer present at the end of recovery and was not associated with degeneration of the tissue; the nondepleted areas of secretory epithelium/submucosa appeared similar to controls.

Changes in red blood cell parameters (reduced hematocrit, hemoglobin; increased reticulocytes) and mild increased hematopoiesis in the spleen, were observed in high dose animals. These

findings likely indicate increased turnover of red blood cells and/or increased removal from circulation, though the mechanism was not evident in this study.

No-observed-adverse-effect level (NOAEL) was 200 mg/kg/day corresponding to Day 28 sex-combined exposures of AUC_{0-t} 1074 µg*h/mL and C_{max} of 107 µg/mL.

Table 52. Information, Study 523980

Study Features and Methods	Details
GLP compliance	Yes
Dose and frequency of dosing	0 (vehicle), 200, 500, and 1000 mg/kg/day
Route of administration	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Rat/Han Wistar
Number/sex/group	10/sex/group main study
Age	9 to 10 weeks at the start of dosing
Satellite groups/unique design	5/sex/group (0 and 1000 mg/kg only) 3/sex/group (TK group) Drug was administered daily for 28 days, until the day before scheduled sacrifice of main study animals. Recovery animals were sacrificed 91 days after the final dose.
Deviation from study protocol affecting interpretation of results	None

Source: Reviewer table from Applicant's study report data
Abbreviations: GLP, good laboratory practice; HCl, hydrochloride

Table 53. Observations and Results, Study 523980

Parameter	Major Findings
Mortality	2 deaths due to gavage errors (high dose females on Day 2 and Day 14)
Clinical signs	Salivation: some low dose animals, all mid and high dose animals Plowing behavior: high dose animals starting on Day 7, mid and low dose animals started on Days 12-17.
Body weights	No test-article related differences
Feed consumption	No test-article related differences
Water consumption	Dose-dependent increase in water consumption At 1000 mg/kg/day, mean water consumption was about double that in the controls. At 500 mg/kg/day, mean water consumption was 50% to 60% higher in females and 25% higher in males. There was no effect in the 200 mg/kg/day dose group.
Ophthalmoscopy	No test-article related differences

Parameter	Major Findings
Hematology	At the end of dosing:

Table 54. Changes in Hematology Parameters

Dose	200		500		1000	
Sex	**M**	**F**	**M**	**F**	**M**	**F**
Metric						
Mean Hb concentration			↓(−5%)	↓(−6%)	↓(−10%)	↓(−8%)
Mean HCT		↓(−4%)	↓(−8%)	↓(−6%)	↓(−11%)	↓(−6%)
Mean RBC count					↓(−6%)	
MCV			↓(−5%)		↓(−5%)	
MCH					↓(−4%)	
MCHC						↓
Reticulocyte counts		↑(+43%)		↑(+59%)	↑(+53%)	↑(+92%)
Mean platelet count					↑(+18%)	

Source: Reviewer generated table from study report data
Abbreviations: ↓, statistically significant decrease compared to controls; ↓, statistically significant increase compared to controls; Hb, hemoglobin; HCT, hematocrit; MCH, mean cell hemoglobin; MCHC, mean cell hemoglobin concentration; MCV, mean cell volume; RBC, red blood cell

At the end of recovery, high mean red cell distribution width was seen in high dose rats of both sexes and red cell count in the high dose females was higher than control.

Changes in white cell counts were reported in both sexes in the 1000 mg/kg/day group at the end of dosing but not the end of recovery. Other inflammatory signs were not reported, so the significance is unclear.

Parameter	Major Findings
Clinical chemistry	Clinical chemistry parameters reported at the end of dosing are as shown in the table below.

Table 55. Changes in Clinical Chemistry Parameters at the End of Dosing

Dose	200		500		1000	
Sex	**M**	**F**	**M**	**F**	**M**	**F**
Metric						
ALP				↑(+33%)	↑(+27%)	
ALT					↑(+26%)	↑(+41%)
Direct bilirubin concentration					↑(+60%)	↑(+29%)
Indirect bilirubin*			+	+	+	+
Cholesterol			↑(29+%)	↑(32+%)	↑(52+%)	↑(56+%)
triglycerides			↓(−35%)	↓(−43%)	↓(−39%)	↓(44-%)
Albumin	↓(−6%)		↓(−6%)	↓(−9%)	↓(−5%)	↓(−10%)
Globulin			↓(−18%)		↓(−17%)	
Albumin: globulin concentration			↑(+15%)		↑(+15%)	↓(−11%)
Total protein concentration	↓(−8%)		↓(−10%)	↓(−6%)	↓(−9%)	↓(−6%)
Mean creatinine			↑(22+%)	↑(26+%)	↑(24+%)	↑(36+%)
Mean urea					↑(38+%)	

Source: Reviewer generated table from study report data
* Indirect bilirubin was detected in one male and two females from the mid dose group and in 11 males and 12 females from the high dose group.
Abbreviations: ↓, statistically significant decrease; ↑, statistically significant increase; +, detected; ALP, alkaline phosphatase; ALT, alanine aminotransferase

At the end of the recovery period, clinical chemistry parameters were similar between control and high dose rats, with the exception that mean plasma creatinine in high dose males was still slightly greater than control.

Urinalysis	End of dosing: 1000 mg/kg/day group: increased urine volume, decreased urinary creatinine concentration, lower specific gravity (females) 200 mg/kg and above group: decreased urinary protein concentration (males), low protein: creatinine ratio End of recovery: 1000 mg/kg/day group: elevated specific gravity (females)

Parameter	Major Findings
Gross pathology	"Material accumulation" was observed in the cecum of one rat/sex in the low and mid dose groups and 4/sex in the 1000 mg/kg/day dose group. This may represent unabsorbed drug. Small seminal vesicles were observed in two 1000 mg/kg/day dose males. At the end of the recovery period, flaccid testis was observed in one 1000 mg/kg/day male.
Organ weights	In high dose males, decreased mean relative epididymides weight and increased mean relative kidney and spleen weights were observed. These differences were not seen at the end of the recovery period.
Histopathology Adequate battery: Yes	Testes: 1000 mg/kg/day: Testicular degeneration (minimal to mild, dose related) 500 and 1000 mg/kg/day: Epididymal cellular debris (minimal to mild, dose related) 200 mg/kg/day and higher: secretory depletion of seminal vesicles (mild to moderate, dose related) Salivary glands: Mandibular glands: acinar hypertrophy (minimal to moderate, dose related) Parotid glands: acinar basophilia (minimal to moderate, dose related) Peritoneal fibroplasia (minimal to mild) adjacent to the mesenteric lymph nodes or pancreas was observed in a few mid and high dose rats. Increased hematopoiesis in the spleen (likely compensatory to the decrease in RBC parameters) Mucosal hyperplasia (minimal to mild) in the cecum or rectum of some mid and high dose rats (may be related to the accumulation of material in this region)

Source: Reviewer table from Applicant's study report data
Abbreviation: RBC, red blood cell

Toxicokinetics

AZ13420914 was below the limit of quantitation (0.02 µmol/L) in control animals.

Similar toxicokinetics were observed between males and females.

Increases in exposure were less than dose proportional.

Exposure (C_{max} and AUC) was between 1.1- to 2.2-fold greater on Day 28 than Day 1, with the difference slightly larger at the high dose compared to the low dose.

Table 56. TK Parameters, Study 523980

		Dose (mg/kg/day)					
		200		500		1000	
Parameter	**Sex**	**Day 1**	**Day 28**	**Day 1**	**Day 28**	**Day 1**	**Day 28**
AUC_{0-t} (µg*h/mL)	Male	834	1068	1813	2364	2096	3485
	Female	990	1082	1818	2569	2510	4158
C_{max} (µg/mL)	Male	83.8	94.6	128.7	173.0	113.1	229.1
	Female	95.5	118.9	124.3	161.3	127.2	283.2
T_{max} (h)	Male	2	2	4	4	4	4
	Female	2	2	2	2	6	6

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study 523996/AZ13420914: 28 Day Oral (Gavage) Toxicity Study With a 3 Month Assessment of Recovery in Dogs (Good Laboratory Practice)

Key Study Findings

One male dog administered 500 mg/kg/day AZ13420914 was euthanized early for low feed consumption and body weight loss. The cause of morbidity in this animal appeared to be due to marked necrosis and inflammation of the ileum and gut-associated lymphoid tissue.

Dose-related increases in incidences of vomiting and salivation were seen in drug-treated animals (all groups).

Treatment with AZ13420914 was associated with increased heart rate (all groups), but not QTc prolongation.

Mean reticulocyte counts were higher in all groups treated with drug compared to pretreatment values and controls. Red blood cell (RBC) parameters were reduced slightly in mid- and high-dose groups and mean bilirubin levels were slightly increased in these groups. These values recovered by the end of a 3-month drug-free period.

Microscopic changes comprised of moderate testicular degeneration and cellular debris in the epididymis of a high-dose dog and mild testicular degeneration in a mid-dose dog were observed. Hypospermatogenesis was observed in 2 dogs in the high-dose group at the end of the recovery period.

The NOAEL in this study was the low dose of 100 mg/kg/day, based on the reduced RBC parameters, increased bilirubin, and testicular degeneration seen in the mid dose group (200 mg/kg/day).

Table 57. Information, Study 523996

Study Features and Methods	Details
GLP compliance	Yes
Dose and frequency of dosing	0 (vehicle), 100, 200, and 500 mg/kg/day once daily
Route of administration	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Dog/beagle
Number/sex/group	3/sex/group
Age	10 to 12 months
Satellite groups/unique design	Three/sex in control and high dose groups for recovery (but only two females in control recovery group due to prestudy rejection of an animal due to seizure)
Deviation from study protocol affecting interpretation of results	None

Source: Reviewer table from Applicant's study report data
Abbreviations: GLP, good laboratory practice; HCl, hydrochloride

Table 58. Observations and Results, Study 523996

Parameter	Major Findings
Mortality	One 100 mg/kg/day group male was found dead before dosing on Day 13. No cause of death could be determined. One 500 mg/kg/day group female was euthanized on Day 15 following reduced feed consumption and weight loss. Necropsy showed necrosis and inflammation of the ileum and gut associated lymphoid tissue (GALT) (considered to be the cause of morbidity); these findings were also observed in the cecum and rectum.
Clinical signs	A dose-related increase in vomiting was observed in test article-treated groups. Occasional increase in salivation was reported in drug-treated groups with a dose-related increase in incidence. Green or yellow discolored feces, sometimes with mucoid material, was observed with a dose-related increase in incidence.
Body weights	No test article-related differences
Feed consumption	No test article-related differences
Water consumption	No test article-related differences
Electrocardiography	Days measured: 2, 23, 27 Heart rate was increased in all test article-administered dose groups starting at 4 hours after administration and continuing for the remainder of the 20-hour measurement period. No drug-related changes to the PR, QRS, QT, or QTc (individual rate correction) intervals were observed.
Hematology	Mean red cell parameters (hemoglobin, hematocrit, RBC count) were slightly reduced in mid-dose males and high-dose dogs of both sexes beginning in Week 3 (males: 6% to 16%, females: 9% to 11%). Mean reticulocyte counts in Week 4 were slightly elevated in all groups treated with AZ13420914. Mean platelet counts were elevated in mid-dose females during Week 3 and in mid- and high-dose dogs of both sexes in Week 4. No test article-related differences were reported at the recovery measurements.
Clinical chemistry	Group mean total bilirubin levels were slightly elevated in mid- and high-dose dogs by Week 3. Indirect bilirubin was detected in some of the high dose dogs. At the end of recovery, changes in bilirubin recovered to pretreatment levels.
Urinalysis	No test article-related differences
Gross pathology	No test article-related differences
Organ weights	No test article-related differences

Parameter	Major Findings
Histopathology Adequate battery: Yes	At the end of dosing: 1 high dose male had moderate testicular degeneration and moderate cellular debris in the epididymis 1 mid-dose male had mild testicular degeneration At the end of recovery: Hypospermatogenesis was observed in 2 high dose males at the end of recovery. This finding can be a background finding or could be a delay of recovery from a drug effect.

Source: Reviewer table from Applicant's study report data
Abbreviations: AZ13420914, zoliflodacin, QTc, corrected QT interval; RBC, red blood cell

Toxicokinetics

Toxicokinetic parameters were similar between sexes.

The increase in exposure was less than dose proportional, especially between 200 and 500 mg/kg/day, suggesting that the high dose may be beyond the plateau for absorption.

There was evidence of drug accumulation between Days 1 and 28 of dosing, particularly in the mid- and high-dose groups.

Table 59. TK Parameters, Study 523996

		Dose (mg/kg/day)					
		100		200		500	
Parameter	Sex	Day 1	Day 28	Day 1	Day 28	Day 1	Day 28
$AUC_{0\text{-}t}$ (µg*h/mL)	Male	405	751	1111	2457	1146	2608
	Female	600	946	809	1862	1102	2954
C_{max} (µg/mL)	Male	41.7	90.7	94.6	210.1	93.6	203.3
	Female	55.6	90.2	79.5	216.9	98.5	244.7
T_{max} (h)	Male	1	1.5	2	4	4	3
	Female	4	2	2	2	2	4

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study 611830/AZD0914: 14 Day Intravenous (1 h) Infusion Toxicity Study With Assessment of 1 Month Recovery in Rats (Good Laboratory Practice)

Study Summary and Key Findings

Wistar Han rats were administered 0 (vehicle), 75, 150, or 250 mg/kg/day AZD0914 by IV infusion over one hour once daily for 14 days (n=10/sex/group [main group], n=3/sex/group [PK group], and n=5/sex/group for high dose and controls recovery groups to assess reversibility).

Dose-dependent increased water consumption was reported; this was statistically significant for mid- and high-dose females and high-dose males. Water consumption was similar across groups in recovery animals.

Body weight gains were reduced about 40% in test article-administered males. During the recovery period the high-dose animals gained more weight than controls.

Neutrophil counts were reduced 34% to 45% in drug-treated males, but the change was not dependent on dose, and a smaller nonsignificant reduction was observed in treated females. At the end of recovery, the neutrophil counts were about 50% lower than controls.

Mean prostate weight was lower in males treated with the mid- and high-doses of AZD0914 compared to controls (−21% and −27% absolute weights compared to controls, respectively; differences relative to body weights were −19% and −25%, respectively). No histopathological correlates were reported. Absolute prostate weights were still statistically significantly lower at the end of recovery in high dose animals (−28%) but not relative to body weights (−23%).

The reported differences were not adverse in this study and the NOAEL was the highest dose tested of 250 mg/kg/day.

Toxicokinetics

Exposure increased greater than dose-proportionality from the low dose to the high dose on Day 1 for both males and females. Exposures did not appear to increase from the mid- to high dose in females on Day 1.

No plasma accumulation was found.

Table 60. TK Parameters, Study 611830

		Dose (mg/kg/day)					
		75		150		250	
Parameter	**Sex**	**Day 1**	**Day 14**	**Day 1**	**Day 14**	**Day 1**	**Day 14**
$AUC_{0\text{-}t}$ (µg*h/mL)	Male	371	315	980	1102	2735	1901
	Female	415	575	2525	1925	2305	3646
C_{max} (µg/mL)	Male	154	102	189	213	306	232
	Female	96	184	281	275	283	377
T_{max} (h)	Male	1.25	1.25	1.25	1.25	1.25	1.25
	Female	2	1.25	2	1.25	0.917	1.25

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study 611521/AZ13420914: 14 Day Intravenous (1h) Infusion Toxicity Study With Assessment of Recovery in Dogs (Good Laboratory Practice)

Key Study Findings

Beagle dogs (males 11 to 15 months old and females 13 to 17 months old) were administered 0 (vehicle control), 25, 50, or 100 mg/kg/day AZD0914 by IV infusion over one hour once daily for 14 days (n=3/sex/group and an additional 3/sex/group for high dose and control recovery animals).

The incidence and frequency of vomiting was greater in the high-dose group than controls, especially within the first hour after dosing.

Increased heart rate (up to 32%) at 100 mg/kg between 1.5 to 8 hours after infusion, particularly in the male animals, on Days 2 and 13.

Mean bilirubin was increased (about 30%) and mean phosphate was increased (10% to 15%) at 100 mg/kg.

Minimal to mild hypospermatogenesis was seen across all treatment groups, including in a control animal. Minimal unilateral testicular tubular segmental hypoplasia was observed in a few drug-treated animals. As the severity of findings and number of affected animals was similar across groups, they were not considered test article-related in this study.

Table 61. Summary Microscopic Findings, Scheduled Euthanasia Animals (Day 15 and Day 43)

	Males (Day 15)				Males (Day 43)	
Group	1	2	3	4	1	4
Dose (mg/kg/day)	**0**	**25**	**50**	**100**	**0**	**100**
No. animals examined	**3**	**3**	**3**	**3**	**3**	**3**
Testis (No. Examined)	(3)	(3)	(3)	(3)	(3)	(3)
Hypospermatogenesis, bilateral						
Minimal	1	1	2	0	0	1
Mild	0	0	0	1	0	0
Hypospermatogenesis, unilateral						
Minimal	0	0	0	0	0	1
Mild	0	0	0	1	1	0
Hypoplasia, segmental, unilateral						
Minimal	0	1	1	2	0	0

Source: Applicant study report

The reported differences were not adverse in this study and the NOAEL was the highest dose tested of 100 mg/kg/day.

Toxicokinetics

Exposures increased approximately proportionate to increasing dose.

There was no evidence of accumulation of the test article.

Exposures were similar in males and females.

Table 62. TK Parameters, Study 611521

		Dose (mg/kg/day)					
		25		50		100	
Parameter	**Sex**	**Day 1**	**Day 14**	**Day 1**	**Day 14**	**Day 1**	**Day 14**
$AUC_{0\text{-}t}$ (µg*h/mL)	Male	173	183	403	391	868	897
	Female	170	186	451	423	877	1048
C_{max} (µg/mL)	Male	30	29	66	64	135	138
	Female	31	30	69	69	143	147
T_{max} (h)	Male	0.917	0.917	0.917	0.917	0.917	0.917
	Female	0.917	0.917	0.917	0.917	0.917	0.917

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study 3494DR/AZ13420914: Maximum Tolerated Dose and 14 Day Oral Toxicity Study in the Rat (Non-Good Laboratory Practice)

Key Study Methods and Findings

In an maximally tolerated dose study, RccHan:WIST rats were administered 500, 1000, or 2000 mg/kg/day AZ13420914 by oral gavage for 2 days, observed for 5 days and euthanized for necropsy on Day 8 (n=3/sex).

One high-dose female was euthanized for humane reasons due to clinical signs of pilo-erection, cold to touch, collapse, hunched posture and extremely decreased motor activity.
Increased reticulocyte counts were seen in all drug-treated female rats.
Histopathology included moderate extramedullary hematopoiesis seen in the spleens of females treated with 2000 mg/kg, accompanied by a moderate increase in pigmented macrophages in one of these rats. In the sternum, an increase in the myeloid:erythroid ratio was seen in the bone marrow of one male treated with 2000 mg/kg and mild sinus dilatation of the bone marrow was seen in a female treated with 2000 mg/kg. A minimal dose-related exfoliation of germinal epithelial cells was seen in the head of the epididymides in males treated with 1000 and 2000 mg/kg.

In a repeat-dose study RccHan:WIST rats were administered 0, 250, 500, or 1000 mg/kg/day zoliflodacin by oral gavage for 14 days (n=3/sex). Two additional animals per sex were dosed with 250, 500 or 1000 mg/kg/day for toxicokinetic evaluation.

Water consumption was increased in the 500 and 1000 mg/kg/day groups 2.2-2.7 x predose.
Increased reticulocyte counts were seen in all drug-treated female rats, with marked increases seen in one animal each treated with 500 mg/kg/day (4-fold) and 1000 mg/kg/day (8-fold) in the repeat-dose phase of the study. These animals also showed slight to moderate (10% to 20%) decreases in hemoglobin, hematocrit, and RBC count. One male rat each in the mid- and high repeat-dose phase groups also had increased reticulocytes.
Drug-treated males had a nondose-related reduction (80% to 90%) in urinary protein on Day 13.
An enlarged spleen was seen in one high-dose female from the repeat-dose phase (same rat that showed numerous changes in hematology parameters. This is indicative of a regeneration of red blood cells to compensate for loss).
Histopathology included moderate to severe extramedullary hematopoiesis in the spleens of rats of both sexes dosed with 1000 mg/kg/day and females treated with 500 mg/kg/day. Dose-related minimal to moderate exfoliation of germinal epithelial cells and immature sperm were seen in the epididymides of males that received 500 and 1000 mg/kg/day. This finding was also seen in the testes of males that received 1000 mg/kg/day (graded minimal).

13.1.5.2. Genetic Toxicology

Table 63. Genetic Toxicology, In Vitro Reverse Mutation Assay in Bacterial Cells (Ames)

Study Features and Methods	Details
Study no.	2895BV
Study title	AZ13420914: Genetic Toxicity Evaluation using a Bacterial Reverse Mutation Test
Conducting laboratory	AstraZeneca R&D Alderley Park, Macclesfield, England

Study Features and Methods	Details
GLP compliance	Yes
Drug, lot #, % purity	AZ13420914 hydrate, batch no. 05-kcrl979-022 P2, 99.7% pure (as hydrate)
Strains:	*Salmonella typhimurium* TA98, TA100, TA102, TA1535, and TA1537
Concentrations in the definitive study/method	0.005, 0.016, 0.05, 0.16, 0.5, 1.6, and 5 µg/plate
Basis of concentration selection	Excessive cytotoxicity at 5 µg/plate
Formulation/vehicle	DMSO
Results	AZ13420914 was mutagenic in *S. typhimurium* strain TA102 ± rat S9 under the conditions of this assay. Mutagenicity was not observed in TA98, TA100, TA1535 or TA1537. Ames positivity has been reported in other DNA gyrase inhibitors in strain TA102. Despite the positive finding in TA102, this drug is unlikely to present an increased risk of mutagenicity to patients because this is a known finding in this bacterial strain with other drugs with similar mechanisms of action.
Validity	Valid

Source: Reviewer table from Applicant's study report data
Abbreviations: AZ13420914, zoliflodacin; DMSO, dimethyl sulfoxide; DNA, desoxyribonucleic acid; GLP, good laboratory practice, no, number

Table 64. Genetic Toxicity, Evaluation Using the Mouse Lymphoma Cell Thymidine Kinase Locus Assay

Study Features and Methods	Details
Study no.	793098
Study title	AZD0914: Genetic Toxicity Evaluation Using the Mouse Lymphoma Cell Thymidine Kinase Locus Assay
Conducting laboratory	(b) (4)
GLP compliance	Yes
Drug, lot #, % purity	AZD0914, batch no. BNG-LSL-12124, 96.4% pure
Cell line	Mouse lymphoma L5178Y cells ($TK^{+/-}$ 3.7.2C)
Concentrations in the definitive study	3 hours -S9: 175.48, 214.47, 253.47, 292.46, 331.46, 370.45, 409.45, 448.44 µg/ml 3 hours + S9: 146.23, 170.60, 194.97, 219.35, 243.72, 268.09, 292.46, 316.83 µg/ml 24 hours -S9 (1): 19.50, 58.49, 97.49, 136.48, 175.48, 214.47, 253.47, 292.46 µg/ml 24 hours -S9 (2): 29.25, 43.87, 58.49, 73.12, 87.74,102.36, 116.98, 131.61, 146.23, 160.85, 175.48, 190.10 µg/ml
Basis of concentration selection	Cytotoxicity testing (cell suspension growth relative to solvent control)- concentrations up to 487.44 µg/ml were tested ± S9 for 3 hours incubation and 24 hours incubation without S9. 1000 µmol/L has excessive toxicity for 3 hours and 500 µmol/L was excessively toxic for 24 hours.
Negative control	DMSO
Positive control	No S9: Methyl methanesulphonate (MMS, 10 µg/ml for 3 hours, 5 µg/ml for 24 hours) With S9: 3-Methylcholanthrene (3-MC, 2.5 and 10 µg/ml)
Formulation/vehicle	DMSO
Incubation and sampling time	3 hours with or without metabolic activation (rat S9) 24 hours without S9
Results	Under the conditions of this assay, AZD0914 was not mutagenic or clastogenic to mouse lymphoma L5178Y cells regardless of metabolic activation with rat S9.

Study Features and Methods	Details
Validity	Valid

Source: Reviewer table from Applicant's study report data
Abbreviations: AZD0914, zoliflodacin; DMSO, dimethyl sulfoxide; GLP, good laboratory practice, no, number

Table 65. Genetic Toxicology, In Vivo Clastogenicity Assay in Rat (Micronucleus Assay)

Study Features and Methods	Details
Study no.	793674
Study title	AZD0914: Genetic Toxicity Evaluation using the Rat Micronucleus Test after Two Oral Doses
Conducting laboratory	(b) (4)
GLP compliance	Yes
Drug, lot #, % purity	AZD0914, batch no. BNG-LSL-12124, 96.4%
Species/strain	Rat/Wistar Han
Number/sex/group	7 males/group for vehicle and all doses of AZD0914 3 males for positive control
Doses in the definitive study	0 (vehicle), 50, 250 and 500 mg/kg
Basis of dose selection	High dose was selected based on bone marrow hematological effects seen in Study 3494DR.
Negative control	(b) (4)
Positive control	20 mg/kg cyclophosphamide, single dose given 24 hours before sacrifice
Formulation/vehicle	(b) (4)
Results	AZD0914 did not induce micronuclei in immature erythrocytes in the bone marrow of male Wistar Han rats after 2 daily consecutive oral doses up to 500 mg/kg/day and is therefore not clastogenic in vivo.
Validity	Yes

Source: Reviewer table from Applicant's study report data
Abbreviations: AZD0914, zoliflodacin; GLP, good laboratory practice, no., number

13.1.5.3. Carcinogenicity

No studies were conducted.

13.1.5.4. Reproductive Toxicology

Study 497076/AZD0914: Oral (Gavage) Fertility and Early Embryonic Development Study in the Male Rat (Good Laboratory Practice)

Key Study Design Features and Findings

Male rats were administered 0, 200, 500, or 1000 mg/kg/day AZD0914 by oral gavage for 4 weeks prior to mating and for 1 week during the first mating period (where the males were paired with untreated females). The same males were then remated with different untreated females for a second mating period after a 4-week recovery period.

Body weight gains were lower in the 500 and 1000 mg/kg dose groups from Day 25 until the end of dosing. Water consumption was increased in these two groups during the dosing period.

During the first mating period, number of matings and time to mating were similar across groups. The male fertility index was reduced to 68.8% in the 500 mg/kg dose group and 0% in the 1000 mg/kg dose group.

From the mating during dosing, there was an increase in pre-implantation loss (24.9% versus 3.2% respectively) and post-implantation loss (12.2% versus 2.3% respectively) in the 500 mg/kg/day dose group, compared to controls.

During the second mating period, mating and pregnancy parameters were similar across groups.

At necropsy at the end of the recovery period, histopathological changes were noted in the 500 and 1000 mg/kg/day dose groups in the epididymides (minimal cellular debris associated with testicular degeneration) and testes (minimal to marked tubular degeneration, minimal tubular vacuolation, and minimal cellular debris).

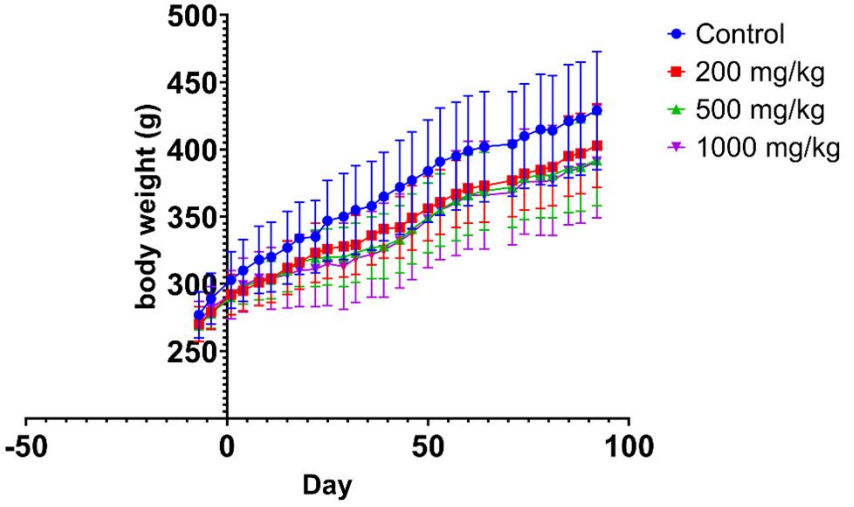
NOAEL was 200 mg/kg which had mean AUC exposures of 795 µg*h/mL and C_{max} of 62 µg/mL.

Table 66. Male Rat FEED, Methods

Parameter	Method Details
Dose and frequency of dosing	0, 200, 500, or 1000 mg/kg/day once daily
Dosing days	4 weeks prior to mating and for the 1-week mating period (Days 1 to 35)
Route of administration:	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Rat/ Han Wistar (b) (4):WI (Han)
GLP compliance	Yes
Number/sex/group	16 males/group
Study design	Only males were dosed. Males were 10 to 11 weeks old at the start of the study (267 to 342 g). Males were dosed for 4 weeks prior to the first mating period of 1 week and dosed until the end of the mating period. Female animals were monitored by morning vaginal lavage for 10 days for stage of estrus cycle prior to initiation of mating. Males and females were paired for mating on a 1:1 basis. The pairing period was for up to 7 nights. Females were monitored early each morning by vaginal lavage until mating occurred as noted by copulatory plug or sperm in the vaginal lavage. Based on fertility effects, following dosing, the males were maintained for a 4-week recovery period and remated with additional females. The males were retained until Day 92 or 93. Necropsy was performed on the reproductive organs for pregnancy parameters in females on Days 12 to 14 of gestation. Male necropsies followed by histological examination were conducted on Days 92 or 93.
Deviation from study protocol affecting interpretation of results	Protocol deviations were minor and were assessed not to affect the evaluation or outcome of this study.

Source: Reviewer table from Applicant's study report data
Abbreviations: (b) (4); FEED, fertility and early embryonic development; GLP, good laboratory practice; (b) (4); w/v, weight/volume

Table 67. Male Rat FEED, Observations and Results

Parameter	Method Details
Mortality	No unscheduled deaths were reported.
Clinical signs	Increased salivation was noted in all test article dose groups from about Days 8 to 10 to the end of dosing, but not during the recovery period.
Body weights	Significantly decreased total male body weights compared to controls were found starting on Day 25 for the higher two dose groups. Male body weight gains during dosing in the mid- and high-dose groups were also significantly reduced compared to controls (−34%, −46%). The low-dose group body weights and body weight gains were lower than controls (−21%), but the differences were not statistically significant. During the recovery period, the weight gains were similar between groups but were still significantly lower in the mid- and high-dose groups compared to controls (9%, 9%). **Figure 4. Total Male Body Weights, Male Rat FEED Study**  Source: Reviewer constructed figure from study data using GraphPad Prism Abbreviation: fertility and early embryonic development Female total body weights (undosed females) were similar across groups during gestation, except in the 1000 mg/kg batch-one mated females (no pregnancies) and 500 mg/kg batch-one mated females, which had a lower number of implants when evaluated on Day 12 to 14 of gestation. Gravid uterine weights were not measured and therefore could not be directly accounted for in the body weight differences.
Feed and water consumption	Feed consumption in all test article-treated groups was slightly reduced for about the first 11 days (up to 17%) and was similar to controls for the remainder of the dosing period and recovery period. Males in the mid- and high-dose groups exhibited increased water consumption by up to 58% compared to controls during the dosing period until about 4 days after dosing.

Parameter	Method Details
Fertility parameters	Number of matings and time to mating were similar across groups during dosing and after recovery. Administration of 500 or 1000 mg/kg AZD0914 reduced the male fertility index to 68.8% and 0%, respectively, during the dosing period; no effect was found in the 200 mg/kg/day dose group. The fertility indices were similar across groups at the recovery period mating. During dosing, in the 500 mg/kg/day dose group, pre-implantation loss was increased over controls (24.9% versus 3.2%) as was post-implantation loss (12.2% versus 2.3%). Low dose pregnancy performance was similar to controls (see Table 68 below). For the recovery mated group, pregnancy performance was similar across all groups (data not shown).

Table 68. Mating Performance and Fertility Indices (Dosing Period)

Number of Nights to Positive Mating Sign	Group (Dose Level mg/kg/day)			
	1 (0)	2 (200)	3 (500)	4 (1000)
	Number of Animals (Number of these not becoming pregnant)			
1	1	2	1	2(2)
2	5	5	7(2)	8(8)
3	4	7	5(1)	3(3)
4	6(1)	1	3(2)	3(3)
No positive mating sign	0	1(1)	0	0
Median number of nights to positive mating sign	3	3	2.5	2
Number passing one oestrus	0	1	0	0
Number of males with positive mating	16	15	16	16
Number of siring males	15	15	11	0
Male Fertility Index (%)	93.8	100	68.8	0

Source: Page 56 of the study report

Clinical pathology	No adverse test article-related effects were noted. Nonadverse decreases in hematocrit in the mid- and high-dose males and increases in mid- and high-dose white cell counts were reported.
Necropsy findings	No test article-related male organ weight changes were reported. Histopathological changes were noted in the mid- and high-dose groups in the epididymides and testes. In the epididymides, minimal cellular debris associated with testicular degeneration was reported in one animal at the 500 mg/kg/day dose and three animals at the 1000 mg/kg/day dose. One animal in the 200 mg/kg/day dose group also had minimal tubular cellular debris but without testicular degeneration and the finding was therefore not considered test article-related. In the testes of animals dosed at 500 and 1000 mg/kg/day, tubular degeneration (2/16 and 8/16 animals, respectively), tubular vacuolation (1/16 and 10/16 animals, respectively) and cellular debris (1/16 animal in each group) were reported.

Source: Reviewer table from Applicant's study report data
Abbreviations: AZD0914, zoliflodacin; FEED, fertility and early embryonic development

Toxicokinetics

Toxicokinetics were assessed on Day 32 from animals that did not have blood drawn for hematology samples (n=3).

Exposures increased with increasing dose.

C_{max} increased less than dose-proportionately.

AUC_{0-24} increased less than dose proportionately from 200 to 500 mg/kg/day and roughly dose-proportionately between 500 and 1000 mg/kg/day.

Half-lives increased with increasing dose (2.7, 4.0, and 7.6 hours for 200, 500 and 1000 mg/kg/day, respectively).

Table 69. TK Parameters, Study 497076

	Dose (mg/kg/day)		
Parameter	**200 Day 32**	**500 Day 32**	**1000 Day 32**
AUC_{0-t} (µg*h/mL)	795	1287	2608
C_{max} (µg/mL)	62	123	178
T_{max} (h)	6	4	6

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study 496884/AZD0914: Oral (Gavage) Fertility and Embryofetal Development Study in the Female Rat (Good Laboratory Practice)

Key Study Findings

Female rats were administered AZD0914 at doses of 0, 200, 500, or 1000 mg/kg/day by oral gavage for 2 weeks prior to mating until gestation day (GD) 16.

Clinical signs in the 1000 mg/kg dose group included salivation and ploughing behavior. Two females in this group had scabbing or reddened skin on muzzle, ears, tail, or extremities at the end of gestation which resolved when dosing stopped before termination.

No significant differences in estrus cycles, mating, or precoital index were reported. In the 500 and 1000 mg/kg/day dose groups, the number of early intrauterine deaths increased. Mean gravid uterine weights were 24% lower in the 1000 mg/kg dose group compared to controls. Mean litter weights were significantly lower in the 200 and 1000 mg/kg dose groups (18% and 29%, respectively).

No test article-related external or visceral fetal abnormalities were reported. In all dose groups, an increase in litters with skeletal variations and a decrease in skeletal ossification was reported.

The no-observed-effect level for maternal toxicity was 200 mg/kg/day. No NOAEL for fetal development could be determined in this study based on the change in litter weights.

Table 70. Female Rat FEED, Methods

Parameter	Method Details
Dose and frequency of dosing	0, 200, 500, or 1000 mg/kg/day daily
Dosing days	From 2 weeks prior to mating until gestation day (GD) 16
Route of administration	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Rat/ Han Wistar (b) (4):WI (Han)
GLP compliance	Yes
Number/sex/group	20 females/group
Study design	Females were dosed in two batches a week apart each with 10 females per dose group. Combined FEED and EFD study with female only dosing.
Deviation from study protocol affecting interpretation of results	No protocol deviations were noted to affect the evaluation or outcome of this study.

Source: Reviewer table from Applicant's study report data
Abbreviations: (b) (4); EFD, embryo-fetal development; FEED, fertility and early embryonic development; GLP, good laboratory practice; w/v, weight/volume

Table 71. Female Rat FEED, Observations and Results

Parameter	Major Findings
Mortality	No unscheduled maternal deaths were reported.
Clinical signs	Occasional salivation and/or ploughing were observed in the 1000 mg/kg/day dose group (19/20 animals). Two females in the 1000 mg/kg/day dose group were observed with scabbing or reddened skin on muzzle, ears, tail, or extremities during the end of gestation which resolved when dosing stopped before termination. One of the two animals was also observed with hunched posture on two occasions during the same time period.
Body weights	During the 2 weeks prior to mating, mean body weight gains in the test article-administered groups were increased compared to controls. After completion of dosing, animals in the 200 and 1000 mg/kg/day dose groups had lower weight gains until termination. Terminal body weights corrected for uterine weights were 8% and 6% higher in the 500 and 1000 mg/kg/day dosing groups, respectively, compared to controls.
Feed and water consumption	Prior to mating, test article-treated animals had reduced feed consumption of 15% to 20% for Days 1 to 3. Water consumption was increased compared to controls in the 500 and 1000 mg/kg/day dosing groups throughout the dosing period. Premating, water consumption increased 26% to 41% and 45% to 85% in the 500 and 1000 mg/kg/day dosing groups respectively, and during gestation increased about 35% and 78% in the 500 and 1000 mg/kg/day dosing groups, respectively.
Fertility parameters	No test-article-related changes were reported in estrus cycles, mating, or precoital index.
Maternal necropsy findings	Three, one, and four females in the 200, 500 and 1000 mg/kg/day groups, respectively, were found not to be gravid, which was not a statistically significant difference.
Caesarean section data	At 500 and 1000 mg/kg/day dosing, the number of early intrauterine deaths was increased from 3.3% in controls to 10.1% and 11.4%, respectively. Mean gravid uterine weights were 24% lower in the 1000 mg/kg/day dose group. Mean litter weights were significantly lower in the 200 mg/kg/day group (18%) and 1000 mg/kg/day group (29%) and were not significantly decreased in the 500 mg/kg/day group mean litter weights (−9%).
Offspring terminal observations	Mean fetal weights were lower in all test article-treated dose groups by 8%, 10%, and 18% in the 200, 500, and 1000 mg/kg/day dose groups, respectively.

Parameter	Major Findings
Offspring necropsy findings	No test article-related external or visceral fetal abnormalities were reported. In all test article-treated groups, there was an about 1.7- to 2.4-fold increase in number of litters with skeletal variations and a decrease in ossification. Additionally, fewer fetuses with supernumerary ribs were noted in the treated groups (0.5- to 0.7-fold). A dose-dependent decrease in skeletal ossification was also evident (2- to 23-fold).

Source: Reviewer table from Applicant's study report data
Abbreviation: FEED, fertility and early embryonic development

Study 496910/AZD0914: Twice Daily Oral (Gavage) Embryofetal Development Study in the Mouse (Good Laboratory Practice)

Key Study Findings

One female in the 1000 mg/kg group was euthanized early due to adverse clinical signs.

Two animals in the 1000 mg/kg/day dose groups were reported to have clinical signs of crackling respiration, piloerection, and anal discharge for 1 day; dosing was skipped for each animal on that day. Two different 1000 mg/kg/day group animals had red discharge and were found at necropsy to be no longer gravid.

Water consumption increased in all test article-treated groups.

A statistically significant reduction in mean numbers of implants and mean live implants was found in the 500 and 1000 mg/kg/day dose groups, and an increase in post-implantation losses was found in the 1000 mg/kg/day group. Mean uterine weight was lower in all test article-treated groups compared to controls. Mean litter male fetus weights, female fetus weights, and combined fetus weights were significantly lower in the 1000 mg/kg/day group compared to controls.

In the 500 and 1000 mg/kg/day dose groups, exencephaly, a rare malformation, was present and considered test article-related. In the 500 and 1000 mg/kg/day dose groups, an increase in delayed ossification was reported.

The NOAEL was 250 mg/kg/day (125 mg/kg/dose) in this study for maternal toxicity and developmental endpoints.

Table 72. Methods, Embryo-Fetal Development Study in Mice

Parameter	Method Details
Dose and frequency of dosing	0, 125, 250 or 500 mg/kg/dose twice daily, 8 hours apart (0, 250, 500, or 1000 mg/kg/day) from gestation day (GD) 5 to 16
Route of administration	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Mouse/CD-1 CRL:CD-1 (ICR)
GLP compliant	Yes
Number/sex/group	22 females/group
Deviation from study protocol affecting interpretation of results	Several protocol deviations were reported. Most were minor and would not affect the reporting of the study. Any that might be more impactful are noted below.

Source: Reviewer table from Applicant's study report data
Abbreviations: CRL, Charles River Laboratories; GLP, good laboratory practice; HCl, hydrochloride; ICR, Institute of Cancer Research; w/v, weight/volume

Table 73. Observations and Results, Embryo-Fetal Development Study in Mice

Parameter	Major findings
Mortality	One animal in the 1000 mg/kg/day dose group was terminated early due to adverse clinical signs. These included a few days of unkempt coat, and on the day of termination abnormal color discharge from the anus, pale skin, piloerection, and irregular crackling respiration. At necropsy the caecum and jejunum were reported as distended by gas, the trachea was froth filled, and the lungs were mottled.
Clinical signs	Two animals in the 1000 mg/kg/day group (#74, 78) were reported to have crackling respiration, piloerection, and anal discharge on GD 13 (#78) or Day 15 (#74). Dosing was skipped on the day that these clinical signs were reported. Two animals in the 1000 mg/kg/day group (#77, #82) were reported to have red discharge on GD 8 and 9. At necropsy, one animal had no live implants and early embryonic death and the other had no live implants, but uterine staining showed 13 implant sites.
Body weights	Mean BW in the 1000 mg/kg/day group were statistically significantly lower from controls on GD 14 to 17 (9% to 10%). Mean BW gain from GD4 to GD17 was significantly lower in this group as well (18%).
Feed and water consumption	Feed consumption data were noted to have errors and was therefore considered unreliable. Water consumption was generally increased in test article-treated animals compared to controls, though the changes were variable and did not appear to have a dose-response relationship (6% to 17% at 250 mg/kg/day, 3% to 52% at 500 mg/kg/day and 0.7% to 36% higher than controls at 1000 mg/kg/day).
Maternal necropsy findings	No test article-related findings.
Cesarean section data	A statistically significant reduction in mean number of implants and mean live implants was found in females in the 500 mg/kg/day and 1000 mg/kg/day group. There were fewer gravid animals at necropsy in the 1000 mg/kg/day group (15) compared to controls (19), but this was not statistically significant. There was in increase in post-implantation loss in the 1000 mg/kg/day group (5% in control, 13% in 1000 mg/kg/day group). Mean gravid uterine weight was lower in all test article-treated groups compared to controls. Mean litter male fetus weights, female fetus weights, and combined fetus weights were significantly lower in the 1000 mg/kg/day group compared to controls. The terminal body weights corrected for the uterine weights were comparable between groups.
Offspring terminal observations and necropsy findings	The rare fetal malformation exencephaly was found in one fetus in each of two litters in the 500 mg/kg/day dose group and two fetuses in one litter in the 1000 mg/kg/day dose group. Exencephaly is a neural tube defect, and a rare malformation. In test article-treated animals, it was present at rates above the Applicant-provided historical control data. Incidence of fetuses with unossified/incomplete ossification of the skull bones, increased ossified vertebral centra and phalangeal elements, incompletely ossified cervical vertebral arches and a lower incidence of rib costal cartilage attached to sternum were increased in the 500 and 1000 mg/kg/day dose groups.

Source: Reviewer table from Applicant's study report data
Abbreviations: BW, body weight; EFD, embryo-fetal development; GD, gestation day

Toxicokinetics

- Toxicokinetics were assessed on Day 16 from three animals at each time point.
- Systemic exposures increased with increasing doses in a roughly dose-proportional way.

Table 74. TK Parameters, Study 497076

	Dose (mg/kg/day)		
Parameter	**250 Day 16**	**500 Day 16**	**1000 Day 16**
$AUC_{0\text{-}t}$ (µg*h/mL)	204	517	941
C_{max} (µg/mL)	27.2	55.1	106.7
T_{max} (h)	8.5	10	8.5

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; TK, toxicokinetic; T_{max}, time to maximum concentration

Study G19182/Zoliflodacin: Study for Effects on Pre and Postnatal Development, Including Maternal Function with Toxicokinetics by Oral Gavage in Wistar Rats (SEGMENT-III STUDY) (Good Laboratory Practice)

Key Study Design Parameters and Study Findings

Mated female Wistar rats were administered 0, 50, 100, or 200 mg/kg/day zoliflodacin from gestation Day 6 until weaning on lactation day 20.

In a motor task assessment, ambulatory counts and distance traveled were higher in high-dose F1 males and mid- and high-dose females compared to controls.

A water M-maze task used in this study was inadequately designed for assessing learning and memory in F1 animals.

No differences in mating, fertility, or pregnancy parameters were reported in the F1 groups. No test article-related abnormalities were reported in the second generation (F2) pups.

Based on the changes in motor activity at ≥100 mg/kg/day, the NOAEL was 50 mg/kg/day in the F1 animals. This determination does not consider any measurement of learning and memory in F1 animals, which were not assessed with a validated test in this study.

Table 75. Methods, Rat PPND Study

Parameter	**Method Details**
Dose and frequency of dosing:	0, 50, 100, 200 mg/kg/day once daily
Route of administration	Oral gavage
Formulation/vehicle	(b) (4)
Species/strain	Rat/Wistar
GLP compliant	Yes
Number/sex/group	25 mated females/group (main) 9 mated females/group [TK group] for test article-administered animals and 5 mated females/group [TK group] for controls
Study design comments	The highest dose should induce some toxicity, which would be higher than the NOAEL, so this is not an ideal selection of high dose. The high dose of 200 mg/kg/day was also not anticipated to lead to exposures >3× the clinical exposure.
Deviation from study protocol affecting interpretation of results	Study deviations were minor and should not affect interpretation of the study.

Source: Reviewer table from Applicant's study report data
Abbreviations: GLP, good laboratory practice; NOAEL, no-observed-adverse-effect level; PPND, pre- and postnatal development; TK, toxicokinetic; w/v, weight/volume

Table 76. F0 Generation Findings, Study G19182

Parameter	Major Findings
Mortality	No unscheduled deaths
Clinical signs	No adverse clinical signs
Body weights	No test article-related differences between groups
Feed Consumption	All test article-treated groups had statistically significantly increased feed consumption during lactation days 18 to 21 but this was not considered adverse.
Pregnancy status	One dam in the 50 mg/kg/day dose group had a total litter loss and one dam in each of the 50, 100, and 200 mg/kg/day dose groups were found not to be gravid. Gestation length, fertility index, implantations, and post-implantation loss were similar between groups.
Necropsy findings	No test article-related gross lesions were reported at necropsy.

Source: Reviewer table from Applicant's study report data

Table 77. F1 Generation Findings, Study G19182

Parameter	Major Finding
Mortality	No unscheduled deaths
Clinical signs	No test article-related clinical signs
Body weights	Male body weights were statistically significantly higher than control on Days 1, 4, 8 in the 50 mg/kg/day group, Days 1, 4, 8, 11, 15, and 18 in the 100 mg/kg/day group, and Days 1, 4, 8, 11, 15, 18, 22, and 29 in the 200 mg/kg/day dose group. The percent differences were small and overall weight gain was similar across groups, so the changes were not considered adverse.
Physical development	Developmental landmarks were achieved in all pups in all litters and the timing was similar between groups.
Sexual maturation	Mean day of female vaginal patency and mean body weight on that day was similar across groups. Mean day and body weights at acquisition of male balano–preputial separation was similar across groups.
Behavior and activity	Sensory observations were normal in all animals in all groups. In an open field type study, ambulatory count and distance traveled during each of the three intervals and total ambulatory counts and distance travelled in the high-dose males and mid- and high-dose females were increased compared to controls. Changes in water M-maze parameters were found compared with controls in males and females. However, the design of the test was not considered to be adequately designed for detecting learning and memory changes and therefore was not used in determination of the NOAEL in this study.
Mating, fertility, and pregnancy parameters	Measured parameters, body weights, body weight gains, feed consumption during gestation or lactation, and mating and fertility parameters, were similar across groups.
Necropsy findings	No test article-related gross lesions were reported at necropsy in the F1 generation.

Source: Reviewer table from Applicant's study report data
Abbreviation: NOAEL, no-observed-adverse-effect level

Table 78. F2 Generation Findings, Study G19182

Parameter	Major Findings
Malformations	No abnormalities were reported on the day of birth. Abnormalities at necropsy were not considered test article-related.
Litter size	Number of implantations, pre- and post-implantation losses, mean number, sexes, and litter size were similar across groups.
Survival	Live birth index and survival at postnatal Day 4 were similar among groups.
Pup weights	Pup weights were similar across groups.

Source: Reviewer table from Applicant's study report data

Toxicokinetics

Exposure as C_{max} and AUC_{0-24h} increased generally greater than dose-proportionately from 50 to 100 mg/kg/day and roughly dose-proportionately from 100 to 200 mg/kg/day during gestation.

Exposures were similar on GD 6 and GD 14 for all three doses. Exposures were lower on lactation day 21 compared to during gestation. The reason for the decrease in exposure during lactation was unclear as reduced exposure with continued dosing was not seen in other repeat dose rat studies. Possible mechanisms include excretion of test article into milk, increased metabolism, or increased excretion via the feces or urine.

Table 79. F0 Dams TK Parameters, Study G19182

	Dose (mg/kg/day)								
	50			100			200		
Parameter	**GD6**	**GD14**	**LD21**	**GD6**	**GD14**	**LD21**	**GD6**	**GD14**	**LD21**
AUC_{0-24h} (µg*h/mL)	55	59.7	42.2	459	416	79.3	801	774	240
C_{max} (µg/mL)	10.6	17.0	12.2	39.0	45.8	22.5	55.4	67.2	45
T_{max} (h)	2	1	1	2	1	0.5	6	4	0.5

Source: Reviewer table from Applicant's study report data
Abbreviations: AUC, area under the concentration-time curve; C_{max}: maximum plasma concentration; GD, gestation day; LD, Lactation day; TK, toxicokinetic; T_{max}, time to maximum concentration

13.1.5.5. Impurities

Summary of Safety of Specified Impurities in Zoliflodacin

Four impurities in zoliflodacin, which are (b) (4) of zoliflodacin, were identified as having the potential to be above the qualification limit and were evaluated for safety as impurities of concern (summarized in Table 80 below). Details on the studies used to qualify the impurities are included following the table.

Table 80. Specified Impurity Qualifications Summary

(b) (4)	Genetic Toxicology Assays	Qualified?
	Bacterial reverse mutation Mouse Lymphoma assay	Yes[a]
	None	Yes[a]
	Bacterial reverse mutation Mouse Lymphoma assay	Yes[a]
	Bacterial reverse mutation Mouse Lymphoma assay	Yes[a]

Source: Reviewer table from submitted information and reviewer analysis
[a] Qualification was based on results of genotoxicity assays and Study G19181. Review summaries are included below.
Abbreviation: RRT, relative retention time

Study G19181: 14-Day Repeated Dose Toxicity and Toxicokinetics Study by Oral Gavage in Wistar Rats (Good Laboratory Practice)

Key Study Methods And Findings

Male and female rats (10/sex/group in the main study, 5/sex/group recovery animals, 3/sex for TK controls and 9/sex/group in test article-administered TK groups) were administered zoliflodacin by oral gavage daily for 14 days at 0, 200, or 500 mg/kg/day. Zoliflodacin was administered from a purified batch alone or from a batch enriched with (b) (4) impurities.

Zoliflodacin alone was 98.5% pure. The enriched batch contained (b) (4)

(b) (4)

Toxicokinetics were measured for zoliflodacin and for only one impurity as representative of the other impurities (b) (4)

No test article-related adverse effects were reported in this study. The NOAEL was 500 mg/kg/day with or without added impurities. This dose corresponded to Day 14 mean C_{max} values for zoliflodacin of 92.1 µg/mL in males and 102 µg/mL in females. The AUC_{last} values of zoliflodacin in males was 1320 µg*hr/mL and in females 1500 µg*hr/mL. The C_{max} values of Impurity (b) (4) in males was (b) (4) µg/mL and in females was (b) (4) µg/mL, respectively. The AUC_{last} values of Impurity (b) (4) in males was (b) (4) µg*hr/mL and in females (b) (4) µg*hr/mL, respectively.

Toxicokinetics

T_{max} ranged from 1 to 6 hours.

Generally, with increasing dose, exposure (C_{max} and AUC_{last}) increased, though in males the increase was more than dose proportional and in females it was less than dose proportional. Sex differences in exposure were only greater than 2-fold on both study days in 200 mg/kg/day groups where females had higher exposures than males.

Accumulation was not seen.

Table 81. Zoliflodacin TK Parameters, Study G19181

Day	Treatment	Sex	Dose (mg/kg/day)	T_{max} (h)	C_{max} (µg/mL)	AUC_{last} (µg*hr/mL)	$T_{½}$ (h)
1	Zoliflodacin without impurity	Male	200	2	61.2	313	NC
		Male	500	4	111	1550	NC
		Female	200	6	85.9	1190	NC
		Female	500	2	123	1590	6.9
14	Zoliflodacin without impurity	Male	200	1	70.6	675	2.2
		Male	500	4	92.1	1320	NC
		Female	200	1	60.9	776	3.32
		Female	500	1	102	1500	7.39
1	Zoliflodacin with impurity	Male	200	(b) (4)			
		Male	500				
		Female	200				
		Female	500				
14	Zoliflodacin with impurity	Male	200				
		Male	500				
		Female	200				
		Female	500				

Source: Reviewer constructed table derived from the table on page 578 of the study report.
NC: not calculated due to improper elimination phase.
Abbreviations: AUC, area under the concentration-time curve; C_{max}; maximum plasma concentration; $T_{½}$, half-life; TK, toxicokinetic; T_{max}, time to maximum concentration

(b) (4) Impurity Toxicokinetics

T_{max} ranged from (b) (4) hours.

Generally, with increasing dose, exposure (C_{max} and AUC_{last}) increased, roughly dose-proportionally.

Accumulation was not seen.

Exposure was about (b) (4) compared to zoliflodacin and did not affect zoliflodacin exposures.

Table 82. Zoliflodacin (b) (4) Impurity, TK Parameters

Day	Treatment	Sex	Group	Dose (mg/kg /day)	T_{max} (h)	C_{max} (µg/mL)	AUC_{last} (µg*hr/mL)	C_{last} (µg/mL)	T_{last} (h)	$T_{½}$ (h)
1	Zoliflodacin without impurity	Male	G2BTK	200	1	1.72	5.7	0.558	6	NC
		Male	G3BTK	500	2	3.04	16.1	0.0743	24	5.21
		Female	G2BTK	200	2	4.26	15.9	1.35	6	NC
		Female	G3BTK	500	1	5.44	46.2	0.197	24	5.02
14	Zoliflodacin without impurity	Male	G2BTK	200	0.5	0.89	3.52	0.427	6	NC
		Male	G3BTK	500	2	3.32	27.7	0.0209	24	3.02
		Female	G2BTK	200	0.5	3.44	7.89	0.784	6	NC
		Female	G3BTK	500	1	6.41	44	0.34	24	7.93

Source: Reviewer constructed table
Abbreviations: AUC, area under the concentration-time curve; C_{max}; maximum plasma concentration; $C_{last,}$ last measurable plasma concentration; $T_{½}$, half-life; NC, not calculated; TK, toxicokinetic; T_{max}, time to maximum plasma concentration, T_{last}, time to last measurable plasma concentration

Genetic Toxicology Testing for (b) (4)

Table 83. Genetic Toxicology, In Vitro Reverse Mutation Assay in Bacterial Cells (Ames) of (b) (4)

Study Features and Methods	Details
Study no.	G19179
Study title	(b) (4): Bacterial Reverse Mutation Test
Conducting laboratory	(b) (4)
GLP compliance	Yes
Drug, lot #, % purity	(b) (4) CZOF/CZOF10-202-0373218, 97.9%
Strains	*Salmonella typhimurium* TA98, TA100, TA102, TA1535, and TA1537 *E. coli* WP2*uvr*A (pKM101)
Concentrations in the definitive study/method	0.3, 1, 3, 10, 31 and 100 µg/plate
Basis of concentration selection	Excessive cytotoxicity at 200 µg/plate
Formulation/vehicle	DMSO

Study Features and Methods	Details
Results	Cytotoxicity (moderate diminution of the bacterial lawn) and reduction in mean revertant colonies was seen at 100 µg/plate ± S9 in the definitive assays. In TA102, the (b) (4) impurity increased colonies under the conditions of this assay, with a 2-fold increase in revertant colonies seen at 100 µg/plate (and close to 2-fold at 31 µg/plate). Mutagenicity was not observed in the other bacterial strains. The parent drug was also positive in strain TA102 in the Ames assay. This impurity was able to be tested at a higher top concentration than the parent drug. As noted for the parent drug, Ames positivity has been reported in other DNA gyrase inhibitors in strain TA102. Despite the positive finding in TA102, this impurity is unlikely to present an increased risk of mutagenicity to patients because this is a known finding in this bacterial strain with other drugs with similar mechanisms.
Validity	Valid

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; DNA, desoxyribonucleic acid; GLP, good laboratory practice; no., number; RRT, relative retention time

Table 84. Genetic Toxicity Evaluation of (b) (4) Using the Mouse Lymphoma Cell Thymidine Kinase Locus Assay

Study Features and Methods	Details
Study no.	G19180
Study title	(b) (4) In Vitro Mammalian Cell Gene Mutation Test Using the Thymidine Kinase Gene
Conducting laboratory	(b) (4)
GLP compliance	Statement that OECD, FDA GLP were followed, but certification is from the (b) (4)
Drug, lot #, % purity	(b) (4) Batch no. CZOF/CZOF10-202-0373218, 97.9% pure
Cell line	Mouse Lymphoma L5178Y $TK^{+/-}$ −3.7.2C
Concentrations in the definitive study	7.62, 30.47, 121.86 and 487.44 µg/mL
Basis of concentration selection	Cytotoxicity: (b) (4) relative total growth in the presence or absence of metabolic activation with 3-hour incubation or absence of metabolic activation with 24-hour incubation.
Negative control	DMSO
Positive control	−S9: 10 µg/mL methyl methanesulfonate +S9: 12 µg/mL cyclophosphamide
Formulation/vehicle	DMSO
Incubation and sampling time	3 hours +S9 3 hours −S9 24 hours −S9
Results	(b) (4) was not genotoxic (mutagenic or clastogenic) under the conditions of this assay. Increases in neither small nor large colonies were observed in the presence of the test article.
Validity	Yes

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; GLP, good laboratory practice; no., number; OECD, Organization for Economic Cooperation and Development; RRT, relative retention time

Genetic Toxicology Testing (b) (4)

Table 85. Genetic Toxicology, In Vitro Reverse Mutation Assay of (b) (4) in Bacterial Cells (Ames)

Study Features and Methods	Details
Study no.	19207
Study title	(b) (4) Bacterial Reverse Mutation Test
Conducting laboratory	(b) (4)
GLP compliance	Yes
Drug, lot #, % purity	(b) (4) CZOF/CZOF10-199-0373213, 99.7% pure
Strains	*Salmonella typhimurium* TA98, TA100, TA102, TA1535, and TA1537 *E. coli* WP2*uvr*A (pKM101)
Concentrations in the definitive study/method	0.2, 0.5, 1.6, 5, 16 and 50 µg/plate
Basis of concentration selection	Excessive cytotoxicity at 100 µg/plate
Formulation/vehicle	DMSO
Results	(b) (4) impurity increased the number of colonies in TA102 under the conditions of this assay, with a 2-fold increase in revertant colonies seen at 50 µg/plate (and close to 2-fold at 16 µg/plate). The parent drug was also positive in strain TA102 in the Ames assay. This impurity was able to be tested at a higher top concentration than the parent drug. As noted for the parent drug, Ames positivity has been reported in other DNA gyrase inhibitors in strain TA102. Despite the positive finding in TA102, this impurity is unlikely to present an increased risk of mutagenicity to patients because this is a known finding in this bacterial strain with other drug with similar mechanisms.
Validity	Valid

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; DNA, desoxyribonucleic acid; GLP, good laboratory practice; no., number; RRT, relative retention time

Table 86. Genetic Toxicity Evaluation of (b) (4) Using the Mouse Lymphoma Cell Thymidine Kinase Locus Assay

Study Features and Methods	Details
Study no.	G19209
Study title	(b) (4) In Vitro Mammalian Cell Gene Mutation Test Using the Thymidine Kinase Gene
Conducting laboratory	(b) (4)
GLP compliance	Statement that OECD, FDA GLP were followed, but certification is from the (b) (4)
Drug, lot #, % purity	(b) (4) Batch no. CZOF/CZOF10-199-0373213, 99.7% pure
Cell line	Mouse Lymphoma L5178Y $TK^{+/-}$ −3.7.2C
Concentrations in the definitive study	7.62, 30.47, 121.86 and 487.44 µg/mL
Basis of concentration selection	Cytotoxicity, (b) (4) inhibition of relative total growth (RTG) observed at highest concentration of test article ±S9 in preliminary test for cytotoxicity
Negative control	DMSO
Positive control	−S9: 10 µg/mL methyl methanesulfonate +S9: 12 µg/mL cyclophosphamide

Study Features and Methods	Details
Formulation/vehicle	DMSO
Incubation and sampling time	3 hours +S9 3 hours −S9 24 hours −S9
Results	(b) (4) was not genotoxic (mutagenic or clastogenic) under the conditions of this assay. Increases in neither small nor large colonies were observed in the presence of the test article.
Validity	Yes

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; GLP, good laboratory practice; no., number; OECD, Organization for Economic Cooperation and Development; RRT, relative retention time

Genetic Toxicology Testing (b) (4)

Table 87. Genetic Toxicology, In Vitro Reverse Mutation Assay of (b) (4) in Bacterial Cells (Ames)

Study Features and Methods	Details
Study no.	G19208
Study title	(b) (4) Bacterial Reverse Mutation Test
Conducting laboratory	(b) (4)
GLP compliance	Yes
Drug, lot #, % purity	(b) (4) CZOF/CZOF10-200-0373217, 99.9%
Strains	*Salmonella typhimurium* TA98, TA100, TA1535, TA1537, and TA102 *E. coli* WP2*uvr*A (pKM101)
Concentrations in the definitive study/method	5, 16, 50, 160, 500 and 1600 µg/plate
Basis of concentration selection	Cytotoxicity: slight diminution of bacterial lawn at 1600 µg/plate, cytotoxicity considered excessive at ≥3200 µg/plate.
Formulation/vehicle	DMSO
Results	(b) (4) was not mutagenic under the conditions of this assay when tested at concentrations up to 1600 µg/plate
Validity	Yes

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; GLP, good laboratory practice; no., number; RRT, relative retention time

Table 88. Genetic Toxicity Evaluation of (b) (4) Using the Mouse Lymphoma Cell Thymidine Kinase Locus Assay

Study Features and Methods	Details
Study no	G19210
Study Title	(b) (4) In Vitro Mammalian Cell Gene Mutation Test Using the Thymidine Kinase Gene
Conducting laboratory	(b) (4)
GLP compliance	Statement that OECD, FDA GLP were followed, but certification is from the (b) (4)
Drug, lot #, % purity	(b) (4) Batch No. CZOF/CZOFI 0-200-0373217, 99.9%
Cell line	Mouse Lymphoma L5178Y $TK^{+/-}$ −3.7.2C
Concentrations in the definitive study	9, 27, 80, and 240 µg/mL

Study Features and Methods	Details
Basis of concentration selection	Cytotoxicity: (b) (4) inhibition of relative total growth (RTG) observed at (b) (4) µg/mL ±S9 in preliminary test for cytotoxicity.
Negative control	DMSO
Positive control	−S9: 10 µg/mL methyl methanesulfonate +S9: 12 µg/mL cyclophosphamide
Formulation/vehicle	DMSO
Incubation and sampling time	3 hours +S9 3 hours −S9 24 hours −S9
Results	(b) (4) was not genotoxic (mutagenic or clastogenic) under the conditions of this assay. Increases in neither small nor large colonies were observed in the presence of the test article.
Validity	Yes

Source: Reviewer table from Applicant's study report data
Abbreviations: DMSO, dimethyl sulfoxide; GLP, good laboratory practice; no., number; OECD, Organization for Economic Cooperation and Development; RRT, relative retention time

(Q)SAR Positive Impurities

For a drug administered at 3 g/day as a single dose, current safety standards would require further assessment of impurities exceeding (b) (4) in the final drug substance for this drug. All the potential mutagenic impurities were sufficiently removed during production to not pose an unreasonable safety risk.

Inactive Ingredients, Elemental Impurities, and Solvent Impurities

Based on the information provided, there are no inactive ingredients, elemental impurities, and solvents of concern at the levels present.

Nitrosamine Risk Assessment

The Applicant conducted a nitrosamine risk assessment and determined the risk to be low. CMC reviewers for this Application agreed with that Assessment (see CMC review.)

13.1.5.6. Other Toxicology

Studies 8257615/AZ13331419, AZ13399885 and AZ13420914: Evaluation of In Vitro Phototoxicity on Balb/c 3T3 Fibroblasts Using the Neutral Red Uptake Assay

Key Study Findings

Test articles were incubated with Balb/c 3T3 mouse fibroblasts for approximately 60 minutes in the dark at 37˚C on duplicate 96-well plates and then one plate was exposed to UV-A (5 J/cm^2) and the other incubated in the dark. The IC_{50} was used to calculate the phototoxic potential.

The phototoxic potential of AZ13420914 (zoliflodacin) was considered low.

Human metabolite M3

The only major circulating metabolite in humans was M3, found at 16.3% of the total radioactivity on a 0 to 24 hour pooled plasma sample in a radiolabeled human absorption, distribution, metabolism, and excretion (ADME) study (see Section 14.2.6).

To determine the safety of the M3 metabolite:

The formation of metabolites in rats was measured in an IV study of rats (Study KMR002) administered 50 mg/kg radiolabeled zoliflodacin, as summarized in Section 13.1.4. In this study, M3 was present in the plasma as 10.3% of the total radiolabeled products.

Direct measurement of the M3 metabolite for pharmacokinetics assessments was not available for any of the repeat-dose nonclinical toxicology studies.

The safety of IV administered zoliflodacin was tested in a 14-day repeat dose IV study in rats, in which the NOAEL was 250 mg/kg/day, the highest dose tested, corresponding to an exposure of 1901 to 3646 µg*h/mL in male and female rats, respectively. Presuming similar metabolism in this IV study to that in the previous IV administered radiolabeled zoliflodacin study, the expected exposure to M3 at the NOAEL would have been 196 to 376 µg*h/mL in male and female rats, respectively.

In a clinical trial using radiolabeled zoliflodacin, M3 was present in the plasma as 16.3% of the total radiolabeled products. The mean exposure calculated for zoliflodacin was 353 µg*h/mL, leading to expected exposure of 58 µg*h/mL.

Therefore, the safety the M3 metabolite at exposures greater than the expected clinical exposures has been adequately tested.

13.2. Individual Reviews of Studies Submitted With the New Drug Application

Studies submitted to the NDA are reviewed in Section 14.

14. Clinical Pharmacology

This section contains the clinical pharmacology review team's assessments and conclusions from the clinical pharmacology pertinent studies submitted by the Applicant. Any differences between the Applicant's and FDA review team's conclusions are noted specifically.

14.1. In Vitro Studies

14.1.1. Pharmacodynamic Studies

The zoliflodacin PK-PD relationship that best correlated with bacterial burden reduction and the suppression of emergence of resistance for *N. gonorrhoeae* was the zoliflodacin *f*$AUC_{0-\infty}$ relative to the MIC.

Neisseria gonorrhoeae lacks an established animal model, as the pathogen is human-specific. Therefore, in Study PC0914-2024-0001, zoliflodacin was studied in a dynamic in vitro HFIM, with *N. gonorrhoeae* reference strains WHO-F (susceptible to all relevant antimicrobials) and WHO-X (extensively drug resistant, including resistance to ceftriaxone) over 7 days (See Table 89 below).

Table 89. Susceptibility, Phenotypic, and Genetic Characteristics of *Neisseria Gonorrhoeae* Strains

Strain characteristics	WHO F (Unemo et al., 2016)	WHO X (Unemo et al., 2016)
Zoliflodacin MIC (microbroth MIC)[a]	0.064 (0.125)	0.125 (0.25)
Ceftriaxone (MIC)[a]	<0.002	2
Cefixime (MIC)[a]	<0.016	4
Ciprofloxacin (MIC)[a]	0.004	>32
Azithromycin (MIC)[a]	0.125	0.5
GyrB codon D429, K450, S467	WT	WT
GyrA codon S91, D95	WT	S91F, D95N
mtrR promoter region 13 bp inverted repeat	WT	deletion of A
mtrR coding region	WT	WT
Mosaic *mtrRCDE*	-	-
PorB1b codon G120, A121	NA	G120K, A121D
NG-MAST	ST3303	ST4220
NG-STAR	ST2	ST226
MLST	ST10934	ST7363

MIC, minimum inhibitory concentration; WT, wild type; NA, not applicable; NG-MAST, *N. gonorrhoeae* multiantigen sequence typing; ST, sequence type; NG-STAR, *N. gonorrhoeae* Sequence Typing Antimicrobial Resistance; MLST, multi-locus sequence typing.
[a]MIC (mg/L) was determined using agar dilution and microbroth methods for zoliflodacin, and Etest (bioMérieux, Marcy-l'Etoile, France) for ceftriaxone, cefixime, ciprofloxacin, and azithromycin.

Source: Study Number: PC0914-2024-0001 Table 1

In HFIM studies, zoliflodacin-containing broth medium solution was introduced by a syringe pump, then isovolumetrically replaced by drug-free medium to simulate drug clearance observed in humans from the plasma (i.e., humanized exposures). HFIM studies included dose-range and dose-fractionation evaluations for relationship between zoliflodacin concentration and bacterial colony forming units over time using humanized exposures for each of the following dosing regimens:

Dose-range study: single doses of 0.5, 1, 2, 3, 4, 6, and 8 g against the WHO-F and WHO-X reference strains

Dose-fractionation study: TDDs of 1, 2, 3, and 4 g per day administered as two or three divided doses (i.e., half of TDD every 12 hours, or one-third TDD every 8 hours) against the WHO-X reference strain

Dose-range study findings: Untreated control arms in the HFIM reached a bacterial density of 10^{10} to 10^{11} colony forming units/mL at the 24-hr time point and maintained growth throughout the 7-day experiments. All doses of zoliflodacin studied resulted in rapid bacterial killing within the first 6.5 hours for both strains, but only single doses ≥1 g zoliflodacin suppressed regrowth of *N. gonorrhoeae* strain WHO-F and single doses ≥2 g zoliflodacin suppressed regrowth of

N. gonorrhoeae strain WHO-X, over the 7-day evaluation period. All regimens which had regrowth selected for zoliflodacin-resistant mutants.

Dose-fractionation study findings: Both single and divided doses of a TDD of ≥2 g were sufficient to prevent regrowth of WHO-X strain for 7 days, confirming that the activity of zoliflodacin was more concentration-dependent than time-dependent.

An *f*$AUC_{0-\infty}$/MIC ratio of 70.6 was associated with the suppression of emergence of resistance for *N. gonorrhoeae.* The Applicant utilized the nonclinical PK-PD information along with PTA analyses to inform the proposed dose selection for the phase 3 trial. The Applicant hypothesized that a single oral dose of 3 g zoliflodacin should be effective in treating most gonococcal infections as well as in suppressing resistance emergence.

The Applicant also conducted additional in vitro and in vivo PK-PD studies using non-*N. gonorrhoeae* pathogens. These studies were not considered for the purposes of this review.

14.1.2. Pharmacokinetic Studies

14.1.2.1. Plasma Protein Binding

Zoliflodacin (AZ13420914) plasma protein binding (PPB) has been studied in dog, rat, mouse, guinea pig, and human plasma (Study KPJ005/AZ13420914).

The reported PPB values were determined by an equilibrium dialysis method using [^{14}C]-AZ13420914 concentrations of 1, 5, and 50µM. PPB estimates were independent of concentration, and broadly similar across species. The unbound zoliflodacin fraction, expressed as a percentage, ranged from 15.8% to 18.4% in human plasma, 20.6% to 23.2% in mouse plasma, 14.1% to 21.9% in rat plasma, 16.8% to 19.9% in dog plasma, and 23.9% to 26.8% in guinea pig plasma.[28]

The review of Study KPJ005/AZ13420914 suggests that zoliflodacin is not highly bound to plasma proteins (<90%) with the bound zoliflodacin fraction (expressed as a percentage) ranging from 81.6% to 84.2% in human plasma. The mean percentage unbound zoliflodacin fraction in human plasma was 16.97%±0.93%, resulting in 83.03% bound fraction.[29] Therefore, the review team agrees with the Applicant's proposal to note a mean value of 83% bound to human plasma proteins in the product labeling, and the use of this PPB value to support PK-PD analyses of unbound exposures.

14.1.2.2. Drug Metabolism

The metabolism of zoliflodacin was investigated in mouse, rat, dog, and human hepatocytes (Study KMN004/AZ13420914), human liver microsomes (Study KMZ227/AZ13420914), and using human recombinant CYP enzymes (Studies SN-000914-2016-01 and KMZ227/AZ13420914).

[28] Table 5 to 9 Study AZ13420914 KPJ005, pages 14 to 16

[29] Reviewer calculated

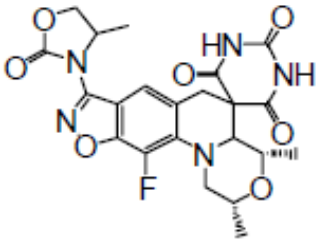
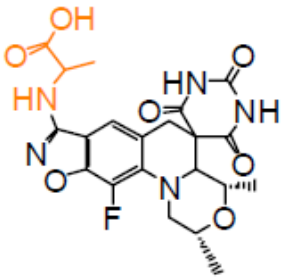
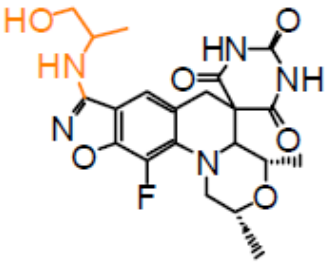
In Study KMN004/AZ13420914, after incubation of zoliflodacin (1µM and 10µM) in mouse, rat, dog, and human hepatocytes for 4 hours, a total of 10 metabolites of zoliflodacin were observed across species. In human hepatocytes, metabolites accounted for less 5% of the total radioactivity, with metabolites M1, M2, and M6 detected each with <2% of the total radioactivity.

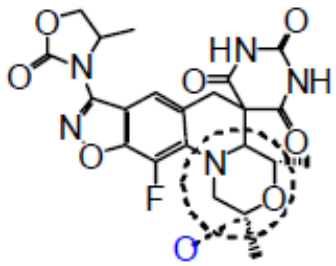
Table 90. Quantitative Estimates of Zoliflodacin and Metabolites Following 4-Hour Incubation in Human Hepatocytes

	Concentration	
	[1µM]	[10µM]
Components	**% Total Peak Area Radioactivity**	
Zoliflodacin	98.7	95.3
M1	ND	1.8
M2	ND	1.0
M6	ND	0.7

Source: Reviewer compiled from Table 1 of Study KMN004/AZ13420914 report (page 16)
Abbreviation: M, metabolite; ND: not detected

Table 91. Structures and Description of Zoliflodacin and Metabolites Following 4-Hour Incubation in Human Hepatocytes

Components	Metabolite description	Structure
Zoliflodacin	Parent	
M1	Decarboxylation	
M2	Decarboxylation	

Components	Metabolite description	Structure
M6	Oxidation	

Source: Reviewer compiled from Tables 1, 2, and 3 of Study KMN004/AZ13420914 report (pages 16 to 19)
Abbreviation: M, metabolite

In Study SN-000914-2016-01, the metabolism of zoliflodacin was studied in vitro using human recombinant enzymes CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A4. Following incubation of zoliflodacin (2µM) with enzymes, zoliflodacin was found to be mainly metabolized by enzyme CYP3A4 (68.4% contribution), with lesser metabolism contributed by CYP1A2 (14.1%), CYP2C9 (10.0%), CYP2C8 (4.9%), and CYP2C19 (2.7%). No metabolites were detected in incubation with CYP2D6.

In Study KMZ227/AZ13420914, the metabolism of zoliflodacin was studied in vitro using human recombinant enzymes CYP1A1, CYP1A2, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, CYP2E1, CYP2J2, CYP3A4, or CYP3A5. In incubations with human recombinant CYP3A4 or CYP3A5 enzymes, metabolites M2 and M6 were observed (see Table 92). In incubations with human liver microsomes and 10µM zoliflodacin concentration with and without ketoconazole, a selective CYP3A inhibitor, only metabolite M6 was observed in the absence of the inhibitor. Formation of metabolite M6 was completely inhibited by the addition of ketoconazole.

Table 92. Quantitative Estimates of Zoliflodacin Metabolites Following Incubation With Recombinant Human Cytochrome P450 Enzymes

Isolate	CYP3A4	CYP3A5
Concentration	[1µM]	[1µM]
Components	% Total Peak Area Radioactivity	
Zoliflodacin	30.0	76.3
M2	8.2	5.3
M6	47.0	18.4

Source: Reviewer compiled from Table 3 of Study KMZ227/AZ13420914 report (page 12)
Abbreviations: CYP, cytochrome P 450, M, metabolite

Overall, the studies' findings showed that zoliflodacin is significantly metabolized and is a substrate of CYP enzyme CYP3A4/5.

14.1.2.3. CYP Enzyme-Mediated Drug-Drug Interactions

The Applicant conducted in vitro enzyme-mediated DDI studies evaluating CYP enzyme inhibition and induction potentials of zoliflodacin.

Zoliflodacin in vitro inhibition studies were conducted with pooled human liver microsomes (Studies 00005CYP_INH_LCMS_2_AZ13420914, ADME-AZS-Wave3-120914, and ADME-AZS-Wave3-130613).

In Study ADME-AZS-Wave3-120914, zoliflodacin's inhibition potential towards CYPs 1A2, 2C9, 2C19, 2D6, and 3A4/5 was evaluated using concentrations ranging from 25µM to 500µM). Zoliflodacin was observed to demonstrate reversible inhibition of CYP2C9 and CYP2C19 with mean IC_{50} estimates of 239µM and 282µM, respectively. No reversible inhibition of CYP1A2, CYP2D6, or CYP3A4/5 was observed.

In Study 00005CYP_INH_LCMS_2_AZ13420914, zoliflodacin was observed to demonstrate reversible inhibition of CYP2C8 with an IC_{50} estimate of 137µM. No reversible inhibition of CYP2B6 or CYP3A4/5 was observed over the evaluated test concentrations (25µM to 500µM).

In Study ADME-AZS-Wave3-130613, no time dependent inhibition was observed for zoliflodacin at 50µM or 500µM concentration towards enzymes CYP1A2, CYP2C9, CYP2C19, CYP2D6, and CYP3A4/5.

Based on the International Council for Harmonisation (ICH) guidance for industry *M12 Drug Interaction Studies*,[30] in vivo thresholds were determined by the reviewer for zoliflodacin as an inhibitor of potentially significant clinical DDIs (see Table 93). IC_{50} estimates utilized for DDI predictions were 137µM for CYP2C8, 239µM for CYP2C9, 282µM for CYP2C19, and >500µM for CYP1A2, CYP2B6, CYP2D6, and CYP3A4/5, with a predicted unbound zoliflodacin C_{max} of 10µM. The unbound C_{max} was predicted based on: (i) zoliflodacin C_{max} of 28500 ng/mL or 58µM following a 3 g zoliflodacin single oral dose and (ii) unbound fraction of 0.17 based on in vitro data in human plasma. No or low CYP inhibition potential was predicted based on using the Drug Interaction Database DDI Calculator® utilizing parameter estimates as follows:

1. Fraction absorbed (F_a) after oral administration assumed as 1,
2. Fraction available (F_g) after intestinal metabolism assumed as 1,
3. Absorption rate constant (k_a) in vivo assumed to be 0.1,
4. Blood-to-plasma ratio of 0.69 based on in vitro data, and
5. Nominal in vitro drug concentrations (e.g., IC_{50}, based on amount of drug that is expected to be present).[31]

Less than 25% of zoliflodacin was consumed when incubated with CYP enzymes in Study SN-000914-2016-01, discussed previously. The inhibition potential of zoliflodacin by the basic model is greater than the threshold of concern (i.e., 1.02) for enzymes CYPs 2C8, 2C9, and 2C19 using IC_{50} values from Study ADME-AZS-Wave3-120914. The inhibition potential of zoliflodacin by the mechanistic static model is between 0.80 to 1.25 for enzymes CYPs 2C8, 2C9, and 2C19, therefore the risk of a clinically relevant interaction is low (Table 93).

Zoliflodacin induction studies were conducted with human donor hepatocyte cultures (Study PR11157/CC0383) incubated with concentrations ranging from 2.06µM to 500µM for 24 hours. Zoliflodacin induced CYP1A2 mRNA 14.4-fold (10.0% of positive control response) Zoliflodacin did not induce CYP2B6 or CYP3A4 mRNA (Table 93).

[30] ICH guidance for industry M12 Drug Interaction Studies (August 2024), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/m12-drug-interaction-studies.

[31] (b) (4)

Table 93. In Vitro Assessment of Zoliflodacin as Substrate, Inhibitor, or Inducer of CYP Enzymes

	In Vitro Findings			In Vivo Potential Predictions[f]		
Enzyme	% Drug Consumed After Incubation[a]	IC_{50}[b,c] (μM)	Induction FC[d] (%Positive Control)	Basic Model Predictions R_1 and/or R_{gut} (Reviewer Analysis)	Mechanistic Static Model Predictions Liver AUCR (Reviewer Analysis)	Interpretation (Substrate/ Inhibitor/ Inducer)
CYP1A2	14	>500	14.4[e] (10.63)	R_1<1.02		Inducer
CYP2B6	NT	>500	NI	R_1<1.02[g]		
CYP2C8	11	137		R_1=1.073	1.084	
CYP2C9	14	239		R_1=1.042	1.048	
CYP2C19	10	282		R_1=1.035	1.041	
CYP2D6	9	>500		R_1<1.02[g]		
CYP3A4/5	17	>500	NI	R_1<1.02[g] R_{gut} <50.241[g]		Inhibitor at GI site

Source: Reviewer's analysis. Reviewer compiled data from Table 8-1 Study SN-000914-2016-01 (page 14), Table 1 of Study ADME-AZS-Wave3-130613 report (page 6), Table 1 of Study ADME-AZS-Wave3-120914 report (page 7), Table 1 of Study 00005CYP tNH LCMS 2 AZ13420914 report (page 7), Table 3 of Study PR11157/CC0383 report (page 12), and Table A2 of Study PR11157/CC0383 report (page 16).

[a] Determined by 100% − (%drug remaining at 25 min).

[b] Highest concentration evaluated was 500μM (50× maximum clinical concentration).

[c] No evidence of time-dependent metabolism (IC_{50}, 30 min incubation / IC_{50}, 30 min incubation + NADPH <2).

[d] Human hepatocytes (mRNA expression); 3 cell lots up to 500μM zoliflodacin (anticipated unbound zoliflodacin concentration =85μM).

[e] Fold change (FC) observed at 500μM (50× $C_{max,u}$); concentration dependent increase in CYP450 enzyme mRNA observed over range of 2 to 500μM.

[f] Predictions based on equations and thresholds specified in the 2024 ICH M12 guidance document using (b) (4) / Accessed September 25, 2025.

[g] Estimate (R1 or R_{gut}) calculated with an assumed IC_{50} value of 500μM.

Abbreviations: ADME, absorption, distribution, metabolism, and excretion; AUCR, ratio of the area under the concentration versus time curve of the substrate drug in the presence and absence of the precipitant drug; $C_{max,u}$, maximum unbound plasma concentration; CYP, cytochrome P450; DDI, drug-drug interaction; FC, fold change; GI, gastrointestinal; IC_{50}, half-maximal inhibitory concentration; ICH, International Council for Harmonisation; mRNA, messenger ribonucleic acid; NADPH, nicotinamide adenine dinucleotide phosphate; NI: no induction; R1, ratio using basic model of reversable inhibition in liver; R_{gut}, ratio using basic model of reversable inhibition in intestine

Overall, zoliflodacin has no or low potential as a direct inhibitor of CYPs 1A2, 2B6, or 2D6, and zoliflodacin has a low risk of a clinically relevant interaction as a direct inhibitor of CYPs 2C8, 2C9, and 2C19. Although zoliflodacin demonstrated induction of CYP1A2 in vitro, the risk of a clinically relevant interaction is unlikely as NUZOLVENCE is a single-dose drug product and zoliflodacin has a median $t_{1/2}$ of approximately 6 hours.

Zoliflodacin theoretically has the potential to inhibit intestinal CYP3A4/5 at concentrations >500μM, the highest concentration evaluated in the in vitro studies. No reversible or time-dependent inhibition of CYP3A4/5 was observed in the in vitro studies at ≤500μM (50× maximum clinical concentration). The potential risk associated with zoliflodacin inhibition of intestinal CYP3A4/5 could be transient as its inhibition effect (if any) may be restricted to the time when the zoliflodacin concentration in intestinal fluid is >500μM. Zoliflodacin has been shown to be rapidly absorbed with a median T_{max} of 2.5 and 4 hours under fasted and fed conditions, respectively, and a $t_{1/2}$ of approximately 6 hours (see Section 14.2.2). While we cannot rule out zoliflodacin's potential to inhibit intestinal CYP3A4/5 based on the data submitted, we will not pursue further studies of the DDI of zoliflodacin as an inhibitor of intestinal CYP3A4/5 considering the available in vitro DDI data up to 500μM concentrations and potential solubility related limitations.

To assess the effect of other medications on zoliflodacin pharmacokinetics following administration of a single 3 g dose, PBPK modeling was used prospectively to predict the likely outcomes of DDIs with CYP3A4/5 inducers and inhibitors. To assess zoliflodacin's inhibition potential following administration of a single 3 g dose, PBPK modeling was used prospectively to predict the likely outcomes of DDIs with substrates of CYP2C9. See Section 14.5.2 for details on PBPK modeling review.

A clinical DDI study was conducted with zoliflodacin and the strong CYP3A4 inhibitor itraconazole as a precipitant, which is discussed in Section 14.2.1.

14.1.2.4. Transporter-Mediated Drug-Drug Interactions

The Applicant conducted in vitro transporter-mediated DDI studies evaluating hepatic and renal transporter substrate and inhibition potentials of zoliflodacin.

Studies evaluated P-gp-, BCRP-, and OATP1 B1/B3-mediated transport of zoliflodacin in overexpressing cell culture monolayers (Study 13ASTRUKP7R7S13).

Studies also evaluated P-gp, OATP1 B1/B3, OAT 1/3, organic cation transporter 2 (OCT2), MATE1, and MATE2K transport inhibition by zoliflodacin in overexpressing cell culture monolayers (Studies INF.000-133-903, 13ASTRUKP7R7S13, and 13072012_OATP1B1_inhib_AZ13420914). BCRP transport inhibition by zoliflodacin was evaluated in Caco-2 cell culture monolayers stably expressing the human efflux transporter BCRP (Study ADME-AZS-Wave3-150213).

Based on the 2024 ICH M12 DDI Guidance document,[32] in vivo thresholds were determined for zoliflodacin as either an object or precipitant of potentially significant clinical DDIs (see Table 94).

The aforementioned in vitro study findings demonstrated zoliflodacin is a substrate of drug transporters P-gp and BCRP (>2 in absence of inhibitor, and net flux ratio significantly decreased in the presence of an inhibitor). Zoliflodacin has inhibition potential for P-gp, BCRP, OATP1 B1/B3, OAT1/3, MATE1, and MATE2K drug transporters. Zoliflodacin was not observed to inhibit the OCT2 drug transporter.

Zoliflodacin did not demonstrate induction of CYP3A, therefore induction of P-gp is unlikely due to common pathways (pregnane X receptors and/or constitutive androstane receptors) per ICH M12 Guidance.[33]

The summary of in vitro assessment of transporters mediated DDIs for zoliflodacin is presented in Table 94. In this summary assessments, the assumptions for predicting unbound C_{max} were same as the assumptions noted above for the CYP enzyme-mediated DDI potential evaluations.

[32] See Footnote 29

[33] Ibid.

Table 94. In Vitro Assessment of Zoliflodacin as Substrate, Inhibitor, or Inducer of Human Uptake and Efflux Transporters

	In Vitro Findings		In Vivo Potential Predictions[e]	
Transporter	Maximum Flux Rate Ratio Range	IC_{50} (µM)	Basic Model Predictions (Reviewer Analysis)	Interpretation (Substrate/ Inhibitor/ Inducer)
BCRP	3.56 to 32.2	55.5	I_1[b]/IC_{50}=0.179, ≥0.02 cut-off limit	Substrate
			I_{gut}[c]/ C_{50}=443.611, ≥10 cut-off limit	Inhibitor
P-gp	19.7 to 373	299	I_1[b]/IC_{50}=0.033, ≥0.02 cut-off limit	Substrate
			I_{gut}[c]/IC_{50}=82.343, ≥10 cut-off limit	Inhibitor
OATP1B1	<2	7.7	I_{in}[d]/IC_{50}=1.291, ≥0.1 cut-off limit	Inhibitor
OATP1B3	<2	9.23	I_{in}[d]/IC_{50}=1.077, ≥0.1 cut-off limit	Inhibitor
OAT1	NT[a]	47.7	I_1[b]/ C_{50}=0.208, ≥0.1 cut-off limit	Inhibitor
OAT3	NT[a]	13.5	I_1[b]/IC_{50}=0.736, ≥0.1 cut-off limit	Inhibitor
OCT2	NT[a]	-		
MATE1	NT[a]	>100	I_1[b]/IC_{50}=0.099, ≥0.02 cut-off limit	Inhibitor
MATE2	NT[a]	>100	I_1[b]/ IC_{50}=0.099, ≥0.02 cut-off limit	Inhibitor

Source: Reviewer's analysis. Reviewer compiled from Table 1 Study 13ASTRUKP7R7S13 (page 11), Table 1 Study13072012_OATP1B1_inhib_AZ13420914 (page 6), Table 1 Study ADME-AZS-Wave3-150213 (page 5), and Table 1 Study INF.000-133-903 (pages 6 to 7).

[a] Renal clearance of parent drug zoliflodacin is <25% of total zoliflodacin clearance.

[b] I1 = C_{max},u is estimated unbound maximum plasma concentration of an inhibitor, C_{max},u = fu,p × C_{max}. The C_{max} 28.5 µg/mL or 10µM and fu,p 0.17 were used in estimation.

[c] Igut = Dose/250 mL.

[d] Iin = C_{max},inlet,u is estimated unbound maximum plasma concentration of an inhibitor at liver inlet. C_{max},inlet,u = fu,p × (C_{max} + (Fa×Fg×ka×Dose)/Qh/RB) The C_{max} 28.5 µg/mL and fu,p 0.17 were used in estimation. Assumptions Fa =1, Fg =1 and k =0.1/minute were used as a worst-case estimates.
max =1.7µM; estimated max unbound plasma concentration of inhibitor at the inlet to the liver.

[e] Predictions based on equations and thresholds specified in the 2024 ICH M12 guidance document using (b) (4) Accessed September 26, 2025.

Abbreviations: BCRP, breast cancer resistance protein; DDI, drug-drug interaction; IC_{50}, half-maximal inhibitory concentration; Imax, u, maximum unbound plasma concentration of interacting drug at steady-state; MATE, multidrug and toxin extrusion transporter; NT, not tested; OAT, organic anion transporter, OATP, organic anion transporting polypeptide; OCT, organic cation transporter; R, ratio using basic model of reversable inhibition in liver and Iin, max; P-gp, P-glycoprotein; (-), not significant

To assess zoliflodacin's inhibition potential following administration of a single 3-g dose, PBPK modeling was used prospectively to predict the likely outcomes of zoliflodacin DDIs with substrates of the following transporters: P-gp, BCRP, OAT1, OAT3, OATP1B1, OATP1B3, OATP1B, and MATE. See Section 14.5.2 for details on PBPK modeling review.

No clinical studies were conducted to further evaluate zoliflodacin DDIs as either a victim or inhibitor of transporters.

14.2. In Vivo Studies

14.2.1. Formulation Development

Formulation Development History

The original formulation developed and manufactured by Astra Zeneca was a zoliflodacin powder for oral suspension (ZoliAZ PFOS) formulation. The phase 2 trial evaluated the efficacy of the ZoliAZ PFOS formulation.

The original formulation, ZoliAZ PFOS, was replaced by a zoliflodacin granules for oral suspension formulation manufactured by Patheon (ZoliPa GFOS). The Applicant did not conduct

a crossover bioavailability (BA) study comparing zoliflodacin BA between the phase 2 formulation ZoliAZ PFOS and formulation ZoliPa GFOS. However, the estimated PK parameters following administration of the same dose (3g) under the same conditions with respect to food (either fed or fasted) were found to be similar between the formulations (See Section 14.2.5 and Section 14.2.2). The phase 3 trial evaluated the efficacy of the ZoliPa GFOS formulation.

The to-be-marketed zoliflodacin granules for oral suspension formulation (ZoliDr GFOS) is manufactured by Dr. Reddy's Laboratories, Ltd, India. The relative BA study between the phase 3 formulation ZoliPa GFOS and the to be marketed formulation ZoliDr GFOS was evaluated in Study STI_Zoli003 (See Section 14.2.2).

A summary of the formulations used in clinical studies included in this review is shown below in Table 95.

Table 95. Zoliflodacin Formulation Development History

Study Type	Phase 1 SAD Study	Phase 1 AME Study	Phase 2 Clinical Trial	Phase 1 PK Study	Phase 1 TQT Study	Phase 1 FE Study	Phase 3 Clinical Trial	Phase 1 BA/BE, FE, DDI Study
Study number	D4930C00001	D4930C00003	DMID 14-0014	DMID 16-0118	DMID 16-0110	STI_Zoli002	STI_Zoli001	STI_Zoli003
Clinical trial start date	Sep 17, 2013	Jan 6, 2015	Nov 25, 2014	Feb 2, 2018	Aug 17, 2018	Oct 3, 2018	Nov 6, 2019	Nov 9, 2022
Clinical trial end date	Mar 3, 2014	Feb 12, 2015	Dec 30, 2015	Mar 2, 2018	Jan 2, 2019	Nov 12, 2018	Mar 16, 2023	Feb 7, 2023
Formulation used	ZoliAZ PFOS[a]	ZoliAZ PFOS[a]	ZoliAZ PFOS[a]	ZoliPa GFOS[b]	ZoliPa GFOS[b]	ZoliPa GFOS[b]	ZoliPa GFOS[b]	ZoliPa GFOS[b] and ZoliDr GFOS[b]
Zoliflodacin dose	200 mg to 4 g	3 g	2 or 3 g	4 g	2 or 4 g	3 or 4 g	3 g	3 g

Source: Reviewer compiled from Table 5, 2.7.1 Summary of Biopharmaceutic Studies and Associated Analytical Methods p. 11
[a] Zoliflodacin PFOS was suspended in water to a concentration of 50 mg/mL and administered with up to 240 mL of water.
[b] Zoliflodacin GFOS were suspended in 60 mL of water before administration, and additional water (60-240 mL) was used for rinsing the dispensing cup.
Abbreviations: AME, absorption, metabolism, excretion; BA, bioavailability; BE, bioequivalence; DDI, drug-drug interaction; FE, food effect; PK, pharmacokinetic; SAD, single ascending dose; TQT, thorough QT; ZoliAZ PFOS, zoliflodacin powder for oral suspension manufactured by Astra Zeneca; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd.

14.2.2. Human Relative Bioavailability, Food Effect, and Drug-Drug Interaction Study

Bridge Between To-Be-Marketed and Clinical Trial/Study Formulations

For the pivotal phase 3 trial, the formulation evaluated was ZoliPa GFOS. The proposed to-be-marketed product, ZoliDr GFOS, is manufactured at a different site with different processes than that used for the manufacturing of the phase 3 clinical trial formulation. Specifically, the ZoliDr GFOS formulation uses an identical formulation composition as the ZoliPa GFOS formulation, except (b) (4) . The to-be-marketed drug product, ZoliDr GFOS, was shown to have comparable zoliflodacin bioavailability (i.e., met the bioequivalent criteria under fed and fasted conditions) with the formulation used in the phase 3 clinical trials, ZoliPa GFOS.

Study STI_Zoli003

Study Design

The study's primary objective was to assess and compare the BA of two formulations of zoliflodacin granules for oral suspension, ZoliDr GFOS and ZoliPa GFOS, in healthy adult male and female participants, following a single 3 g dose under fasted and fed conditions. Additional secondary objectives included to assess the food effect (FE) on pharmacokinetics, safety, and tolerability of the two formulations under evaluation. Another secondary objective was to assess the DDI effect of multiple doses of itraconazole, a strong CYP3A4 inhibitor, on the PK parameters of a single 3-g dose of ZoliPa GFOS in fasted healthy participants.

Study STI_Zoli003 was a phase 1, open-label, single-site, randomized design with two parallel cohorts in healthy participants. Cohort 1 assessed the BA, FE, Safety, and Tolerability objectives, while Cohort 2 assessed the DDI objective.

Cohort 1 had a four-way crossover design, where participants were randomized to receive a single 3 g dose of zoliflodacin granules under either fed or fasted conditions, with a 72-hour washout between periods. For fed conditions, participants were administered a moderate-fat/moderate-calorie standardized meal, and were expected to consume the entire meal, 30 minutes prior to treatment administration. Moderate fat meal contained approximately 400 to 500 kcal with 51%, 10%, and 39% calories from carbohydrates, protein, and fat, respectively. For fasted conditions, participants fasted overnight for at least 10 hours prior to treatment administration.

Cohort 1 participants were administered the following four treatments:

Treatment A: ZoliPa GFOS fasted

Treatment B: ZoliDr GFOS fasted

Treatment C: ZoliPa GFOS fed

Treatment D: ZoliDr GFOS fed

Cohort 1 participants were randomized into four sequences of the four treatments to examine sequence and period effect.

BA and FE were assessed by a comparison of the geometric mean ratio (GMR) and the associated 90% CI of the observed zoliflodacin PK parameters, C_{max}, $AUC_{0-\infty}$. For the relative BA assessment, the reference product is ZoliPa GFOS, and for FE, the reference is fasted conditions.

Cohort 2 (DDI) had a two-period two-treatment fixed sequence crossover design, where participants received a single 3 g dose of ZoliPa GFOS under fasted conditions, followed by a 72-hour washout. Then on Day 4, participants received a 400 mg itraconazole loading dose, followed by 200 mg itraconazole once daily from Days 5 to Day 8, under fed conditions. On Day 9 participants received a 200 mg itraconazole dose followed 1 hour later by a single 3 g dose of ZoliPa GFOS, both under fasted conditions. Participants continued to receive a once daily dose of 200 mg itraconazole on Days 10 to Day 11, under fed conditions.

The DDI evaluation assessed the trough concentration (C_{trough}) of itraconazole on Day 4 to 11, and the GMR and CI of the observed zoliflodacin PK parameters $AUC_{0-\infty}$ and C_{max}, when comparing between administration with and without the precipitant drug, itraconazole.

Drug Product Formulations

ZoliPa GFOS and ZoliDr GFOS formulations described in the preceding section were evaluated in Cohort 1. ZoliPa GFOS formulation described in the preceding section and commercially available 100 mg itraconazole capsules were evaluated in Cohort 2.

PK Sampling

Blood samples for zoliflodacin plasma concentration were collected for each participant under fasting conditions (Cohort 1 Treatments A and B, Cohort 2 period 1 and 2) predose and at 0.5, 1, 2, 2.5, 3, 3.5, 4, 5, 6, 7, 9, 12, 24, and 48 hours postdose. Additional zoliflodacin blood samples were collected for Cohort 2 period two participants at 5.5, 6.5, and 72 hours postdose.

Blood samples for zoliflodacin plasma concentration were collected for each participant under fed conditions (Cohort 1 Treatments C and D) predose and at 0.5, 1, 2, 3, 4, 5, 5.5, 6, 6.5, 7, 9, 12, 24, 48 hours postdose.

Blood samples for itraconazole plasma concentration were collected for each participant in Cohort 2 period 2 predose on Days 4 through 8 and on Day 9.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by liquid chromatography with tandem mass spectrometry (LC-MS/MS) using the method TP70542 with associated validation report V1241802P1. Itraconazole concentrations were assayed by LC-MS/MS using the method TP-80142 with associated validation report V3231903P1.

Results

Cohort 1 enrolled 32 participants (n=8 per sequence; including 11 (34.4%) females) with a mean (range) age of 39.5 (25 to 55) years and the mean ± standard deviation weight of 74.19±11.218 kg. All 32 participants received at least one dose of study medication, and 31 of which received all four treatments and completed the study. One participant in Cohort 1

withdrew from the study after receiving one dose of study medication and experiencing an adverse event (rash) related to the study medication.

Cohort 2 enrolled 18 participants (including 3 (16.7%) females) with a mean (range) age of 42.3 (25 to 55) years and the mean ± standard deviation weight of 78.19±11.332 kg. All 18 participants received all planned doses of study medication and completed the study.

The descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral dose of the treatment administered under either fasted or fed conditions are summarized in Table 96. The statistical comparison of zoliflodacin's exposure parameters is summarized in Table 97.

As shown in Table 97, the to-be-marketed formulation ZoliDr GFOS (test product) and the formulation used in the phase 3 efficacy trial ZoliPa GFOS (reference product) demonstrated comparable zoliflodacin BA under fasting and fed conditions. The GMR and corresponding 90% CIs were wholly contained within the bioequivalence range criteria of 80.00% to 125.00% for the pharmacokinetic parameters C_{max}, and $AUC_{0-\infty}$. The two formulations demonstrated similar pharmacokinetic profiles (Figure 5 [fasted state] and Figure 6 [fed state]) as well as similar T_{max} and $t_{1/2}$ estimates (Table 96).

With respect to the FE on zoliflodacin pharmacokinetics, zoliflodacin BA was significantly increased due to FE, as shown in Table 97 below. The GMRs (fed/fasted) for both formulations evaluated were approximately 1.5 for both the C_{max} and $AUC_{0-\infty}$ and the 90% CIs of the GMRs were not contained within the range of 80.00% to 125.00%. Additionally, food also delayed absorption by approximately 1.5 hr for both formulations evaluated (Table 96).

With respect to the DDI findings, the administration of zoliflodacin with the strong CYP3A4 inhibitor, itraconazole, at steady state did increase the exposure of zoliflodacin with the GMRs (+itraconazole/alone) of approximately 1.03 and 1.38 for C_{max} and $AUC_{0-\infty}$, respectively, when administered under fasted conditions (Table 99).

Table 96. Summary Statistics of Plasma Zoliflodacin Pharmacokinetic Parameters by Treatment

	Geometric Mean (%GCV)					
	Cohort 1				Cohort 2	
Parameter (Unit)	Treatment A ZoliPa GFOS (Fasted) N=31	Treatment B ZoliDr GFOS (Fasted) N=31	Treatment C ZoliPa GFOS (Fed) N=32	Treatment D ZoliDr GFOS (Fed) N=31	ZoliPa GFOS alone (fasted) Day 1 N=18	ZoliPa GFOS + Itraconazole (fasted) Day 9 N=18
C_{max} (ng/mL)	19400 (24.3)	17900 (29.4)	28900 (26.2)	26700 (25.4)	17600 (31.1)	18100 (34.3)
T_{max} (h)^	3.0 (1.0 to 9.0)	2.5 (1.0 to 4.0)	4.5 (3.0 to 5.5)	4.0 (3.0 to 5.5)	2.5 (2.0 to 3.5)	3.3 (2.0 to 5.0)
$t_{1/2}$ (h)	6.17 (16.3)	6.43 (20.4)	5.44 (12.2)	5.49 (14.0)	7.26 (19.6)	9.09 (22.9)
$AUC_{0-\infty}$ (h*ng/mL)	166000 (25.6)	157000 (28.8)	250000 (28.8)	241000 (27.8)	164000 (33.7)	226000 (31.6)
CL/F (L/h)	18.1 (25.6)	19.1 (28.8)	12.0 (28.8)	12.5 (27.8)	18.3 (33.7)	13.3 (31.6)
Vz/F (L)	161 (31.8)	177 (26.6)	94.1 (22.6)	98.7 (24.1)	192 (30.6)	174 (32.7)

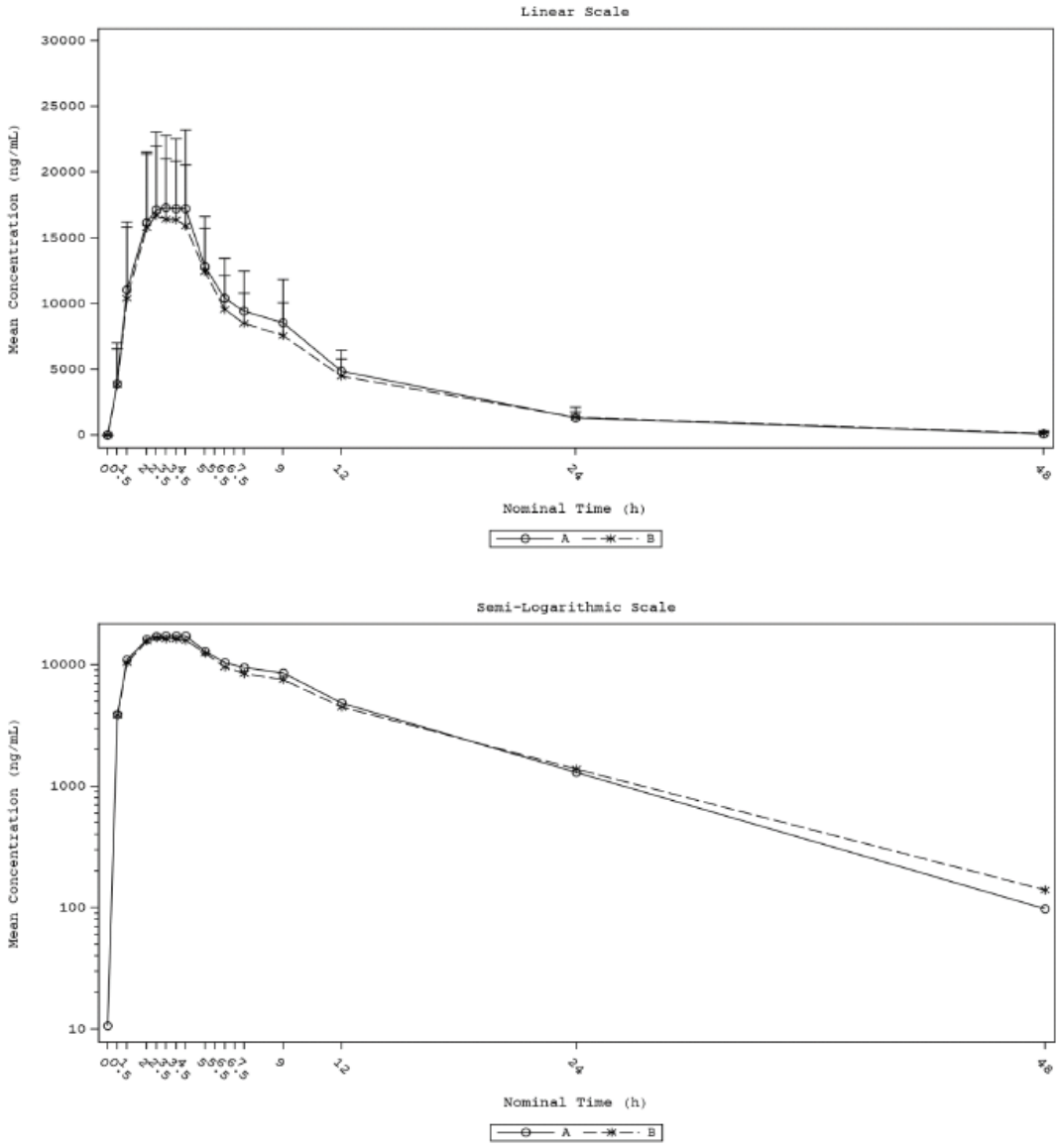
Source: Reviewer compiled from Table 12 STI_Zoli003 CSR (page 64) and Table 13 STI_Zoli003 CSR (page 66)
^Median (minimum to maximum).
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CL/F, apparent clearance; C_{max}, maximum plasma concentration; GCV, geometric coefficient of variation; N, number of participants in treatment arm; $t_{1/2}$, Half-life; T_{max}: time to maximum plasma concentration; Vz/F, apparent volume of distribution; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd

Table 97. Statistical Analysis of Plasma PK Parameters for Zoliflodacin Test and Reference Formulations

Food condition	Parameter (Unit)	Test ZoliDr GFOS GM (90% CI), n	Reference ZoliPa GFOS GM (90% CI), n	Ratio (%) (ZoliDr GFOS/ZoliPa GFOS) GMR (90% CI)	Intra CV%	p-Value
Fasted	C_{max} (ng/mL)	17900 (17000, 18800), n=31	19400 (18500, 20300), n=31	92.340 (86.20, 98.92)	16.0	0.0588
	$AUC_{0-\infty}$ (h*ng/mL)	157000 (151000, 163000), n=31	165000 (159000, 172000), n=31	95.057 (89.93, 100.47)	12.9	0.1307
Fed	C_{max} (ng/mL)	26,700 (25600, 27900), n=31	28200 (27000, 29500), n=31	94.631 (89.03, 100.58)	14.2	0.1351
	$AUC_{0-\infty}$ (h*ng/mL)	241,000 (236000, 246000), n=31	246000 (242000, 251000), n=31	97.637 (94.92, 100.44)	6.5	0.1614

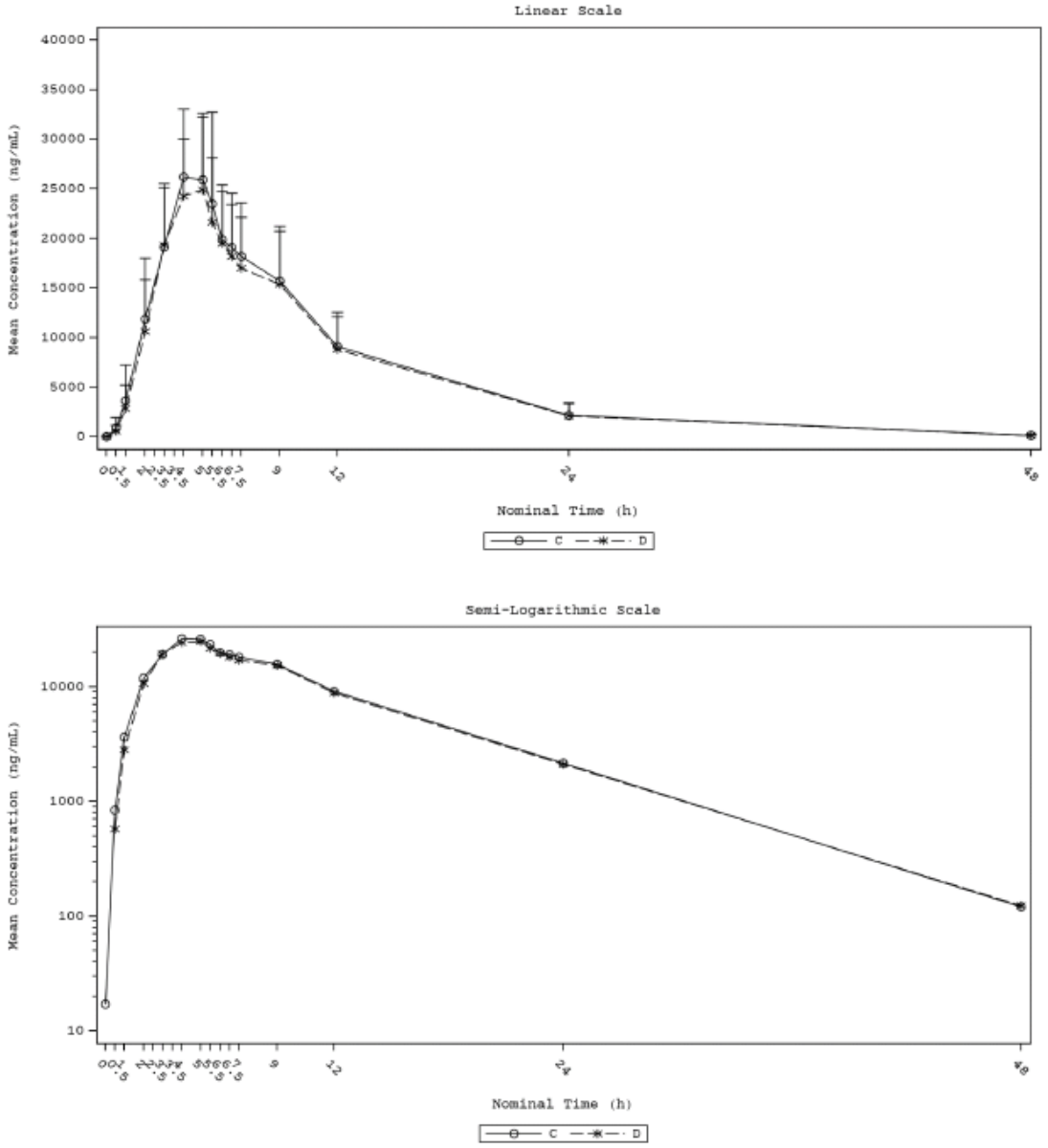
Source: Table 14 STI_Zoli003 CSR (page 67)
Abbreviations, $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CI, Confidence Interval; C_{max}, maximum plasma concentration; CV, Coefficient of Variation; GM, Geometric Mean; GMR, Geometric Mean Ratio; n, number of participants with a specific treatment parameter; PK, pharmacokinetic; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd

Figure 5. Mean (+SD) Zoliflodacin Plasma Concentration vs. Time for Treatment B (ZoliDr GFOS) vs. A (ZoliPa GFOS), Fasted State, Linear Scale and Semi-Logarithmic Scale



Source: Figure 1 STI_Zoli003 CSR (page 56)
Abbreviations: SD, standard deviation; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Figure 6. Mean (+SD) Zoliflodacin Plasma Concentration Time Data for Treatment D (ZoliDr GFOS) vs. C (ZoliPa GFOS), Fed State, Linear Scale and Semi-Logarithmic Scale



Source: Figure 2 STI_Zoli003 CSR (page 57)
Abbreviations: SD, standard deviation; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd.; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Table 98. Statistical Analysis To Assess the Effect of Food on Plasma Pharmacokinetic Parameters of Zoliflodacin

Formulation	Parameter (Unit)	Test Fed GM (90% CI), n	Reference Fasted GM (90% CI), n	Ratio (%) (Fed/Fasted) GMR (90% CI)	Intra CV%	p-Value
ZoliPa GFOS	C_{max} (ng/mL)	28200 (26800, 29600), n=31	19400 (18400, 20300), n=31	145.667 (135.81, 156.24)	16.3	<0.0001
	$AUC_{0-\infty}$ (h*ng/mL)	246000 (236000, 257000), n=31	165,000 (158000, 172000), n=31	149.060 (140.38, 158.27)	13.9	<0.0001
ZoliDr GFOS	C_{max} (ng/mL)	26700 (25300, 28200), n=31	17900 (17000, 18900), n=31	149.182 (138.48, 160.71)	17.3	<0.0001
	$AUC_{0-\infty}$ (h*ng/mL)	241000 (232000, 250000), n=31	157000 (152000, 163000), n=31	152.901 (145.20, 161.01)	12.0	<0.0001

Source: Table 15 STI_Zoli003 CSR (page 69)
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CI, Confidence Interval; C_{max}, maximum concentration; CV, Coefficient of Variation; GM, Geometric Mean; GMR, Geometric Mean Ratio; n, number of participants with a specific parameter for a treatment; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon; ZoliDr GFOS, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd.

Table 99. Statistical Analysis To Assess the Effect of Coadministration of Itraconazole on Plasma Pharmacokinetic Parameters of Zoliflodacin

Parameter (Unit)	Test ZoliPa GFOS + Itraconazole/ GM (90% CI), n	Reference ZoliPa GFOS Alone GM (90% CI), n	Ratio (%) (ZoliPa GFOS + Itraconazole/ /ZoliPa GFOS alone) GMR (90% CI)	Intra CV%
C_{max} (ng/mL)	18100 (15900, 20600), n=18	17600 (15400, 20000), n=18	103.159 (94.46, 112.66)	15.3
$AUC_{0-\infty}$ (h*ng/mL)	226000 (198000, 257000), n=18	164000 (144000, 186000), n=18	137.845 (126.64, 150.04)	14.7

Source: Table 16 STI_Zoli003 CSR (page 70)
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CI, Confidence Interval; C_{max}, maximum concentration; CV, Coefficient of Variation; GM, Geometric Mean; GMR, Geometric Mean Ratio; n, number of participants with a specific parameter for a treatment; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Assessment of Study STI_Zoli003

The Applicant's overall findings from this study are consistent with the reviewer's analysis.

An inspection of the clinical and bioanalytical sites was requested for Study STI_Zoli003.

With respect to the analytical sites, the Office of Study Integrity and Surveillance (OSIS) declined to conduct an inspection for the bioanalytical site (see memo dated June 12, 2025 (archived in DARRTS June 17, 2025)). OSIS determined that an inspection was not needed for the site, as OSIS had conducted an inspection for the analytical site in (b) (4). OSIS noted two observations of objectionable conditions during the inspection, but after review of the objectionable conditions and the written response from the site, OSIS determined that the

reported value for one study was invalid but that the remaining data from the reviewed studies were reliable.

With respect to the clinical site, the Office of Inspections and Investigations conducted an inspection for the clinical site in May 2024. OSIS concluded that data from the reviewed study were reliable.

Overall, from a perspective of clinical pharmacology, the review team agrees with following:

The study findings demonstrated comparable zoliflodacin BA between the proposed to-be-marketed product ZoliDr GFOS and the formulation used in the pivotal phase 3 trial ZoliPa GFOS, and supports a bridge between the clinical trial formulation (for the pivotal clinical study) and the proposed to-be-marketed product.

The results of the DDI study support the conclusion that dosage adjustment is not required if zoliflodacin is coadministered with CYP3A4 inhibitors.

The results of the FE assessments demonstrate a significant increase in zoliflodacin BA when administered with food. These findings are in agreement with the findings observed in other zoliflodacin FE studies STI_Zoli002 and D4930C00001, discussed below in Sections 14.2.3 and 14.2.5, respectively. The proposed administration instructions with respect to patient weight accounts for this FE, as discussed in the PTA analysis discussed below (see Section 14.5.1.1).

Study STI_Zoli003 collected standard 12-lead electrogram predose (at screening and at admission visits), and 6 hours postdose of zoliflodacin following Treatment A, B, C, and D, and at the follow-up visit. The electrogram was recorded after at least 10 minutes in the supine position. The analysis of the electrocardiogram data included an evaluation of QT prolongation. Data from this study were used in analysis that evaluated QTc prolongation risk for a single oral dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more (See Section 14.2.9).

14.2.3. Human Relative Bioavailability and Food Effect

Study STI_Zoli002

Study Design

The study's primary objective was to assess the PK parameters of zoliflodacin granules for oral suspension, ZoliPa GFOS, in healthy adult male and female participants, following a single 3 g or 4 g dose under fasted and fed conditions. Additional secondary objectives included to assess the FE, safety, and tolerability of the formulation under evaluation. Another secondary objective was to assess the change from predose Baseline QT interval corrected for heart rate using Fridericia's formula (QTcF) of the formulation under fasted and fed conditions.

The study was a phase 1, parallel, open-label, single-site, randomized, crossover design with two parallel cohorts in healthy participants. Study participants were administered the following treatment regimens:

Cohort 1 Treatment A: ZoliPa GFOS, 3 g, fasted

Cohort 1 Treatment B: ZoliPa GFOS, 3 g, fed

Cohort 2 Treatment A: ZoliPa GFOS, 4 g, fasted

Cohort 2 Treatment B: ZoliPa GFOS, 4 g, fed

Participants were randomized to each Cohort and received both treatments (i.e., A or B), with a 4-day washout between periods. For fed conditions, participants were administered a high-calorie, high-fat breakfast, and were expected to consume the entire meal, 30 minutes prior to treatment administration. For fasted conditions, participants fasted overnight for at least 10 hours prior to treatment administration.

FE was assessed by a comparison of the GMR and the associated 90% CI of the observed zoliflodacin PK parameters, C_{max}, and $AUC_{0-\infty}$. For FE evaluation, the reference treatment arm was fasted conditions (Treatment A).

Drug Product Formulations

ZoliPa GFOS formulation described in the preceding section was evaluated.

PK Sampling

Blood samples for zoliflodacin plasma concentration were collected for each participant predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 9, 12, 24, and 48 hours postdose.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method TP70542 with associated validation report V1241802P1 (see Section 14.3).

Results

Cohort 1 enrolled 24 participants (including 11 (45.8%) females) with a mean (range) age of 38.0 (19 to 53) years and the mean ± standard deviation weight of 75.13±11.40 kg. Cohort 2 enrolled 24 participants (including 12 (50.0%) females) with a mean (range) age of 41.4 (19 to 55) years and the mean ± standard deviation weight of 72.75±11.87 kg. In total, 48 participants were enrolled and received at least one dose of study medication (n=24 for each Cohort), and 47 participants completed the study.

Summaries of the descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral dose of the treatment administered under either fasted or fed conditions are presented in Table 100.

Zoliflodacin BA was significantly increased when administered with food, as shown in Table 101 below. The exposure parameter estimate GMRs (fed/fasted) were approximately 1.5 for C_{max} and 2.0 for $AUC_{0-\infty}$ when a 3 g single dose of zoliflodacin is administered with a high-calorie, high-fat meal. The FE is even more pronounced with a 4 g single dose of zoliflodacin administered with a high-calorie, high-fat meal, with GMRs of approximately 1.9 for C_{max} and 2.1 for $AUC_{0-\infty}$. The 90% CIs of the GMRs were not contained within the range of 80.00% to 125.00%. Additionally, as shown in Table 100, food delayed absorption by between 1.5 to 3 hr for the doses evaluated.

Table 100. Summary Statistics of Plasma Zoliflodacin Pharmacokinetic Parameters by Treatment

	Geometric Mean (%GCV), n			
	Cohort 1		Cohort 2	
Parameter (Unit)	Treatment A ZoliPa GFOS (3 g Fasted) N=24	Treatment B ZoliPa GFOS (3 g Fed) N=23	Treatment A ZoliPa GFOS (4 g Fasted) N=24	Treatment B ZoliPa GFOS (4 g Fed) N=24
C_{max} (ng/mL)	17600 (21.3), n=24	27100 (24.7), n=23	19900 (28.1), n=24	37500 (24.7), n=24
T_{max} (h)^	3.0 (1.0 to 9.0), n=24	6.0 (3.0 to 12.0), n=24	2.5 (1.0 to 12.0), n=24	4.0 (2.5 to 9.0), n=24
$t_{1/2}$ (h)	6.474 (16.7), n=24	6.015 (13.5), n=22	6.945 (23.9), n=22	5.838 (13.4), n=23
$AUC_{0-\infty}$ (h*ng/mL)	171000 (29.4), n=24	343000 (21.2), n=22	224000 (36.6), n=22	486000 (24.0), n=23
CL/F (mL/min)	292 (29.4), n=24	146 (21.2), n=22	298 (36.6), n=22	137 (24.0), n=23
Vz/F (L)	163 (30.0), n=24	75.8 (15.6), n=22	179 (44.6), n=22	69.3 (24.1), n=23

Source: Reviewer compiled from Table 8 of STI_Zoli002 CSR (pages 35 to 39)
^Median (Minimum to Maximum).
Abbreviations: $AUC_{0-\infty}$: area under the time-concentration curve from zero to infinity; CL/F: apparent clearance; C_{max}: maximum concentration; GCV: geometric coefficient of variation; N: The number of participants included in the pharmacokinetic population for each treatment; n: the number of participants with a specific parameter; $t_{1/2}$: Half-life; T_{max}: time C_{max} observed; Vz/F: apparent volume of distribution; ZoliDr GFOS: zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd; ZoliPa GFOS: zoliflodacin granules for oral suspension manufactured by Patheon

Table 101. Statistical Analysis To Assess the Effect of Food on Plasma Pharmacokinetic Parameters of Zoliflodacin

		Test: Fed	Reference: Fasted	Ratio (%) (Fed/Fasted)		
Formulation and Dose	Parameter (Unit)	GM, n	GM, n	GMR (90% CI)	Intra CV%	p-Value
ZoliPa GFOS 3 g	C_{max} (ng/mL)	27100 n=23	17800 n=23	152.39 (139.30, 166.71)	17.8	<0.001
	$AUC_{0-\infty}$ (h*ng/mL)	345000 n=22	172000 n=22	200.87 (185.10, 217.99)	15.8	<0.001
ZoliPa GFOS 4 g	C_{max} (ng/mL)	37500 n=24	19900 n=24	188.72 (166.42, 214.00)	26.4	<0.001
	$AUC_{0-\infty}$ (h*ng/mL)	480000 n=21	225000 n=21	213.61 (182.03, 250.66)	30.6	<0.001

Source: Reviewer compiled from Table 9 of STI_Zoli002 CSR (page 41) and Table 10 of STI_Zoli002 CSR (page 43)
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CI: Confidence Interval; C_{max}: maximum concentration; CV, Coefficient of Variation; GM, Geometric Mean; GMR, Geometric Mean Ratio; n, number of participants with a specific parameter for a treatment; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Assessment of Study STI_Zoli002

The Applicant's overall findings from this study are consistent with the reviewer's analysis.

Study STI_Zoli002 Cohorts 1 and 2, both Periods, collected standard 12-lead electrogram predose (at screening) in triplicate approximately 2 min apart after the participant was in the supine position for a minimum of 10 minutes at the predose time point. Additionally, Study STI_Zoli002 Cohorts 1 and 2, both Periods, collected continuous 12-lead Holter electrogram 1 hr before dosing until 24-hr postdose for all treatments administered. The continuous 12-lead Holter electrogram was recorded after at least 20 minutes in the supine or semi-supine position. The analysis of the electrocardiogram data included an evaluation of QT prolongation. Data from this

study were also used in analysis that evaluated QTc prolongation risk for a single oral dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more (See Section 14.2.9).

14.2.4. Human Relative Bioavailability of Oral Zoliflodacin Formulation

Study DMID 16-0118

Study Design

The study was a phase 1, open-label, single-site, nonrandomized study in healthy participants. The study's primary objective was to assess the PK parameters of zoliflodacin granules for oral suspension, ZoliPa GFOS, following a single 4 g dose under fasted conditions. The secondary objectives included to assess the safety and tolerability. For fasted conditions, participants fasted overnight for at least 8 hours prior to treatment administration.

Drug Product Formulations

ZoliPa GFOS described in the preceding section was evaluated.

PK Sampling

Blood samples for zoliflodacin plasma concentration were collected for each participant predose and at 0.5, 1, 2, 3, 4, 6, 8, 12, 24, 36, 48, and 72 hours postdose.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method TP70474 with associated validation report VDPVC1701P1 (see Section 14.3).

Results

In total, eight participants were enrolled (including 4 (50%) females) with a mean (range) age of 26 (22 to 31) years and the mean ± standard deviation weight of 74.71±11.09 kg. All participants received at least one dose of study medication and completed the study. Summaries of the descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral dose administered under fasted conditions are presented in Table 102.

Table 102. Summary Statistics of Plasma Zoliflodacin Pharmacokinetic Parameters

Parameter (Unit)	Geometric Mean (%GCV), n ZoliPa GFOS (4 g Fasted) N=8
C_{max} (ng/mL)	19857 (34), n=8
T_{max} (h)^	4 (2 to 12), n=8
$t_{1/2}$ (h)	6.5 (10), n=8
$AUC_{0-\infty}$ (h*ng/mL)	213505 (38), n=8
CL/F (L/h)	18.7 (38), n=8
Vz/F (L)	174.4 (37), n=8

Source: Table 18 DMID 16-0118 CSR (page 74)
^Median (minimum to maximum)
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CL/F, apparent clearance; C_{max}: maximum concentration; GCV, geometric coefficient of variation; N, The number of participants included in the pharmacokinetic population for each treatment; n, the number of participants with a specific parameter; $t_{1/2}$, half-life; T_{max}:,time time to maximum plasma concentration; Vz/F, apparent volume of distribution; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Assessment of Study DMID 16-0118

The Applicant's overall findings from this study are consistent with the reviewer's analysis.

Study DMID 16-0118 collected standard 12-lead electrogram and a 10-second rhythm strip predose (at screening and within 1 hour prior to dosing) and at 1, 2, and 4 hours postdose on Day 1 and on Day 4. The electrogram was recorded after at least 5 minutes in the supine position. The analysis of the electrocardiogram data included an evaluation of QT prolongation. Data from this study were used in analysis that evaluated QTc prolongation risk for a single oral dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more (See Section 14.2.9).

14.2.5. Single Ascending Dose Studies and Dose Proportionality

Study D4930C00001

Study Design

The study was a phase 1, first-in-human, randomized, placebo-controlled, single-site, sequential Cohort study conducted in healthy participants in two parts. The first part, Part A, evaluated the study's primary objective, to assess the safety and tolerability of zoliflodacin powder for oral suspension, ZoliAZ PFOS, in healthy adult male and female participants. Part A evaluated a single ascending dose (SAD), ranging from 200 mg to 4000 mg, under fasted conditions in Cohort 1. A secondary objective included to assess the SAD PK parameters in healthy participants under fasted conditions. For fasted conditions, participants fasted overnight for at least 10 hours prior to treatment administration. Cohort 1 participants at each dose level were randomized in a 6:2 ratio to receive ZoliAZ PFOS or placebo.

The second part, Part B, evaluated a secondary objective of evaluating the FE on zoliflodacin pharmacokinetics in healthy participants in Cohort 2. Cohort 2 had two arms evaluating two different doses, and a two-way crossover design within each arm. Participants were randomized to receive a single dose of zoliflodacin (ZoliAZ PFOS) under either fed or fasted conditions, with a 4-day washout between periods. For fed conditions, participants were administered a high-fat

breakfast, and were expected to consume the entire meal, 30 minutes prior to treatment administration. For fasted conditions, participants fasted overnight for at least 10 hours prior to treatment administration.

Cohort 2 participants were randomized to the following four treatments as single dose:

Treatment A: 1500 mg ZoliAZ PFOS fasted

Treatment B: 1500 mg ZoliAZ PFOS fed

Treatment C: 3000 mg ZoliAZ PFOS fasted

Treatment D: 3000 mg ZoliAZ PFOS fed

FE was assessed by a comparison of the GMR and the associated 90% CI of the observed zoliflodacin PK parameters, C_{max}, and $AUC_{0-\infty}$. For FE the reference treatment is respective fasted condition treatment with the same dose.

Drug Product Formulations

All study parts evaluated ZoliAZ PFOS formulation described in the preceding section.

PK Sampling

Blood samples for zoliflodacin plasma concentration were collected for each participant predose and at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 12, 24, 36, 48, and 72 hours postdose. Urine samples for zoliflodacin urine concentration were collected for each participant at collection intervals from −12 to 0 hours (predose), and 0 to 4, 4 to 8, 8 to 12, 12 to 24, 24 to 48, and 48 to 72 hours postdose.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method 914HPP with associated validation reports 8288182 (see Section 14.3). Urine samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method 914HUP with associated validation reports 8288183 (see Section 14.3).

Results

Cohort 1 (Part A) enrolled 48 participants (including 2 (4.2%) females) with a mean (range) age of 31 (19 to 55) years and the mean ± standard deviation weight of 77.4±10.8 kg. Cohort 2 (Part B) enrolled 18 participants (including 0 (0%) females) with a mean (range) age of 30 (22 to 48) years and the mean ± standard deviation weight of 77.4±11.2 kg. In total, 66 participants (48 in Cohort 1 and 18 in Cohort 2) were randomized and 65 participants (47 in Cohort 1 and 18 in Cohort 2) completed the study. One participant in Cohort 1, 800-mg dose level, was lost to follow-up.

In Cohort 1, 36 participants received a single dose of zoliflodacin (n=6 for each dose level) and 12 received a single placebo-dose. In Cohort 2, 18 participants (n=8 for 1500 mg dose level, and n=10 for 3000 mg dose level) received a single dose of zoliflodacin.

Summaries of the descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral dose administered under either fasted or fed conditions are presented in Table 103 and Table 104.

Cohort 1 SAD PK exposure parameters generally increased with increasing dose in the 200 to 4000 mg dose range studied. Exposure parameters generally demonstrated dose proportionality between 200 to 800 mg, and less than dose proportionality above 800 mg to the highest dose range studied, 4000 mg. Urinary excretions were <5.0% of the total dose delivered, with a decreasing fraction extracted (fe) observed as dose increased.

Zoliflodacin was found to have a significant FE with respect to $AUC_{0-\infty}$, as shown in Table 105 below. The GMRs (fed/fasted) for $AUC_{0-\infty}$ were approximately 1.2 and 1.4 when administered with a high-fat breakfast for 1500 mg and 3000 mg doses, respectively. The 90% CIs of the GMRs for $AUC_{0-\infty}$ did not contain 100%, indicating a statistically significant FE. The GMRs (fed/fasted) for C_{max} when administered with a high-fat breakfast were 0.8 and 1.0 of the C_{max} when fasted, which is not clinically significant for this formulation. As shown in Table 104, food also delayed absorption by approximately 1.5 hrs at both doses evaluated.

Table 103. Summary Statistics of Zoliflodacin (ZoliAZ PFOS Fasted) Pharmacokinetic Parameters by Treatment (Part A SAD)

	Geometric Mean (Geometric CV%)					
Parameter (Unit)	**200 mg N=6**	**400 mg N=6**	**800 mg N=6**	**1600 mg N=6**	**3000 mg N=6**	**4000 mg N=6**
Plasma						
C_{max} (ng/mL)	2010 (12.1)	4870 (14.4)	10000 (18.7)	13400 (36.7)	20200 (19.0)	26100 (46.4)
t_{max} (h)^	2.26 (1.03 to 3.02)	2.00 (1.50 to 2.50)	2.00 (1.02 to 3.00)	1.50 (1.02 to 6.02)	2.25 (1.02 to 4.02)	2.04 (1.50 to 4.02)
$t_{1/2}$ (h)	5.58 (11.8)	5.22 (16.6)	6.25 (15.3)	5.81 (18.9)	6.11 (7.0)	5.99 (15.7)
$AUC_{0-\infty}$ (h*ng/mL)	14600 (10.5)	31300 (13.9)	68300 (22.5)	108000 (25.3)	183000 (17.5)	209000 (44.7)
CL/F (L/h)	13.7 (10.6)	12.8 (13.8)	11.7 (22.5)	14.8 (25.3)	16.4 (17.4)	19.1 (44.8)
Vz/F (L)	110 (12.2)	96.5 (28.3)	106 (14.5)	124 (37.7)	145 (21.9)	165 (34.6)
Urine						
Cumulative Ae (mg)	9.11 (15.1)	16.5 (28.9)	32.4 (34.2)	46.4 (24.5)	106 (23.7)	104 (44.1)
Cumulative fe (%)	4.56 (15.2)	4.12 (28.9)	4.05 (34.0)	2.90 (24.5)	3.54 (23.8)	2.59 (44.3)
CLr (L/h)	0.625 (18.0)	0.527 (25.1)	0.474 (47.1)	0.430 (30.8)	0.582 (12.8)	0.496 (11.1)

Source: Table 11.2.3 D4930C00001CSR (pages 108 to 113) and Table 11.2.9 D4930C00001CSR (pages 138 to 140)
^Median (minimum to maximum).
Abbreviations: Ae, amount excreted; $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CL/F, apparent clearance; CLr, renal clearance; C_{max}: maximum plasma concentration; fe, fraction excreted; N, number of participants in treatment arm; $t_{1/2}$, half-life; t_{max}, time to maximum plasma concentration; SD, standard deviation; Vz/F, apparent volume of distribution; ZoliAZ PFO,: zoliflodacin powder for oral suspension manufactured by Astra Zeneca

Table 104. Summary Statistics of Zoliflodacin Pharmacokinetic Parameters by Treatment (Part B FE)

	Geometric Mean (Geometric CV%)			
Parameter (Unit)	**1500 mg ZoliAZ PFOS (Fasted) N=8**	**1500 mg ZoliAZ PFOS (Fed) N=8**	**3000 mg ZoliAZ PFOS (Fasted) N=10**	**3000 mg ZoliAZ PFOS (Fed) N=10**
Plasma				
C_{max} (ng/mL)	15100 (18.2)	12100 (14.8)	22900 (31.6)	24000 (27.2)
t_{max} (h)^	2.50 (2.00 to 4.00)	4.00 (4.00 to 4.00)	2.50 (1.50-4.00)	4.00 (1.50 to 8.02)
$t_{1/2}$ (h)	5.41 (20.3)	5.53 (18.3)	6.14 (5.9)	6.17 (6.6)
$AUC_{0-\infty}$ (h*ng/mL)	109000 (19.1)	129000 (21.5)	195000 (36.4)	281000 (22.9)
CL/F (L/h)	13.8 (19.1)	11.6 (21.5)	15.4 (36.4)	10.7 (23.1)
Vz/F (L)	107 (15.3)	92.5 (16.0)	136 (34.3)	94.8 (23.3)
Urine				
Cumulative Ae (mg)	52.7 (48.5)	76.5 (36.3)	107 (37.1)	157 (21.1)
Cumulative fe (%)	3.51 (48.5)	5.10 (36.2)	3.56 (37.0)	5.24 (21.1)
CLr (L/h)	0.484 (36.9)	0.592 (24.4)	0.548 (27.2)	0.559 (23.1)

Source: Table 11.2.4 D4930C00001CSR (pages 114 to 117) and Table 11.2.10 D4930C00001CSR (pages 141 to 142)
^Median (minimum to maximum).
Abbreviations: Ae, amount excreted; $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CL/F, apparent clearance; CLr, renal clearance; C_{max}, maximum plasma concentration; fe, fraction excreted; N, The number of participants included in the pharmacokinetic population for each treatment and the number of participants with a specific parameter; $t_{1/2}$, half-life; t_{max}, time to maximum plasma concentration; SD, standard deviation; Vz/F, apparent volume of distribution; ZoliAZ PFOS, zoliflodacin powder for oral suspension manufactured by Astra Zeneca

Table 105. Statistical Analysis To Assess the Effect of Food on Plasma Pharmacokinetic Parameters of Zoliflodacin (Part B FE)

		Test:	Reference:	Ratio (%) (Fed/Fasted)		
Formulation and Dose	**Parameter (Unit)**	**Fed GM, n**	**Fasted GM, n**	**GMR (90% CI)**	**Intra CV%**	**p-Value**
ZoliAZ PFOS 1.5 g	C_{max} (ng/mL)	12080 n=8	15080 n=8	80.11 (70.62, 90.88)	NR	NR
	$AUC_{0-\infty}$ (h*ng/mL)	129300 n=8	108900 n=8	118.78 (111.43, 126.62)	NR	NR
ZoliAZ PFOS 3 g	C_{max} (ng/mL)	23960 n=10	22870 n=10	104.74 (86.30, 127.12)	NR	NR
	$AUC_{0-\infty}$ (h*ng/mL)	281400 n=10	194700 n=10	144.57 (122.34, 170.85)	NR	NR

Source: Table 18 D4930C00001CSR (page 60)
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CI, Confidence Interval; C_{max}, maximum concentration; CV, Coefficient of Variation; GM, Geometric Mean; GMR, Geometric Mean Ratio; n, number of participants with a specific parameter for a treatment; NR, not reported; ZoliAZ PFOS, zoliflodacin powder for oral suspension manufactured by Astra Zeneca

Assessment of Study D4930C00001

The Applicant's overall findings from this study are consistent with the reviewer's analysis.

Study D4930C00001 Cohort 1 collected standard 12-lead electrogram (paper) predose (at screening and prior to dosing on Day 1), and 1, 2, 3, 4, 6, 12, and 24 hours postdose on Day 1, and on Days 2, 3, 4, and between Day 7 to 11 days, and collected standard 12-lead electrogram (digital) predose (prior to dosing on Day 1), and 1, 2, 3, 4, 6, 12, and 24 hours postdose on Day 1, and on Day 2. Study D4930C00001 Cohort 2, both Periods, collected standard 12-lead

electrogram (paper) predose (at screening and prior to dosing on Day 1), and 12 hours postdose on Day 1, and on Days 2, 3, 4, 5, 6, 7, 8, 9, and 7 to 10 days following last dose. The electrogram was recorded after at least 10 minutes in the supine position. The analysis of the electrocardiogram data included an evaluation of QT prolongation. Data from this study were used in analysis that evaluated QTc prolongation risk for a single oral dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more (See Section 14.2.9).

14.2.6. Human Mass Balance Study

Study D4930C00003

Study Design

Study D4930C00003 was a phase 1, open-label, nonrandomized, mass balance study. The study's objectives were to assess the absorption, metabolism, excretion, pharmacokinetics, safety, and tolerability of a single radiolabeled oral dose of ZoliAZ PFOS in healthy male participants, administered under fasted conditions. For fasted conditions, participants fasted overnight for at least 8 hours prior to treatment administration.

Drug Product Formulations

The study medication was ZoliAZ PFOS mixed with radiolabeled [^{14}C]-zoliflodacin drug substance, such that a single oral dose of 3 g zoliflodacin contains ~500 μCi.

PK Sampling

Blood, urine, and fecal samples were collected up to 240 hours postdose to measure total radioactivity (in whole blood, plasma, pooled urine, and pooled feces samples) as well to characterize zoliflodacin and its metabolic profiles (pooled plasma, urine, and feces samples).

Blood samples for zoliflodacin plasma concentration and total radioactivity in blood and plasma were collected for each participant predose and at 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, 24, 36, 48, 72, 96, 120, 144, and 168 hours postdose.

Blood samples for metabolite plasma concentration were collected for each participant predose and at 0.5, 2, 4, 6, 12, 24, 48, and 72 hours postdose.

Urine samples for zoliflodacin urine concentration and total radioactivity in urine were collected for each participant at collection intervals from −12 to 0 hours (predose), and 0 to 6, 6 to 12, 12 to 24, 24 to 48, 48 to 72, 72 to 96, 96 to 120, 120 to 144, 144 to 168, 168 to 192, 192 to 216, 216 to 240 hours postdose.

Fecal samples for zoliflodacin fecal concentration and total radioactivity in feces were collected for each participant at collection intervals from −24 to 0 hours (predose), and 0 to 24, 24 to 48, 48 to 72, 72 to 96, 96 to 120, 120 to 144, 144 to 168, 168 to 192, 192 to 216, 216 to 240 hours postdose.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method 914HPP with associated validation reports 8288182 see Section 14.3). Urine samples were

assayed for zoliflodacin concentrations by LC-MS/MS using the method 914HUP with associated validation reports 8288183 (see Section 14.3).

Blood, plasma, urine, and fecal samples were assayed for total radioactivity by liquid scintillation counters.

Results

Six male participants were enrolled with a mean (range) age of 34 (21 to 53) years and weight of 83.6 (73.7 to 90.6) kg. In total, six participants received a single 3 g dose of zoliflodacin (~500 μCi), and all participants completed the study.

The following are summary findings. Percentages described below refer to percent of dose:

Mean radioactive recoveries in total excreta (urine and feces) was estimated to be 98% over 192 hours, with 18% and 80% of the total recovery in urine and feces, respectively.

The blood to plasma total radioactivity ratio ranged from 0.659 to 0.712.

From the circulating total radioactivity in plasma, zoliflodacin was the most abundant circulating component (72.3%) and all metabolites accounted for ≤21% (M3 (16.4%), M7(1.39%), M2(1.2%), M37(1.2%), and M4 (0.655%)).

Mean terminal $t_{1/2}$ for zoliflodacin in plasma, total radioactivity in plasma, and total radioactivity in whole blood was 6.454 hours, 7.511 hours, and 6.420 hours, respectively.

Parent/Metabolite profiling and identification in excreta (urine or feces):

Zoliflodacin accounted for <5% of radiolabeled components excreted (urine and feces).

Urinary excretion is a minor elimination route. Mean ± standard deviation radioactive recoveries in urine were 18.15±2.559% of the total recovery, with 2.5% as parent drug, with other minor metabolites (all <10% of the dose).

Fecal excretion is major elimination route. Mean ± standard deviation radioactive recoveries in feces were 79.60±2.440% of the total recovery, with 1.5% as parent drug, with other minor metabolites (all <10% of the dose).

Metabolism was localized predominantly to the morpholine ring. Primary metabolism of zoliflodacin was mediated by reduction and hydrolysis, to a lesser extent, oxidation and N-dealkylation. The most abundant metabolite is plasma and urine, M3, resulted from oxidation of the morpholine ring with cleavage of the tetrahydropyridine ring.

Assessment of Study D4930C00003

The Applicant's overall findings from this study are consistent with the reviewer's analysis.

Study D4930C00003 collected standard 12-lead electrogram, recorded in triplicate, predose (at screening and prior to dosing on Day 1), and postdose on Days 1 to 8, and at discharge. The electrogram was recorded after at least 10 minutes in the supine position. The analysis of the electrocardiogram data included an evaluation of QT prolongation. Data from this study were used in analysis that evaluated QTc prolongation risk for a single oral dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more (See Section 14.2.9).

14.2.7. Participants With Uncomplicated Urogenital Gonorrhea Pharmacokinetic-Pharmacodynamic Studies

Trial STI_Zoli001

Study Design

Trial STI_Zoli001 was the pivotal phase 3 noninferiority efficacy trial that evaluated and compared the efficacy and safety of zoliflodacin to dual therapy with ceftriaxone and azithromycin, for the treatment of uncomplicated urogenital gonorrhea. See Section 6.2.2 for more information on study design.

Drug Product Formulations

Trial STI_Zoli001 evaluated ZoliPa GFOS formulation described in the preceding section, and commercially available ceftriaxone and azithromycin.

PK Sampling

In a PK substudy, blood samples for zoliflodacin plasma concentration were collected for each participant predose and at the following intervals: 15 minutes to 1 hour, 2 to 2.5 hours, 4.5 to 5 hours, 10 to 12 hours, and 24 to 36 hours postdose.

Bioanalytical Method

Trial STI_Zoli001 plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method TP70542 with associated validation report V1241802P1 (see Section 14.3).

Results

Participant disposition, study demographics, and study results are described in Section 6.2.2.4. Zoliflodacin was noninferior to ceftriaxone-azithromycin (See Table 8) for the primary efficacy endpoint of microbiological cure at TOC at the urogenital site evaluated in the micro-ITT (urogenital) population. A subgroup analysis for the primary endpoint is presented in Section 6.2.2.4 Table 9. The microbiological cure at TOC by anatomical site and baseline *N. gonorrhoeae* zoliflodacin MIC (micro-ITT) is presented in Table 131.

With respect to the PK substudy, 24 participants (3.9% of zoliflodacin arm, age 20 to 50 years of age weighing between 48.2 and 112.3 kg) participated. All participants in the PK substudy received a single 3 g dose of zoliflodacin under fed conditions and had at least one PK sample collected and analyzed for zoliflodacin plasma concentration. One participant (Participant (b) (6) only had one PK blood sample drawn, at the first timepoint. All other participants had sampling per protocol for PK blood samples.

Summaries of the descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral dose administered under fed or fasted conditions are presented in Table 106.

Table 106. Summary Statistics of Plasma Zoliflodacin PK Parameters

Parameter (Unit)	Geometric Mean (%GCV) ZoliPa GFOS (3 g Fed) N=23
C_{max} (ng/mL)	31800 (27.3)
t_{max} (h)^	4.62 (2.03 to 11.3)
t_{last} (h)^	24.4 (24.0 to 26.9)
AUC_{0-last} (h*ng/mL)	330000 (26.5)
CL/F (L/h)	9090 (26.4)
Vz/F (L)	85.4 (24.5)

Source: Table 6 PK report B3231901P1-PK (page 19)
^Median (minimum to maximum).
Abbreviations: AUC_{0-last}, area under the time-concentration curve from zero to last timepoint collected; CL/F, apparent clearance; C_{max}, maximum concentration; GCV, geometric coefficient of variation; N, number of participants included in the PK population for each treatment; PK, pharmacokinetic; $t_{1/2}$, half-life; t_{last}, time to last PK sample collected with a quantifiable zoliflodacin plasma concentration; t_{max}, time to maximum plasma concentration; Vz/F, apparent volume of distribution; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

Assessment of Trial STI_Zoli001

The Applicant's overall findings from this PK substudy are consistent with the reviewer's analysis. See discussion in Section 6.1 regarding efficacy of the proposed dosage. Additional assessments regarding the proposed dosage with respect to efficacy and safety data from Trial STI_Zoli001 are discussed below.

Trial STI_Zoli001 Data Assessment Related to the Applicant's Proposal to Dose Patients Between >35 to <50 kg in the Fasted State To Mitigate Potential QT Prolongation Risk

Trial STI_Zoli001 participants weight range was 36.5 to 138.6 kg. All participants with a reported baseline weight of <50 kg (n=35) received the assigned study treatment under fed conditions (n=24 zoliflodacin arm, n=11 ceftriaxone-azithromycin arm). Per the proposed zoliflodacin dosing instructions, patients weighing <50 kg would have been dosed under fasted conditions rather than fed conditions. Only one of these participants (Participant (b) (6), weight of 48.2 kg) was enrolled in the PK substudy. This participant had an observed C_{max} of 42.7 µg/mL at 2.03 hours, which exceeds the TQT threshold of 42.1 µg/mL determined in the TQT study (See Section 14.2.9). None of the participants weighing below 50 kg reported any cardiac-associated adverse events.

Data Assessment of Need to Dose Patients Between >50 kg in the Fed State To Ensure an Efficacious Dose

For the phase 3 trial, participants with uncomplicated suspected or confirmed urogenital gonorrhea received a single 3 g dose of zoliflodacin under either fed conditions (n=555/621, 89%), fasted conditions (n=64/621, 10%), or unknown conditions with respect to food status (n=2/621, 0.3%). No clear trend was observed between the food status and the primary endpoint, microbiological cure at test of cure (Day 6±2 days) for this study.

The PTA analysis discussed in Section 14.5.1 below predicted >90% target attainment for a PK-PD target associated with efficacy for patients weighing <175 kg received a single 3 g dose of zoliflodacin under fasted conditions for MICs of ≤0.25 mg/L.

Extrapolation To Support Indicated Population (Age ≥12 Years and Weight ≥35 kg)

The phase 3 trial data and population PK analyses provide support for the indicated population age and weight cutoffs based on the following:

The pivotal phase 3 trial provided efficacy and safety data for the participants in the zoliflodacin treatment arm ranged from 16 to 73 years of age, and a weight range 36.5 to 138.6 kg.

The population PK analysis (see Section 14.5.1) found that body weight is the primary determinant for the PK differences between adult and adolescent patients.

Age (19 to 55 years old) was not a significant determinant of the pharmacokinetics of zoliflodacin once body weight was considered.

The PTA analysis (see Section 14.5.1) supports the extrapolation of use of zoliflodacin for treatment of urogenital gonorrhea down to an age of 12 years and a weight of 35 kg.

14.2.8. Participants With Uncomplicated Urogenital Gonorrhea Pharmacodynamic Studies

Trial DMID 14-0014

Trial Design

Trial DMID 14-0014 was a multicenter phase 2 randomized, open-label efficacy trial that evaluated the efficacy by microbiological cure rate of 2000 mg or 3000 mg zoliflodacin compared to 500 mg ceftriaxone for the treatment of uncomplicated urogenital gonorrhea. See Section 6.2.3 for information on trial design.

Drug Product Formulations

Trial DMID 14-0014 evaluated ZoliAZ PFOS formulation described in the preceding section, and commercially available ceftriaxone formulation.

PK Sampling

The trial did not include PK assessments.

Results

Participant disposition, study demographics, and study results are described in Section 6.2.3.4. The microbiological cure at TOC at urogenital Site evaluated in the micro-ITT (urogenital) population for zoliflodacin was >95% for all doses studied (See Table 17).

14.2.9. QT Assessment

Study DMID 16-0110

Study Design

Study DMID 16-0110, the TQT/QTc study, was a phase 1, single-site, randomized, placebo and positive-controlled, double-blinded (except for the use of positive control moxifloxacin), four-

period, four-treatment, crossover design in healthy male and female participants. The study's objectives were to assess the effect of zoliflodacin on cardiac repolarization, pharmacokinetics, safety, and tolerability of zoliflodacin granules for oral suspension, ZoliPa GFOS, in healthy adult male and female participants, following a single dose of zoliflodacin, under fasted conditions.

In the study's four-way crossover design, participants received the following four treatments with an 8-day minimum washout between periods:

Treatment A: 2 g ZoliPa GFOS, fasted

Treatment B: 4 g ZoliPa GFOS, fasted

Treatment C: Placebo to match zoliflodacin, fasted

Treatment D: 400 mg tablet of moxifloxacin hydrochloride, fasted

Participants were randomized using a 12-sequence design balanced for period and first-order carryover effects to examine sequence and period effect. For fasted conditions, participants fasted overnight for at least 10 hours prior to treatment administration.

The effect of zoliflodacin on cardiac repolarization, was assessed by the length of the electrocardiogram (ECG) onset of QRS complex to end of T-wave, or QTcF. The QTcF effect of zoliflodacin or moxifloxacin was studied following single doses of 2 g or 4 g zoliflodacin or 400 mg oral moxifloxacin, administered under fasted conditions. The primary endpoint was based on a comparison of the upper bound of the one-sided 95% CI of the estimated mean placebo-adjusted change of QTcF from Baseline (ΔΔQTcF) for each zoliflodacin dose group in the 24-hr period postdose, to a limit of <10 msec. Other secondary endpoints included determination of time-matched, placebo-corrected, baseline-adjusted non-QT intervals (PR, QRS, and RR intervals) and heart rate collected in the 24-hr period postdose, a comparison of the one-sided lower bound of the 95% CI of ΔΔQTcF for each zoliflodacin dose group in the time period between 1-hr and 4-hr postdose, to a limit of >5 msec, incidence of abnormal T-wave morphology using predefined categories.

Drug Product Formulations

The study evaluated ZoliPa GFOS formulation, placebo to match zoliflodacin granules for oral suspension, and commercially available moxifloxacin tablets.

PK Sampling

Blood samples for zoliflodacin plasma concentration assessment were collected for each participant predose and at 0.5, 1, 2, 3, 4, 6, 8, 12, and 24, hours postdose.

Bioanalytical Method

Plasma samples were assayed for zoliflodacin concentrations by LC-MS/MS using the method TP70474 with associated validation report VDPVC1701P1 (see Section 14.3).

Results

Seventy-two participants were enrolled (including 26 [36%] females) with a mean (range) age of 31.7 (19 to 44) years and the mean ± standard deviation weight of 74.53±12.37 kg. In total, 72

participants were randomized (6 per sequence) and all participants were included in the safety population. Nine participants terminated early or voluntarily withdrew for the following reasons: failure to meet eligibility criteria (two participants), lost to follow-up (three participants), voluntary withdrawal (one participant), noncompliance with protocol required procedures (two participants), and physician decision (one participant). Sixty-four participants completed all four periods of study (n=69 Treatment A, n=67 Treatment B, n=70 Treatment C, and n=71 Treatment D). One participant who did not receive zoliflodacin was excluded from ECG analysis population and PK-ECG analysis population. One additional participant was excluded from PK-ECG analysis population due to protocol deviation of not having at least one ECG and PK concentration at the same nominal time point.

Summaries of the descriptive statistics of selected plasma zoliflodacin PK parameters following a single oral 2 g or 4 g dose administered under fasted conditions are presented in Table 107.

Table 107. Summary Statistics of Zoliflodacin Pharmacokinetic Parameters by Treatment, Study DMID 16-0110

	Geometric Mean (%GCV)	
Parameter (Unit)	**2 g ZoliPa GFOS (Fasted) N=68**	**4 g ZoliPa GFOS (Fasted) N=66**
C_{max} (ng/mL)	11770 (25), n=68	19360 (31), n=66
t_{max} (h)^	3.02 (1.1-6.0), n=68	3.02 (1.1-12.0), n=66
$t_{1/2}$ (h)	5.49 (12), n=49	5.92 (12), n=41
$AUC_{0-\infty}$ (h*ng/mL)	103500 (30), n=49	174900 (29), n=41
CL/F (L/h)	19.32 (30), n=49	22.86 (29), n=41
Vz/F (L)	153.01 (32), n=49	195.23 (29), n=41

Source: Table 55 and 56 DMID 16-0110 CSR (page 394)
^Median (minimum to maximum).
Abbreviations: $AUC_{0-\infty}$, area under the time-concentration curve from zero to infinity; CL/F, apparent clearance; C_{max}, maximum concentration; GCV, geometric coefficient of variation; N, number of participants included in the pharmacokinetic population for each treatment; n, number of participants with a specific parameter; $t_{1/2}$, half-life; t_{max}, time to maximum plasma concentration; Vz/F, apparent volume of distribution; ZoliPa GFOS, zoliflodacin granules for oral suspension manufactured by Patheon

The primary analysis in this TQT study demonstrated that the estimated mean ΔΔQTcF at the geometric mean plasma concentrations for the zoliflodacin doses had CI upper bounds of <10 msec. The highest least square mean value of ΔΔQTcF (msec) was 1.42 (upper confidence limit of 3.044, timepoint Day 1 3 hr) and 3.04 (upper confidence limit of 4.674, timepoint Day 1, 4 hr) for the 2 g and 4 g treatment groups, respectively.

Assessment of Study DMID 16-0110

The Applicant's overall findings from this study are consistent with the reviewer's analysis. See associated June 26, 2025, memo from Interdisciplinary Review Team for Cardiac Safety Studies related to the review of zoliflodacin QT prolongation risk, and the conclusion that the totality of data adequate to exclude clinically significant QTc prolongation for a single dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more.

It was noted in the initial interdisciplinary review team (IRT) review of TQT study (July 9, 2020, memo from Interdisciplinary Review Team for Cardiac Safety Studies under IND (b) (4) that Applicant's ΔΔQTcF results are consistent with results from reviewer's analysis. Additionally, none of the events identified to be of clinical importance per the ICH E14 guidelines (i.e.,

seizure, significant ventricular arrhythmias or sudden cardiac death) occurred in this study.[34] No participants experienced QTcF >480 msec or ΔQTcF >60 msec after receiving Zoliflodacin 2 g or 4 g under fasted conditions.

The memorandum dated June 26, 2025 (DARRTS date June 27, 2025), from the Interdisciplinary Review Team for Cardiac Safety Studies noted disagreement with the Applicant's proposal to use the TQT study data to derive a threshold concentration for QTc prolongation of 42 µg/mL. The memorandum noted considerable uncertainty in the proposed threshold concentration of 42 µg/mL due to limited concentration-QTc prolongation data above 30 µg/mL and that the totality of data was adequate to exclude clinically significant QTc prolongation for a single dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg. Therefore, an information request (IR) was sent to the Applicant requesting justification of the proposal of administration under fasted conditions for patients weighing ≥35 kg to <50 kg. In their subsequent response, the Applicant acknowledged that the theoretical safety threshold of 42.1 µg/mL is very likely conservative and noted the lack of any observed exposure-related safety signal. Nevertheless, the Applicant retained reliance on a theoretical TQT signal for dose justification purposes. The review team did not pursue further analysis given the Applicant's proposal for relying on a conservative approach.

As noted in the June 26, 2025, IRT Consult under this NDA, the mean exposure estimated for 3 g zoliflodacin administered under fed conditions, observed in FE studies STI_Zoli002 and STI_Zoli003, exceeds that observed in this study for healthy volunteers administered 4 g zoliflodacin under fasted conditions. The Applicant conducted an additional assessment of QT prolongation in healthy volunteers administered either 3 g or 4 g under both fed and fasted conditions in Study STI_Zoli002. No clinically significant QTc prolongation was observed following administration of 4 g zoliflodacin administered under fed conditions. Study STI_Zoli002 was not placebo-controlled or positive-controlled with respect to QTc prolongation evaluation.

The drug development program collected electrocardiogram data from all clinical studies except for the phase 2 and 3 efficacy trials. In these studies, no QTc change from Baseline >60 msec or QTc >500 msec was observed in any participants, even at 4 g doses administered under fed condition.

Finally, no clinically concerning cardiovascular adverse events were reported in the completed phase 3 trial for zoliflodacin.

Based on the totality of evidence, the review team concurs with the IRT Consult conclusion that the totality of data is adequate to exclude clinically significant QTc prolongation for a single dose of 3 g of zoliflodacin administered with or without food to patients weighing 35 kg or more.

[34] ICH guidance for industry E14 Clinical Evaluation of QT/QTc Interval Prolongation and Proarrhythmic Potential for Non-Antiarrhythmic Drugs (October 2005), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/e14-clinical-evaluation-qtqtc-interval-prolongation-and-proarrhythmic-potential-non-antiarrhythmic-0.

14.3. Bioanalytical Method Validation and Performance

Clinical studies utilized validated high-performance liquid chromatography with tandem mass spectrometry bioanalytical methods for quantification of zoliflodacin in plasma and urine PK samples. In general, the bioanalytical methods' validation and performance, as summarized in Table 108, met the criteria recommended in the ICH Technical Requirements for Pharmaceuticals for Human Use M10 Bioanalytical Method Validation and Study Sample Analysis Guidance for Industry.[35]

Specificity, selectivity, matrix effect for plasma and urine samples, hemolytic and lipemic effects for plasma samples, carryover, precision and accuracy at the low-, medium-, and high-quality control concentrations evaluated, dilution linearity, and extraction efficiency (recovery) were found to be acceptable for all methods submitted. The validated calibration curves and associated dilution factors for each method were sufficient to cover the reported concentration ranges for the bioanalytical samples from the clinical studies. Overall, the methods were adequately validated for the intended purpose.

The concentrations reported in the bioanalytical performance reports were precisely and accurately measured with samples stored and processed in the time frame supported by stability data (see Table 108). Incurred sample re-analyses were conducted, except for Study D4930C00003 BA urine samples, and findings are acceptable. The bioanalytical performance reports included chromatograms, except for report B1241804P1 (see below). The findings from bioanalysis reports review are summarized below.

Bioanalysis Report Assessment

Bioanalysis report 8312578, associated with Study D4930C00003, conducted incurred sample reanalysis for the plasma samples, but not for the urine samples.

Bioanalysis report B1241804P1, associated with Study STI_Zoli002, did not contain chromatograms, however, representative chromatograms of the method were submitted for the other bioanalyses using this method and in the method validation report. All performance measures reported in report B1241804P1 indicate that the method met other requirements, therefore there is no impact of this finding on the study results.

Bioanalysis report 8290499 associated with Study D4930C00001, Run 17, from January 30, 2014, had assay values of 2.93 and 1120 ng/mL for the two injections of the lowest quality control (QC) (3.00 ng/mL Dilution =1). Although a value of 1120 ng/mL is notably an outlier for this QC check, the report did not indicate that an investigation was conducted, and the value was not omitted from descriptive statistics, resulting in an interday accuracy of 666% relative error and interday precision of 647.8% coefficient of variance for n=56 injections. Omitting this outlier, values for this QC level across all 40 runs ranged from 2.42 ng/mL to 3.85 ng/mL, with interday accuracy of 100.9% relative error and interday precision of 7% coefficient of variance for n=55 injections.

[35] See Footnote 17

Bioanalysis report 8290499, associated with Study D4930C00001, had an interday precision of ≤23% CV for the lowest QC level (15 ng/mL). All reported assay values for the lowest QC level (range 7.02 to 8.46) for Run 39 from March 10, 2014 had >15% bias. A single reported assay value for the lowest QC level from both Run 29 from February 14, 2014 (17.4) and Run 43 from March 18, 2014 (18.5) also had >15% bias. The report did not indicate that an investigation was conducted for any of these assayed results, and the values were not omitted from descriptive statistics. Interday precision was <15% CV for all other QC levels in Bioanalysis report 8290499. These findings have no impact on the interpretation of study results.

Table 108. Summary of Bioanalytical Method Validation and Performance for Zoliflodacin

	Method Details Per Trial/Study			
Method Parameters	**Trials STI_Zoli001, STI_Zoli002, and STI_Zoli003**	**Studies NIAID 16-0110 and NIAID 16-0118**	**Study D4930C00001**	**Study D4930C00003**
Drug	Zoliflodacin (also referred to as ETX0914, AZD0914, or CZOF11)			
Biological matrix (anticoagulant)	Plasma (K2EDTA)			Urine (N/A)
Extraction methods	Protein precipitation extraction	Liquid-liquid extraction	Liquid-liquid extraction	Liquid-liquid extraction
Internal standard[c]	ETX0914-13C2 15N2		AZ13420914-010 [13C2 15N2]	
Validation range (ng/mL)	10.0 to 30,000	1.00 to 5,000	1.00 to 5,000	5.00 to 10,000
QC levels (ng/mL)	30.0, 12000, and 24,000	3.00, 300, 1500, and 4,000	3.00, 110, and 4,000	15.0, 350, and 8,000
Dilution QC (ng/mL) [dilution factor]	120,000 [5-fold]	4,000 [2-fold], 20,000 [5-fold], 20,000 [25-fold], and 40,000 [10-fold]	50,000 [100-fold]	100,000 [100-fold]
Interday accuracy (RE%)[a]	−6.7 to 7.5	−3.5 to 6.7	−5.5 to 666% (See notes above related to an outlier)	−8.4 to 10.7%
Interday precision (CV%)[a]	<12.9%	<11.7%	<647.8% (See notes above related to an outlier)	<23.6% (See notes above related outliers)
Incurred sample reanalysis[b]	>94.2%	>99.3%	100%	87%
Long term matrix stability at −70°C	630 days	318 days	407 days	405 days
Long term matrix stability at −20°C	622 days	431 days	407 days	405 days
Validation report(s)	V1241802P1	VDPVC1701P1 and addendums	8288182 and addendum	8288183 and addendum
Bioanalysis report(s)	B3231901P1, B1241804P1, B3231903P1, and B3231903P2	BDPVC1701P2 and BDPVC1701P1	8290499 and 8312578	

Source: Reviewer's analysis
[a] The interday accuracy and precision values from validation and performance reports are combined and presented as an overall range.
[b] At least 10% of samples reanalyzed.
[c] Zoliflodacin is also known as ETX0914 and AZ13420914.
Abbreviations: CV%, coefficient of variation expressed as percent; K2EDTA, dipotassium ethylenediaminetetraacetic acid; N/A, not applicable; no., number; QC, quality control; RE%, relative error expressed as percent.

14.4. Immunogenicity Assessment—Impact of PK/PD, Efficacy, and Safety

No immunogenicity assessments were conducted for zoliflodacin.

14.5. Pharmacometrics Assessment

14.5.1. Population PK Analysis

Review Summary

The Applicant conducted 1) a PopPK analysis of zoliflodacin based on the PK data from healthy participants in six phase 1 studies and participants with uncomplicated gonorrhea in a phase 3 trial, and 2) PK-PD target attainment analyses using the final PopPK model, nonclinical PK-PD targets for efficacy, the in vitro surveillance data, and the MIC data from the phase 3 trial to support the proposed dosing regimen and the criteria for the in vitro susceptibility testing for zoliflodacin against *N. gonorrhoeae*. The proposed dosing regimen is a single oral administration of zoliflodacin 3 g, which should be taken with a meal for patients weighing ≥50 kg and in a fasted state for patients weighing 35 kg to <50 kg. In the pivotal phase 3 trial, participants were instructed to take 3 g in a fed state regardless of body weight. The pharmacometrics review was focused on whether the submitted analyses support the Applicant-proposed dosing regimen.

Table 109. Anticipated Model Risk for PopPK Analysis

Elements	Details
Question of interest	What is the appropriate zoliflodacin dosage for treatment of uncomplicated *Neisseria gonorrhoeae* in adult and pediatric patients 12 years and older?
Context of use	PopPK analysis to predict the PK profiles in adolescent and adult patients and to conduct the PTA analysis to support the proposed dosage
Model influence	Medium • In the pivotal phase 3 trial, most participants with gonorrhea received a single oral dose of 3 g zoliflodacin in a fed state. The efficacy, safety, and PK data were available for this dosing regimen. The food effect on zoliflodacin pharmacokinetics was characterized in dedicated food effect studies. • The proposed dose (3 g in a fasted state) for patients weighing 35 kg to <50 kg was not studied in the phase 3 trial. There were no PK data in adolescent patients and limited PK data in participants weighing <50 kg. Therefore, the PK prediction for assessment of QT prolongation risk, and PTA analysis in this participant group is relied on the PopPK analysis and simulation
Decision consequence	Medium to high • If the decision on the dosage based on the totality of evidence is wrong, patients may be potentially associated with increased risk of QT prolongation.
Model risk	Medium/high

Source: Reviewer's assessment
Abbreviations: PK, pharmacokinetic; PopPK, population PK, PTA, probability of target attainment

Applicant's PopPK analysis adequately described the observed PK data following administration of zoliflodacin in healthy participants and adult participants with uncomplicated gonorrhea

weighing ≥50 kg. The PopPK analysis reasonably captured the food effect on zoliflodacin pharmacokinetics. While the predictive performance of the PopPK model is acceptable, to simulate PK profiles in unstudied scenarios such as adolescent patients, and participants weighing <50 kg with varying food conditions (fasted or fed state), the following assumptions were made: 1) there is no age effect on zoliflodacin pharmacokinetics and a body size (e.g., body weight) is the primary determinant for the PK difference between adult and adolescent patients, 2) the extent of food effect is similar between adult and adolescent patients, and 3) there are no significant differences in the PK-PD relationship for QT prolongation risk and antibacterial activity between adult and adolescent patients.

The simulations show that the estimated PK exposures in patients weighing 35 kg to <50 kg who take 3 g in a fasted state are comparable to those weighing ≥50 kg who take 3 g in a fed state. Based on and predicted PK exposures and the concentration-QTc interval relationship, the proposed dose for the weight group of 35 kg to <50 kg (3 g in a fasted state) is not expected to result in additional QT prolongation risk compared to the dosing regimen studied in the phase 3 trial (3 g in a fed state). The PK-PD target attainment analyses suggest that the probability of the target (free-drug $AUC_{0-\infty}$: MIC ratio of 70.6) attainment is >90% across the body weight group (35 kg to <50 kg and ≥50 kg) for the MIC ≤0.25 µg/mL, when patients receive the proposed dosing instruction. Overall, the proposed dosing regimen is supported by the PopPK analysis, the PTA analysis, and the efficacy and safety data from the phase 3 trial.

Data

The PopPK dataset includes zoliflodacin plasma concentration-time data collected from healthy participants enrolled in six phase 1 studies and participants with uncomplicated gonorrhea enrolled in a phase 3 trial. The dataset included a total of 261 participants and 6043 concentration records. The phase 1 studies included a range of doses (200 mg to 4 g) and investigated the impact of formulation type, manufacturer, predose meal type, and a concomitant CYP3A4 inhibitor on zoliflodacin PK. The data from a subset (n=24) of adult participants with uncomplicated gonorrhea from the phase 3 trial were included.

Table 110. Description of Studies Included in the PopPK Analysis

Study Title	Zoliflodacin Dose	Formulation (Manufacturer)
NCT01929629 (D4930C00001) Phase 1, Randomized, Placebo-Controlled, Single-Center Study To Assess the Safety, Tolerability, And PK of Zoliflodacin as a Powder for Oral Suspension After a Single Oral Administration and To Assess the Effect of Food in Healthy Adult Volunteers	Single oral administration Part A: Single ascending 200 mg to 4 g or Placebo Part B: Food effect 1.5 g and 3 g	(b) (4) powder (AstraZeneca)
NCT02298920 (D4930C00003) Phase 1, Open-Label Study To Assess ADME, Safety and Tolerability of a Single Oral Dose of Radiolabeled [^{14}C]-Zoliflodacin as a Powder for Oral Suspension in Healthy Male Subjects	Single oral dose of radiolabeled [^{14}C]-zoliflodacin, 3 g	(b) (4) powder (AstraZeneca)
NCT03404167 (DMID 16-0118) Phase 1 Clinical Trial To Evaluate the Plasma PK, Safety, and Tolerability of a Single Oral Dose of Zoliflodacin in Healthy Male and Female Subjects	Single oral dose of zoliflodacin 4 g	Granule for suspension (Patheon Pharma Services)

Study Title	Zoliflodacin Dose	Formulation (Manufacturer)
NCT03613649 (DMID 16-0110) Randomized, Double-Blinded, Phase 1, Four-Period Crossover, Thorough QT/QTc (TQT) Clinical Trial To Evaluate the Effect of Zoliflodacin as Granules for Oral Suspension, on Cardiac Repolarization in Healthy Male and Female Subjects	A: Zoliflodacin 2 g B: Zoliflodacin 4 g	Granule for suspension (Patheon Pharma Services)
NCT03718806 (STI_Zoli002) Phase 1, Open-Label Study To Investigate PK, Effect Of Food (FE) and Safety of A Granules for Oral Suspension Formulation of Zoliflodacin in Healthy Adult Subjects	Single oral dose of zoliflodacin 3 g or 4 g	Granule for suspension (Patheon Pharma Services)
NCT05635305 (STI_Zoli003) Phase 1, Single-Dose, Open-Label, Randomized, 4-Way Crossover Zoliflodacin BE Study of the Reference Product (Zolipa) With the Test Product (Zolidr) in Healthy Adult Volunteers Under Fasted and [Specific] Fed Conditions Paired With an Investigation of the Effect of CYP450 3A4 Inhibition by Itraconazole On 3 g Zoliflodacin (Zolipa) Single-Dose PK	Single oral dose of zoliflodacin 3 g	Granule for suspension (Patheon Pharma Services or Dr Reddy's)
NCT03959527 (STI_Zoli001) Multicenter, Randomized, Open-Label, Noninferiority Phase 3 Trial To Evaluate the Efficacy and Safety of Single Oral Dose of Zoliflodacin (Granules for Oral Suspension) Compared to the Combination of a Single IM Dose of Ceftriaxone and a Single Oral Dose of Azithromycin in the Treatment of Patients With Uncomplicated Gonorrhea	Zoliflodacin 3 g versus Azithromycin 1 g plus a single IM dose of ceftriaxone 500 mg	Granule for suspension (Patheon Pharma Services)

Source: Adapted from Applicant's PopPK analysis report, Table 1
Abbreviations: ADME, absorption, distribution, metabolism, and excretion; BE, bioequivalence; [^{14}C], carbon 14, CYP, cytochrome P 450; IM, intramuscular; PK, pharmacokinetic; PopPK, population PK; QTc, corrected QT interval; TQT, thorough QT; ZoliDr, zoliflodacin granules for oral suspension manufactured by Dr. Reddy's Laboratories, Ltd; ZoliPa, zoliflodacin granules for oral suspension manufactured by Patheon

Table 111. Summary Statistics of Participant Characteristics for PopPK Population

	NCT01929629	NCT02298920	NCT03613649	NCT03404167	NCT03718806	NCT05635305	NCT03959527	Pooled
Age (y)	31 (9.93) 27 (19-53)	34.2 (14.8) 28 (21-53)	31.7 (6.67) 31 (19-44)	26 (3.63) 26 (22-31)	39.7 (11) 40.5 (19-55)	40.5 (10.7) 40.5 (25-55)	28.6 (8.14) 27.5 (20-50)	34.3 (10.4) 32 (19-55)
Height (cm)	175 (6.73) 175 (160-190)	176 (4.56) 177 (170-181)	172 (9.83) 173 (148-192)	171(12.5) 172 (151-187)	169 (9.27) 169 (152-189)	176 (9.16) 177 (156-195)	170 (7.56) 170 (150-180)	173 (9.11) 173 (148-195)
Weight (kg)	76.5 (10.9) 76.5 (51.7-100)	83.5 (6.27) 85.2 (72.3-90.5)	74.7 (12.4) 72.5 (51.3-101)	74.7 (11.1) 70 (63-90.7)	73.9 (11.6) 74.8 49.2-97)	75.6 (11.3) 74.3 (51.5-93.8)	64.4 (14) 61.8 (48.2-112)	74.4 (12.2) 74 (48.2-112)
BSA (m^2)	1.92 (0.153) 1.90 (1.51-2.25)	2 (0.084) 2.01 (1.88-2.11)	1.87 (0.196) 1.88 (1.49-2.24)	1.87 (0.211) 1.81 (1.59-2.16)	1.84 (0.185) 1.86 (1.47-2.19)	1.91 (0.176) 1.89 (1.56-2.20)	1.74 (0.181) 1.72 (1.46-2.31)	1.87 (0.185) 1.88 (1.46-2.31)
BMI (kg/m^2)	24.9 (3) 25.5 (19.0-30.4)	27 (2.14) 27.4 (23.4-29.7)	25.1 (2.94) 25.7 (18.6-30)	25.4 (1.7) 25.7 (22.2-27.7)	25.8 (2.81) 26 (19.7-30.0)	24.5 (3.06) 24.5 (18.0-29.5)	22.3 (4.24) 21.4 (16.8-34.7)	24.9 (3.17) 25.4 (16.8-34.7)
CLcr (mL/min)	112 (23.6) 108 (71.9-176)	113 (30.3) 108 (80.9-150)	99 (18.2) 95.9 (71.9-156)	110 (14.1) 111 (89.2-127)	99.7 (18.3) 97.2 (66.8-137)	110 (22.2) 113 (69.6-165)	107 (16.5) 109 (78.0-134)	105 (20.9) 103 (66.8-176)
Sex								
Male	54/54 (100%)	6/6 (100%)	45/71 (63.4%)	4/8 (50%)	25/48 (52.1%)	36/50 (72%)	20/24 (83.3%)	190/261 (72.8%)
Female			26/71 (36.6%)	4/8 (50%)	23/48 (47.9%)	14/50 (28%)	4/24 (16.7%)	71/261 (27.2%)
Race								
White	28/54 (51.9%)	5/6 (83.3%)	33/71 (46.5%)	7/8 (87.5%)	41/48 (85.4%)	45/50 (90%)		159/261 (60.9%)
Black	23/54 (42.6%)		35/71 (49.3%)	1/8 (12.5%)	7/48 (14.6%)	2/50 (4%)	5/24 (20.8%)	73/261 (28%)
Asian	1/54 (1.85%)						19/24 (79.2%)	20/261 (7.66%)
American Indian/ Alaska Native	2/54 (3.7%)		3/71 (4.23%)					5/261 (1.92%)
Other		1/6 (16.7%)				3/50 (6%)		4/261 (1.53%)
Region								
US/EU	54/54 (100%)	6/6 (100%)	71/71 (100%)	8/8 (100%)	48/48 (100%)	50/50 (100%)		237/261 (90.8%)
Africa							5/24 (20.8%)	5/261 (1.92%)
Southeast Asia							19/24 (79.2%)	19/261 (7.28%)

Source: Adapted from Applicant's PopPK analysis report, Table 5, and Table 6
Abbreviations: BMI, body mass index, BSA, body surface area; CLcr, creatinine clearance; EU, European Union; PopPK, population pharmacokinetic; US, United States

Base Model

Gastrointestinal transit chain was described by three parameters: the transit compartments, the mean transit time (MTT) within the transit chain, and the rate of transfer through the transit compartments. The number of transit compartments was not constrained to be an integer allowing flexible fitting of complex absorption profiles. A first order k_a was also included to describe the movement from the transit chain to the central compartment. Relative bioavailability was parametrized as 'BIO'. Elimination from the central compartment was parameterized using a linear, first-order CL parameter. A proportional residual variability model was determined to be most appropriate.

As a correlation was identified between meal types and the interindividual variability (IIV) on MTT, categorical shifts were implemented for administration of zoliflodacin after a low-, medium-, or high-fat meal. As there was a correlation between formulation type (powder versus granule) and the IIV on BIO, the impact of administration of the granule formulation relative to the powder formulation were included in the model. The two granule formulations had similar relative bioavailability estimates.

Covariate Analysis

Covariate model development was carried out based on a forward selection followed by a backward elimination procedure. The covariates evaluated in the PopPK analysis included age, body weight, height, body surface area, body mass index, formulation type (granule for suspension versus powder), formulation manufacturer, food effect/meal composition (fasted

status, fed status with high-, medium-, or low-fat contents), CYP3A4 inhibitor (itraconazole), sex, race, and study phase (phase 1 versus phase 3).

Final Model

The final model contained an oral depot compartment with a relative bioavailability parameter, a transit chain compartment to describe the absorption profile, and one systemic compartment with linear elimination. Interindividual variability was estimated for k_a, CL, volume of central compartment (Vc), MTT, and BIO. Interoccasional variability was estimated for MTT, k_a, and BIO. Age and race were not identified as statistically significant covariates. The equations below describe the identified covariate-PK parameter relationships. The final PopPK parameter estimates are provided in Table 112 along with the resample statistics from a sampling-importance-resampling analysis.

Equation 1. Equations for Typical Pop-PK Parameter Values and Significant Covariates

$$Ka\ (1/h) = 1.82 \cdot (1-0.391)^{(Low\ or\ Medium\ Fat)} \cdot (1-0.610)^{(High\ Fat)} (1-0.479)^{PHASE}$$

$$CL\ (L/h) = 15.1 \cdot (1-0.246)^{Itraconazole} \cdot (1-0.139)^{Female} \cdot (WTKG/74)^{0.604}$$

$$V_C(L) = 124 \cdot (WTKG/74)^{0.811}$$

$$NN = 2.61$$

$$MTT\ (h) = 0.276 \cdot (1+1.63)^{(Low\ or\ Medium\ Fat)} \cdot (1+1.20)^{(High\ Fat)} (1+0.537)^{Patheon} \cdot (1+0.645)^{Dr.Reddy's}$$

$$BIO = 1 \cdot (1-0.236)^{(granules)} \cdot (1+0.412)^{PHASE} \cdot (1+0.496)^{(Low\ or\ Medium\ Fat)} \cdot (1+0.739)^{(High\ Fat)}$$

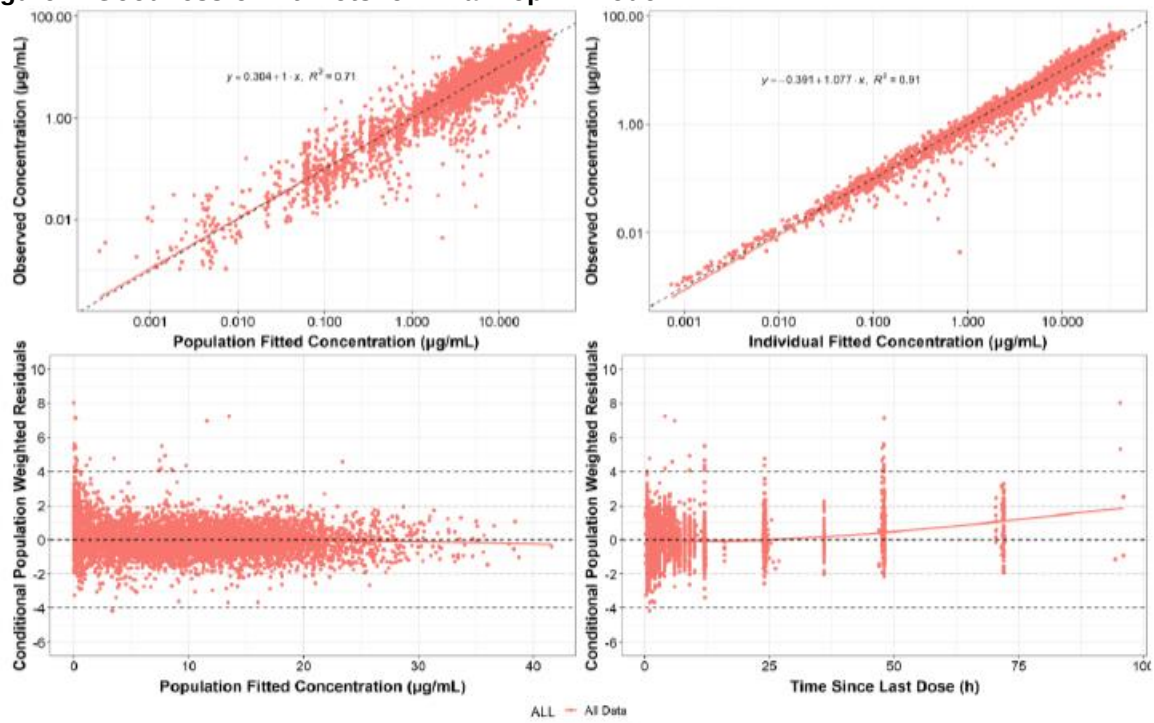
Source: Applicant's PopPK analysis report, Equations 11 to 16 on page 62.

Table 112. Parameter Estimates for Final PopPK Model

Parameter	Final Model			SIR statistics			
	Estimate[a]	%SEM	Shrinkage	Mean	Median	%CV	90% CI
Ka (1/h)	1.82	6.35		1.82	1.81	5.23	[1.66, 1.98]
Ka:Low/medium-fat meal shift	-0.391	21.9		-0.385	-0.386	17.9	[-0.498, -0.266]
Ka:High-fat meal shift	-0.610	6.94		-0.606	-0.606	5.99	[-0.664, -0.543]
Ka:Phase shift	-0.479	29.0		-0.473	-0.483	23.2	[-0.637, -0.271]
CL (L/h)	15.1	3.32		15.1	15.1	2.76	[14.5, 15.8]
CL:Itraconazole shift	-0.246	2.46		-0.247	-0.246	2.94	[-0.258, -0.235]
CL:Sex shift	-0.139	15.0		-0.137	-0.138	14.0	[-0.168, -0.103]
CL:WTKG coefficient	0.604	16.1		0.604	0.603	15.9	[0.449, 0.763]
Vc (L)	124	3.17		124	124	2.59	[119, 129]
Vc:WTKG coefficient	0.811	11.5		0.813	0.812	10.8	[0.676, 0.967]
NN	2.61	1.65		2.61	2.61	1.88	[2.53, 2.69]
MTT (h)	0.276	7.80		0.275	0.275	7.58	[0.241, 0.311]
MTT:Low/medium-fat meal shift	1.63	14.0		1.65	1.64	12.5	[1.32, 2.00]
MTT:High-fat meal shift	1.20	12.4		1.21	1.20	11.0	[1.00, 1.42]
MTT:Patheon shift	0.537	24		0.544	0.539	23.6	[0.341, 0.763]
MTT:Dr. Reddy's shift	0.645	36.2		0.650	0.647	31.6	[0.330, 0.990]
BIO[b]	1						
BIO:granules shift	-0.236	11.7		-0.235	-0.236	9.77	[-0.274, -0.196]
BIO:Phase shift	0.412	29.8		0.409	0.409	23.3	[0.254, 0.572]
BIO:Low/medium-fat meal shift	0.496	12.6		0.498	0.495	8.41	[0.433, 0.568]
BIO:High-fat meal shift	0.739	6.79		0.739	0.738	6.20	[0.668, 0.820]
ω^2_{Ka}	0.129 (35.9%)	35.7	47.9	0.132	0.132	20.7	[0.0885, 0.176]
ω^2_{CL}	0.0144 (12.0%)	18.8	21.5	0.0145	0.0145	16.0	[0.0108, 0.0184]
ω^2_{Vc}	0.00826 (9.09%)	31.2	39.0	0.00819	0.00799	27.9	[0.00461, 0.0122]
ω^2_{MTT}	0.0238 (15.4%)	109	68.7	0.0269	0.0250	68.4	[0.00341, 0.0561]
ω^2_{BIO}	0.0205 (14.3%)	24.8	34.2	0.0208	0.0204	23.0	[0.0136, 0.0293]
IOV_{MTT}[c]	0.265 (51.5%)	11.8	27.4	0.266	0.263	11.2	[0.220, 0.317]
IOV_{Ka}[d]	0.291 (54%)	14.7	37.6	0.293	0.287	15.6	[0.229, 0.380]
IOV_{BIO}[e]	0.0278 (16.7%)	10.2	27.9	0.0281	0.0279	10.2	[0.0237, 0.0329]
σ^2_{prop}	0.0573 (23.9%)	1.04	10.2	0.0573	0.0573	1.16	[0.0562, 0.0584]

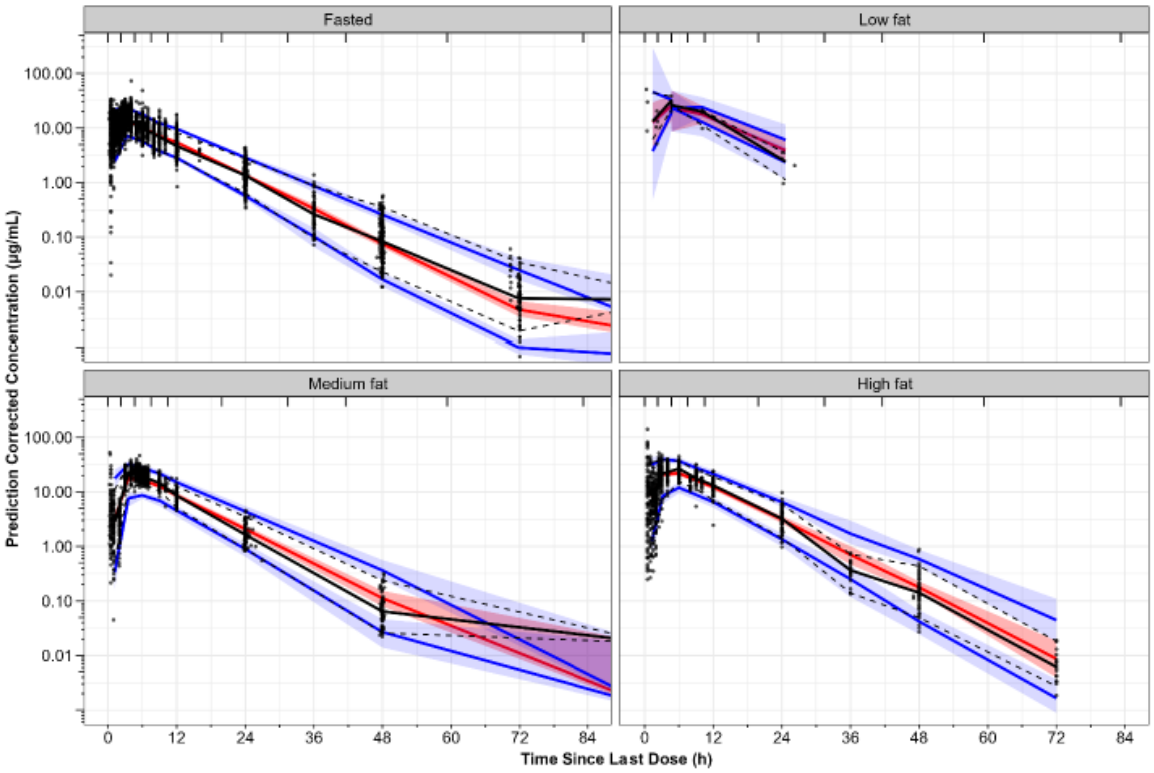
Source: Applicant's Population PK analysis report, Table 11
[a] Values presented in parentheses are %CV.
[b] The reference condition for bioavailability is the powder formulation in phase 1 participants administered zoliflodacin in the fasted state.
[c] MTT period 2, 3, and 4 shrinkages are 31.5, 67.3, and 63.5 respectively.
[d] Ka period 2, 3, and 4 shrinkages are 47.9, 75.2, and 73.4 respectively.
[e] BIO period 2, 3, and 4 shrinkages are 33.7, 70, and 72.6 respectively.
Abbreviations: BIO, bioavailability; CI, confidence interval; CL, clearance; CV, coefficient of variation; IOV, Interoccasional variability; Ka, absorption rate constant; MTT, mean transit time; popPK, population pharmacokinetic; NN, hypothetical number of transit compartments; SEM, standard error of mean; SIR, sampling-importance-resampling, Vc, volume of distribution in the central compartment; WTKG, body weight in kg; σ, standard deviation; ω, estimated variability

Figure 7. Goodness-of-Fit Plots for Final PopPK Model



Source: Applicant's Population PK analysis report, Figure 10
Abbreviation: popPK, population pharmacokinetic

Figure 8. Prediction-Corrected Visual Predictive Check Plot for Final Model Stratified by Meal Type



Note: Circles are observed concentrations, black solid lines are the median observed concentrations, black dashed lines are the 5th and 95th percentiles of the observed concentrations. Black pip marks at top of figure indicate the times since dose at which the data were binned for the calculation of summary statistics. Red and blue shaded regions are the 90% confidence intervals for the median, 5th, and 95th percentiles from the simulations.

Source: Applicant's IR response received on July 23, 2025

The PopPK model adequately describes the observed PK data following administration of single dose of zoliflodacin in healthy participants and participants with uncomplicated gonorrhea. The structural PK parameters (i.e., k_a, CL, Vc, and MTT) were estimated with good precision (%RSE <10%). The resampled parameter means are in line with those estimated in the final model fit. The interindividual variability parameters were estimated with acceptable precisions. The goodness of fit plots did not show obvious model misspecifications. The prediction corrected visual predictive check plots show that the final model captures the median tendency and the variability of observed time course of PK profiles. The shrinkage for IIV of clearance is low to moderate (21.5%). The PopPK analysis is acceptable to derive the post hoc PK exposure metrics for a given participant.

The covariate effect parameters were estimated with acceptable precisions. The interindividual variability for CL, Vc, MTT, and BIO were 12%, 9%, 15%, and 14%, respectively. The interindividual variability in k_a was more pronounced (36%). The goodness of fit plots and the prediction corrected visual predictive check plots stratified by the key covariates (i.e., formulation type, meal type, study, sex) show an agreement between the model predicted and the observed data in each subgroup. The key covariate effects are summarized below:

CL decreases with a decreasing body weight, female (versus male), and concomitant itraconazole administration.

Vc decreases with a decreasing body weight.

k_a decreases with food consumption with low-/medium-fat or high-fat and with the PK data from the phase 3 trial.

MTT increases with food consumption with low-/medium-fat or high-fat.

The relative bioavailability increases by 50% with a low- or medium-fat meal, and by 74% with a high-fat meal compared to the drug administration in a fasted condition. The estimated effect of a medium-fat meal is consistent with that observed in Study Zoli003. The estimated effect of a high-fat meal is lower than those observed in Study Zoli002.

The relative bioavailability in the phase 3 trial was estimated 41% higher than that in the phase 1 studies. No plausible explanation for this difference was identified.

Simulations

A virtual patient population resembling the characteristics of the 913 participants who were screened for Trial STI_Zoli001 was generated by resampling the participants from this study population with replacement. Virtual patients were assumed to receive the granule formulation for oral suspension. Zoliflodacin total-drug plasma concentration-time profiles were simulated for each virtual participant after a single oral administration of zoliflodacin 3 g. Each simulation was performed using the phase 1 bioavailability estimate and the phase 3 bioavailability estimate, sex (male or female), and food condition (in a fasted condition or following a meal with low to medium fat contents) across the body weight categories: ≥20 to <35 kg, ≥35 to <50 kg, ≥50 to <75 kg, ≥75 to <100 kg, ≥100 to <125 kg, ≥125 to <150 kg, ≥150 to <175 kg, and ≥175 to ≥200 kg.

The PopPK analysis showed that the PK exposures were higher for the participants enrolled in the phase 3 trial compared to those enrolled in phase 1 study or phase 2 trial, and the PK exposures in female participants were higher compared to male participants. Therefore, taking a conservative approach, when assessing the concentration-dependent QT prolongation risk, the C_{max} derived for female patients using the relative BA estimate for the phase 3 PK data, which is a high-exposure scenario, was primarily considered. Similarly, when assessing the probability of the PK-PD target attainment, the AUC derived for male patients using the relative bioavailability estimate for the PK data from phase 1 studies, which is a low-exposure scenario, was considered.

For female patients weighing 35 to <50 kg receiving 3 g in a fed state, the mean (5th and 95th percentiles) C_{max} was predicted to be 40.4 (28.5, 54.8) µg/mL, which is considerably higher than those predicted for patients weighing ≥50 kg who receive the same dose in a fed state. When female patients weighing 35 to <50 kg receive 3 g in a fasted state, the mean (5th and 95th percentiles) C_{max} was predicted to be 30.6 (21.8, 41.3) µg/mL, which is comparable to those

weighing ≥50 kg receiving the same dose in a fed condition. The proposed dosage (3 g in a fasted state) for patients weighing 35 to <50 kg is not likely lead to an additional QT prolongation risk compared to the clinical experience in the phase 3 trial.

Table 113. Simulated Total-Drug C_{max} Following 3 g in Fasted State (Assuming Phase 3 BA)

Variable	Summary statistics for zoliflodacin total-drug plasma C_{max} (µg/mL) by sex and body weight group (kg)															
	Male								Female							
	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Count	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000
Minimum	18.1	12.4	11.0	8.56	6.45	6.38	5.88	4.85	18.9	12.8	11.4	8.90	6.65	6.55	6.06	5.03
Maximum	97.3	55.3	41.7	32.1	27.1	21.9	18.9	17.7	102	56.6	42.6	33.1	27.8	22.5	19.3	18.2
Mean	41.8	29.6	22.1	17.1	14.0	12.0	10.5	9.46	43.2	30.6	22.8	17.6	14.4	12.4	10.8	9.72
%CV	21.9	19.5	20.4	18.6	18.5	18.3	17.9	17.6	21.8	19.3	20.2	18.5	18.4	18.2	17.7	17.5
Geometric mean	40.8	29.1	21.6	16.8	13.8	11.8	10.4	9.32	42.3	30.0	22.3	17.3	14.2	12.2	10.7	9.58
Geometric %CV	21.7	19.3	20.2	18.5	18.5	18.2	17.8	17.5	21.6	19.2	20.0	18.3	18.4	18.0	17.7	17.4
Percentile																
1st	25.0	18.7	13.6	11.1	8.79	7.75	6.86	6.14	25.9	19.4	14.1	11.6	9.07	7.99	7.12	6.34
5th	28.5	21.1	15.5	12.4	10.1	8.75	7.75	6.99	29.6	21.8	16.0	12.8	10.5	9.04	7.98	7.20
10th	30.8	22.7	16.6	13.2	10.9	9.41	8.26	7.44	32.0	23.6	17.3	13.6	11.2	9.68	8.50	7.66
15th	32.6	23.8	17.5	13.8	11.4	9.77	8.63	7.80	33.9	24.6	18.1	14.2	11.7	10.0	8.87	8.01
20th	34.0	24.8	18.2	14.3	11.8	10.1	8.92	8.07	35.2	25.6	18.8	14.8	12.2	10.4	9.20	8.29
25th	35.1	25.6	18.9	14.8	12.2	10.4	9.17	8.30	36.4	26.4	19.5	15.3	12.6	10.8	9.43	8.54
30th	36.3	26.3	19.5	15.2	12.6	10.7	9.42	8.52	37.5	27.2	20.2	15.7	13.0	11.1	9.70	8.77
35th	37.4	27.0	20.1	15.6	12.9	11.0	9.67	8.73	38.7	27.8	20.8	16.1	13.3	11.3	9.95	8.97
40th	38.6	27.7	20.6	16.0	13.2	11.3	9.91	8.93	40.0	28.6	21.3	16.5	13.6	11.6	10.2	9.19
45th	39.7	28.3	21.1	16.3	13.5	11.6	10.2	9.11	41.1	29.3	21.8	16.8	13.9	11.9	10.4	9.35
50th	40.8	29.0	21.7	16.8	13.9	11.9	10.4	9.31	42.2	30.0	22.3	17.3	14.3	12.2	10.7	9.56
55th	41.9	29.8	22.1	17.2	14.2	12.1	10.7	9.52	43.4	30.7	22.9	17.7	14.6	12.5	11.0	9.78
60th	43.2	30.5	22.8	17.6	14.5	12.4	10.9	9.76	44.7	31.5	23.5	18.1	14.9	12.8	11.2	10.0
65th	44.5	31.3	23.4	18.1	14.8	12.7	11.2	9.97	46.0	32.3	24.1	18.6	15.2	13.0	11.5	10.2
70th	45.9	32.2	24.0	18.6	15.2	13.0	11.4	10.2	47.3	33.2	24.8	19.1	15.6	13.3	11.8	10.5
75th	47.4	33.2	24.7	19.0	15.6	13.3	11.7	10.5	49.0	34.3	25.5	19.6	16.0	13.7	12.1	10.8
80th	49.3	34.2	25.6	19.6	16.1	13.8	12.1	10.8	51.0	35.3	26.3	20.2	16.5	14.1	12.4	11.1
85th	51.8	35.4	26.6	20.3	16.7	14.3	12.5	11.2	53.5	36.5	27.4	20.9	17.2	14.7	12.8	11.4
90th	54.2	37.3	28.1	21.2	17.5	14.9	13.0	11.6	55.9	38.4	28.9	21.8	18.0	15.3	13.4	11.9
95th	57.9	40.0	30.3	22.6	18.6	15.9	13.9	12.5	59.9	41.3	31.2	23.3	19.2	16.3	14.2	12.7
99th	66.2	45.6	34.4	25.5	21.1	18.1	15.7	14.0	68.0	47.1	35.7	26.4	21.7	18.4	16.1	14.3

Source: Applicant's PTA analysis report, Table 12
This simulation assumes phase 3 bioavailability.
Abbreviations: BA, bioavailability; C_{max}, maximum plasma concentration; CV, coefficient of variation

Table 114. Simulated Total-Drug C_{max} Following 3 g in Fed State (Assuming Phase 3 BA)

Variable	Summary statistics for zoliflodacin total-drug plasma C_{max} (µg/mL) by sex and body weight group (kg)															
	Male								Female							
	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Count	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000
Minimum	22.8	16.4	13.9	10.8	8.50	8.30	7.64	6.36	24.0	17.1	14.7	11.5	8.84	8.70	8.05	6.66
Maximum	121	73.0	55.4	42.5	36.0	30.1	25.6	23.7	128	75.9	57.6	44.3	37.5	31.0	26.4	24.6
Mean	54.3	38.7	29.0	22.5	18.6	16.0	14.1	12.6	56.8	40.4	30.3	23.5	19.3	16.6	14.6	13.1
%CV	22.3	20.0	20.9	19.1	19.0	18.9	18.4	18.1	22.1	19.8	20.7	18.9	18.8	18.6	18.2	17.9
Geometric mean	52.9	38.0	28.4	22.1	18.3	15.7	13.8	12.4	55.4	39.7	29.6	23.0	19.0	16.3	14.4	12.9
Geometric %CV	22.2	20.0	20.7	19.0	19.0	18.8	18.3	18.0	22.0	19.7	20.5	18.8	18.8	18.6	18.1	17.8
Percentile																
1st	31.8	23.8	17.6	14.4	11.6	10.0	8.94	8.05	33.5	25.0	18.5	15.1	12.1	10.5	9.37	8.42
5th	36.5	27.2	20.3	16.1	13.3	11.5	10.2	9.23	38.4	28.5	21.1	16.9	13.9	12.0	10.7	9.59
10th	39.7	29.4	21.7	17.3	14.3	12.4	10.9	9.85	41.7	30.9	22.7	18.1	14.9	12.9	11.4	10.3
15th	42.2	31.0	22.8	18.1	15.0	12.9	11.4	10.4	44.2	32.3	23.9	18.9	15.6	13.4	11.9	10.8
20th	44.0	32.3	23.8	18.9	15.6	13.4	11.8	10.7	46.1	33.7	24.9	19.6	16.2	13.9	12.3	11.1
25th	45.5	33.3	24.6	19.5	16.2	13.8	12.2	11.1	47.6	34.8	25.8	20.3	16.8	14.3	12.6	11.5
30th	46.9	34.3	25.5	20.1	16.6	14.2	12.5	11.4	49.1	35.8	26.6	20.9	17.3	14.8	13.0	11.8
35th	48.4	35.2	26.3	20.6	17.0	14.6	12.9	11.6	50.8	36.8	27.5	21.4	17.7	15.2	13.4	12.1
40th	50.0	36.0	27.0	21.1	17.5	15.0	13.2	11.9	52.4	37.7	28.2	22.0	18.2	15.6	13.7	12.4
45th	51.5	37.0	27.7	21.6	18.0	15.4	13.5	12.2	53.9	38.6	28.9	22.5	18.7	16.0	14.0	12.6
50th	53.0	37.9	28.5	22.2	18.4	15.8	13.9	12.4	55.5	39.6	29.7	23.1	19.1	16.4	14.4	12.9
55th	54.6	38.8	29.3	22.7	18.8	16.1	14.2	12.7	57.1	40.6	30.4	23.7	19.5	16.8	14.8	13.2
60th	56.2	40.0	30.0	23.3	19.2	16.5	14.6	13.1	58.6	41.7	31.3	24.2	20.0	17.2	15.1	13.5
65th	57.7	40.9	30.9	24.0	19.7	16.9	14.9	13.4	60.5	42.7	32.2	24.9	20.5	17.5	15.5	13.8
70th	59.6	42.3	31.6	24.6	20.2	17.3	15.3	13.7	62.4	44.1	33.0	25.6	21.0	17.9	15.8	14.2
75th	61.7	43.5	32.6	25.2	20.7	17.8	15.7	14.1	64.4	45.3	33.9	26.2	21.5	18.5	16.3	14.6
80th	64.1	44.8	33.6	25.9	21.4	18.4	16.2	14.5	67.0	46.8	35.0	26.9	22.2	19.0	16.7	15.0
85th	67.2	46.5	35.2	26.8	22.3	19.0	16.7	15.0	70.2	48.4	36.5	27.9	23.1	19.8	17.3	15.5
90th	70.7	48.9	37.1	28.2	23.3	19.8	17.5	15.6	73.8	51.0	38.6	29.3	24.3	20.6	18.1	16.2
95th	75.8	52.8	40.0	30.2	24.9	21.2	18.6	16.7	79.2	54.8	41.5	31.3	25.8	22.0	19.2	17.3
99th	86.8	59.9	45.7	34.2	28.2	24.3	21.3	18.8	90.5	62.5	47.4	35.3	29.2	25.1	21.9	19.3

Source: Applicant's PTA analysis report, Table 13
This simulation assumes phase 3 bioavailability.
Abbreviations: BA, bioavailability; C_{max}, maximum plasma concentration; CV, coefficient of variation

PK-PD PTA analyses were carried out using the final PopPK model, nonclinical PK-PD targets for efficacy, in vitro surveillance data, and MIC data from a phase 3 clinical trial to support the proposed dosing regimen and the criteria for the in vitro susceptibility testing for zoliflodacin against *N. gonorrhoeae*. As a conservative approach, the $AUC_{0-\infty}$ values simulated assuming the phase 1 bioavailability estimate, the low exposure scenario, are mainly discussed. The free-drug $AUC_{0-\infty}$ ($fAUC_{0-\infty}$) values were calculated by multiplying the total-drug $AUC_{0-\infty}$ by the plasma unbound fraction of 0.17. The PK-PD target was $fAUC_{0-\infty}$: MIC ratio of 70.6, which were determined to be associated with suppression of mutant amplification in *N. gonorrhoeae* (See Section 14.1.1). The probability of PK-PD target attainment was further calculated for each fixed MIC value ranging from 0.008 to 1 µg/mL. The Applicant also calculated the weighted average of percent probabilities of PK-PD target attainment using the zoliflodacin MIC distribution ranges from the phase 3 trial and in the vitro surveillance data.

The summary of the simulated free-drug $AUC_{0-\infty}$ following a single dose of zoliflodacin 3 g PO using the phase 1 relative bioavailability estimate is presented in Table 115 (fasted status) and Table 116 (fed status). The percent probabilities of PK-PD target attainment were presented in Table 117 (in male participants) and Table 118 (in female participants).

Table 115. Simulated Free-Drug AUC Following Zoliflodacin 3 g in a Fasted State (Assuming Phase 1 BA)

Variable	Summary statistics for zoliflodacin free-drug plasma $AUC_{0-\infty}$ (µg•h/mL) by sex and body weight group (kg)															
	Male								Female							
	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Count	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000
Minimum	21.7	19.0	14.8	11.4	10.1	8.67	8.12	7.81	25.2	22.0	17.2	13.3	11.8	10.1	9.43	9.07
Maximum	104	74.1	63.8	44.1	39.4	32.5	35.4	26.9	120	86.0	74.2	51.3	45.7	37.7	41.1	31.2
Mean	48.4	36.9	29.2	23.9	20.5	18.1	16.4	15.0	56.2	42.8	33.9	27.8	23.8	21.0	19.1	17.4
%CV	21.3	19.9	20.5	19.2	19.5	19.0	19.1	18.6	21.3	19.9	20.5	19.2	19.5	19.0	19.1	18.6
Geometric mean	47.3	36.2	28.6	23.5	20.1	17.8	16.1	14.7	54.9	42.0	33.2	27.3	23.4	20.7	18.7	17.1
Geometric %CV	21.2	19.6	20.2	19.1	19.3	18.9	18.9	18.6	21.2	19.6	20.2	19.1	19.3	18.9	18.9	18.6
Percentile																
1st	28.8	22.9	18.2	15.0	13.1	11.7	10.7	9.47	33.5	26.6	21.1	17.4	15.2	13.5	12.4	11.0
5th	33.2	26.1	20.6	17.0	14.6	13.0	11.9	10.8	38.5	30.3	24.0	19.8	17.0	15.1	13.8	12.5
10th	35.9	28.1	22.1	18.4	15.7	13.9	12.6	11.6	41.8	32.7	25.7	21.4	18.2	16.2	14.7	13.5
15th	37.9	29.5	23.1	19.3	16.6	14.6	13.2	12.2	44.1	34.3	26.9	22.5	19.3	17.0	15.4	14.1
20th	39.5	30.6	24.0	20.1	17.2	15.2	13.7	12.6	45.9	35.6	27.9	23.4	20.0	17.6	15.9	14.7
25th	41.1	31.7	24.8	20.7	17.7	15.7	14.2	13.0	47.7	36.8	28.8	24.1	20.6	18.2	16.5	15.2
30th	42.3	32.7	25.6	21.3	18.2	16.1	14.6	13.4	49.1	38.0	29.7	24.8	21.2	18.7	17.0	15.6
35th	43.6	33.6	26.4	21.9	18.7	16.6	15.0	13.8	50.7	39.0	30.6	25.4	21.8	19.2	17.4	16.0
40th	45.0	34.4	27.1	22.4	19.2	16.9	15.4	14.1	52.2	40.0	31.5	26.1	22.3	19.7	17.9	16.4
45th	46.4	35.2	27.8	23.0	19.6	17.3	15.7	14.4	53.9	40.9	32.3	26.7	22.8	20.1	18.3	16.8
50th	47.6	36.1	28.5	23.4	20.1	17.7	16.1	14.8	55.3	41.9	33.2	27.2	23.4	20.6	18.7	17.1
55th	48.8	36.9	29.4	24.1	20.6	18.2	16.5	15.1	56.7	42.9	34.1	28.0	24.0	21.1	19.2	17.6
60th	50.0	37.9	30.1	24.6	21.1	18.6	16.9	15.5	58.1	44.1	35.0	28.6	24.5	21.6	19.7	18.0
65th	51.2	39.0	31.1	25.2	21.7	19.1	17.4	15.9	59.5	45.3	36.1	29.3	25.2	22.2	20.2	18.5
70th	52.8	40.0	32.0	26.0	22.3	19.6	17.8	16.3	61.3	46.5	37.1	30.2	25.9	22.7	20.7	18.9
75th	54.6	41.2	32.9	26.7	22.9	20.2	18.3	16.7	63.4	47.9	38.2	31.1	26.6	23.4	21.2	19.4
80th	56.5	42.6	34.0	27.6	23.7	20.9	18.9	17.2	65.6	49.4	39.5	32.1	27.5	24.2	21.9	20.0
85th	58.7	44.5	35.3	28.7	24.5	21.6	19.6	17.8	68.2	51.7	41.1	33.3	28.5	25.1	22.7	20.7
90th	62.0	46.4	37.2	30.2	25.8	22.6	20.5	18.6	72.0	53.9	43.2	35.1	30.0	26.3	23.8	21.6
95th	66.7	49.8	39.7	32.2	27.9	24.2	22.0	20.0	77.5	57.8	46.2	37.4	32.4	28.1	25.6	23.2
99th	77.1	57.0	45.5	36.7	32.0	27.6	25.0	22.4	89.5	66.2	52.8	42.6	37.2	32.0	29.1	26.0

Source: Applicant's PTA analysis report, Table 6
This simulation assumed the phase 1 bioavailability.
Abbreviations: AUC, area under the concentration-time curve; BA, bioavailability; CV, coefficient of variation

Table 116. Simulated Free-Drug AUC Following 3 g in a Fed State (Assuming Phase 1 BA)

Variable	Summary statistics for zoliflodacin free-drug plasma $AUC_{0-\infty}$ (µg•h/mL) by sex and body weight group (kg)															
	Male								Female							
	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200	≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Count	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000	3000
Minimum	32.5	28.4	22.2	17.1	15.1	13.0	12.1	11.7	37.7	32.9	25.8	19.9	17.6	15.1	14.1	13.6
Maximum	155	111	95.5	66.0	58.9	48.6	53.0	40.2	180	129	111	76.7	68.4	56.4	61.5	46.7
Mean	72.3	55.1	43.7	35.8	30.7	27.1	24.6	22.4	84.0	64.0	50.7	41.6	35.7	31.5	28.5	26.1
%CV	21.3	19.9	20.5	19.2	19.5	19.0	19.1	18.6	21.3	19.9	20.5	19.2	19.5	19.0	19.1	18.6
Geometric mean	70.7	54.1	42.8	35.2	30.1	26.6	24.1	22.1	82.2	62.8	49.7	40.9	35.0	30.9	28.0	25.6
Geometric %CV	21.2	19.6	20.2	19.1	19.3	18.9	18.9	18.6	21.2	19.6	20.2	19.1	19.3	18.9	18.9	18.6
Percentile																
1st	43.1	34.3	27.2	22.4	19.5	17.4	16.0	14.2	50.1	39.8	31.5	26.0	22.7	20.3	18.5	16.5
5th	49.6	39.0	30.9	25.5	21.8	19.4	17.8	16.2	57.6	45.3	35.8	29.6	25.4	22.6	20.7	18.8
10th	53.8	42.1	33.1	27.5	23.4	20.9	18.9	17.3	62.5	48.9	38.5	32.0	27.2	24.2	21.9	20.1
15th	56.7	44.1	34.6	28.9	24.8	21.9	19.8	18.2	65.9	51.2	40.2	33.6	28.8	25.4	23.0	21.1
20th	59.1	45.8	35.9	30.1	25.7	22.7	20.5	18.9	68.7	53.2	41.7	35.0	29.9	26.4	23.8	21.9
25th	61.5	47.4	37.1	31.0	26.5	23.5	21.2	19.5	71.4	55.1	43.1	36.1	30.7	27.3	24.6	22.7
30th	63.3	48.9	38.3	31.9	27.3	24.1	21.9	20.1	73.5	56.8	44.5	37.1	31.7	28.0	25.4	23.4
35th	65.3	50.3	39.4	32.7	28.0	24.8	22.4	20.6	75.8	58.4	45.8	38.0	32.5	28.8	26.0	24.0
40th	67.3	51.5	40.6	33.6	28.7	25.4	23.0	21.1	78.1	59.8	47.1	39.0	33.3	29.4	26.7	24.5
45th	69.4	52.7	41.6	34.4	29.4	25.9	23.5	21.6	80.6	61.2	48.3	39.9	34.1	30.1	27.3	25.1
50th	71.2	54.0	42.7	35.1	30.1	26.5	24.1	22.1	82.7	62.7	49.6	40.7	35.0	30.8	28.0	25.7
55th	73.0	55.3	43.9	36.0	30.9	27.2	24.7	22.6	84.8	64.2	51.0	41.8	35.8	31.6	28.7	26.3
60th	74.8	56.8	45.0	36.8	31.6	27.8	25.3	23.2	86.9	65.9	52.3	42.8	36.7	32.3	29.4	26.9
65th	76.6	58.3	46.5	37.8	32.4	28.6	26.0	23.8	89.0	67.7	54.0	43.8	37.6	33.2	30.2	27.6
70th	78.9	59.9	47.8	38.9	33.4	29.3	26.7	24.3	91.7	69.6	55.5	45.1	38.7	34.0	31.0	28.3
75th	81.7	61.7	49.2	40.0	34.2	30.2	27.3	25.0	94.9	71.6	57.2	46.5	39.8	35.1	31.8	29.1
80th	84.5	63.7	50.8	41.3	35.4	31.2	28.2	25.8	98.1	74.0	59.0	48.0	41.2	36.2	32.8	29.9
85th	87.9	66.5	52.9	42.9	36.7	32.3	29.3	26.6	102	77.3	61.4	49.8	42.6	37.6	34.0	30.9
90th	92.7	69.5	55.7	45.2	38.6	33.8	30.6	27.9	108	80.7	64.7	52.5	44.8	39.3	35.6	32.4
95th	99.8	74.4	59.5	48.1	41.8	36.2	33.0	29.9	116	86.5	69.0	55.9	48.5	42.1	38.3	34.7
99th	115	85.2	68.0	54.9	47.9	41.2	37.4	33.5	134	99.0	79.0	63.7	55.6	47.9	43.5	38.9

Source: Applicant's PTA analysis report, Table 7
This simulation assumed the phase 1 bioavailability.
Abbreviations: AUC, area under the concentration-time curve; BA, bioavailability; CV, coefficient of variation

Table 117. Percent Probability of PK-PD Target Attainment by MIC Following 3 g in Male Simulated Patients (PK-PD Target: *f*AUC/MIC Ratio =70.6)

Fasted or Fed	MIC (µg/mL)[b]	Percent probabilities of PK-PD target attainment by body weight group (kg)							
		≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Fasted	≤ 0.008	100	100	100	100	100	100	100	100
	0.015	100	100	100	100	100	100	100	100
	0.03	100	100	100	100	100	100	100	100
	0.06	100	100	100	100	100	100	100	100
	0.12	100	100	100	100	100	100	99.9	99.7
	0.25	100	100	99.4	93.1	75.4	50.9	31.7	16.1
	0.5	91.4	54.5	15.2	1.57	0.17	0	0.03	0
	1	2.47	0.03	0	0	0	0	0	0
	Overall								
	Phase 3 data[c]	>99.9	99.8	99.6	99.3	98.6	97.6	96.8	96.1
	In vitro surveillance[d]	100	100	>99.9	99.7	99.1	98.1	97.4	96.7
Fed	≤ 0.008	100	100	100	100	100	100	100	100
	0.015	100	100	100	100	100	100	100	100
	0.03	100	100	100	100	100	100	100	100
	0.06	100	100	100	100	100	100	100	100
	0.12	100	100	100	100	100	100	100	100
	0.25	100	100	100	>99.9	99.9	98.8	95.4	88.0
	0.5	>99.9	98.4	82.1	48.7	20.7	6.83	2.33	0.37
	1	51.8	8.53	0.67	0	0	0	0	0
	Overall								
	Phase 3 data[c]	>99.9	>99.9	99.9	99.8	99.7	99.6	99.4	99.1
	In vitro surveillance[d]	100	100	100	>99.9	>99.9	>99.9	99.8	99.5

Source: Applicant's PTA analysis report, Table 17
Shaded cells indicate percent probabilities of PK-PD target attainment >90%
[b]. Zoliflodacin MIC value
[c, d] The weighted percent probability of PK-PD target attainment over an MIC distribution from the phase 3 data and the in vitro surveillance data
Abbreviations: *f*AUC, free area under the concentration time curve; MIC, minimum inhibitory plasma concentration; PK-PD, pharmacokinetic-pharmacodynamic

Table 118. Percent Probability of PK-PD Target Attainment by MIC Following 3 g in Female Simulated Patients (PK-PD Target: *f*AUC/MIC Ratio =70.6)

Fasted or Fed	MIC (µg/mL)[b]	Percent probabilities of PK-PD target attainment by body weight group (kg)							
		≥20-<35	≥35-<50	≥50-<75	≥75-<100	≥100-<125	≥125-<150	≥150-<175	≥175-≤200
Fasted	≤ 0.008	100	100	100	100	100	100	100	100
	0.015	100	100	100	100	100	100	100	100
	0.03	100	100	100	100	100	100	100	100
	0.06	100	100	100	100	100	100	100	100
	0.12	100	100	100	100	100	100	100	100
	0.25	100	100	99.9	98.8	92.6	79.8	62.3	44.4
	0.5	98.2	81.0	38.1	9.40	1.83	0.23	0.07	0
	1	11.8	0.40	0.03	0	0	0	0	0
	Overall								
	Phase 3 data[c]	>99.9	99.9	99.8	99.6	99.3	98.8	98.1	97.3
	In vitro surveillance[d]	100	100	>99.9	>99.9	99.7	99.2	98.6	97.9
Fed	≤ 0.008	100	100	100	100	100	100	100	100
	0.015	100	100	100	100	100	100	100	100
	0.03	100	100	100	100	100	100	100	100
	0.06	100	100	100	100	100	100	100	100
	0.12	100	100	100	100	100	100	100	100
	0.25	100	100	100	100	>99.9	99.8	99.3	97.3
	0.5	100	99.8	95.8	78.2	48.3	23.8	10.9	4.17
	1	76.4	27.5	4.00	0.20	0	0	0	0
	Overall								
	Phase 3 data[c]	100	>99.9	>99.9	99.9	99.8	99.7	99.6	99.5
	In vitro surveillance[d]	100	100	100	100	>99.9	>99.9	>99.9	99.9

Source: Applicant's PTA analysis report, Table 18
The simulations were conducted assuming the phase 1 BA estimate
Shaded cells indicate percent probabilities of PK-PD target attainment >90%
[b]. zoliflodacin MIC value
[c, d] The weighted percent probability of PK-PD target attainment over an MIC distribution from the phase 3 data and the in vitro surveillance data.
Abbreviations: *f*AUC, free area under the concentration time curve; MIC, minimum inhibitory plasma concentration; PK-PD, pharmacokinetic-pharmacodynamic

At the proposed dosage for patients weighing >50 kg (3 g in a fed state), the percent probabilities of PK-PD target attainment are predicted to exceed 90% at the MIC value of 0.25 µg/mL in male and female participants across weight groups, except for a weight group (≥175 to 200 kg) in male participants. At the proposed dosage for patients weighing 35 kg to <50 kg (3 g in a fasted state), the percent probabilities of PK-PD target attainment are predicted to exceed 90% at an MIC value of 0.25 µg/mL for both male and female patients. The weighted average of percent probabilities of PK-PD target attainment using the zoliflodacin MIC distribution from the phase 3 trial and in the vitro surveillance data also are predicted to be ≥90% across the body weight groups. Overall, with the proposed dosing regimen, it is expected to achieve >90% probability of target attainment (*f*AUC/MIC ratio =70.6) across body weight for the MIC values ≤0.25 µg/mL.

14.5.2. Physiologically Based Pharmacokinetic Analysis

The objective of this review is to evaluate the adequacy of the Applicant's PBPK analyses to predict the impact of the following intrinsic and extrinsic factors on zoliflodacin PK: 1) CYP3A inhibition with or without food effect; 2) CYP3A induction; 3) mild, moderate, and severe hepatic impairment. The predicted impact of zoliflodacin as a precipitant on the PK of

transporter (i.e., P-gp, BCRP, OAT1, OAT3, OATP1B1, OATP1B3, MATEs) substrates was also assessed.

The Division of Pharmacometrics has reviewed the submitted PBPK reports, modeling and simulation files, and response to the FDA's IR received on July 2, 2025 to conclude the following:

The PBPK model of zoliflodacin adequately described observed plasma PK profile and parameters of single dose zoliflodacin in healthy adult participants under fasted and fed conditions.

DDI simulations of strong CYP3A inhibition under fasted condition show zoliflodacin AUC would increase by ~1.4-fold with coadministration with itraconazole. Food is not expected to impact the magnitude of DDI.

DDI simulations of strong and moderate CYP3A induction indicate zoliflodacin AUC would decrease 56% and 41% with coadministration of rifampin (strong) or efavirenz (moderate) CYP3A4 inducer, respectively.

The likelihood of interaction with P-gp, BCRP, MATEs, OAT1, OAT3, OATP1B1, and OATP1B3 substrates cannot be ruled out. However, due to zoliflodacin's single dose administration and short $t_{1/2}$, the overall DDI risk is considered low and no dose adjustments have been proposed when zoliflodacin is coadministered.

The hepatic impairment simulations were not reviewed due to limited relevancy; hepatic impairment is not considered a significant clinical risk for this single dose administration drug.

Background

Zoliflodacin inhibits bacterial deoxyribonucleic acid type II topoisomerase and is active in vitro against *N. gonorrhoeae*. Zoliflodacin is indicated for the treatment of uncomplicated gonorrhea. The Applicant proposed the following dosage: 3 g administered as a single oral dose with food in patients weighing ≥50 kg or 3 g administered as a single oral dose on an empty stomach (1 hour before or 2 hours after food) in patients weighing ≥35 to <50 kg.

Zoliflodacin is a biopharmaceutics classification system class II compound (i.e., high permeability and low solubility). Zoliflodacin exposure was shown to increase in a dose-proportional manner up to 800 mg and less than dose-proportionally between 800 mg and 4 g. Zoliflodacin was rapidly absorbed with a median T_{max} of 1.50 to 4.00 hours in the fasted state. The effect of food on the absorption of zoliflodacin was evaluated in three clinical studies: Study D4930C00001 (high-fat, high-calorie meal), Study STI_Zoli002 (high-fat, high-calorie meal), and Study STI_Zoli003 (moderate-fat, moderate-calorie meal). A consistent trend was observed across all studies when zoliflodacin 3 g was administered in a fed or fasted state; food generally increased zoliflodacin C_{max} by approximately 1.5-fold and AUC by approximately 1.5- to 2-fold and delayed T_{max}. Population PK analysis showed that clinically relevant covariates included body weight and prandial state.

The geometric mean apparent volume of distribution ranged from 76 to 94 L under fed conditions and 161 to 163 L under fasted conditions in healthy participants, suggesting distribution to the extravascular space. In vitro human plasma protein binding was 83% (17% unbound). In vitro blood-to-plasma ratio was 0.69.

In healthy participants, the geometric mean elimination $t_{1/2}$ of zoliflodacin was approximately 6 hours and the geometric mean apparent clearance was approximately 17 L/h. In patients with gonorrhea, the geometric mean elimination $t_{1/2}$ of zoliflodacin was similar (5.36 hours) and the geometric mean apparent clearance was approximately 14 L/h.

In the human ADME Study D4930C00003, following administration of a single oral 3 g dose of [^{14}C]-zoliflodacin, 18.2% of total radioactive dose was recovered in urine (with 2.5% as parent drug) and 79.6% of the dose was recovered from feces (with approximately 1.5% as parent drug). Zoliflodacin is minimally renally eliminated. Zoliflodacin undergoes extensive biotransformation to 31 metabolites. The primary systemic routes of biotransformation were CYP-mediated oxidation and non-CYP mediated hydrolysis. Because these pathways produce the same metabolites, the exact contribution of each to the overall metabolic clearance is not clear. Metabolites formed via reduction in the intestine, likely by gut microflora, were present in feces.

In vitro, zoliflodacin is predominately metabolized by CYP3A4/5 (68.4%), with lesser contributions from CYPs 1A2, 2C9, 2C8, and 2C19.

Zoliflodacin is an in vitro inhibitor of P-gp (IC_{50}=299μM), BCRP (IC_{50}=55.5μM), OATP1B1 (IC_{50}=7.7μM), OATP1B3 (IC_{50}=9.23μM), OAT1 (IC_{50}=47.7μM), and OAT3 (IC_{50}=13.5μM). Zoliflodacin showed limited inhibition of MATE1 and MATE2K, with IC_{50} values >100μM. Zoliflodacin is not an inhibitor of OCT2.

Study STI_Zoli003 included a DDI evaluation (Cohort 2) to investigate the effect of strong CYP3A4 inhibition by itraconazole on the pharmacokinetics of zoliflodacin 3 g single dose. Itraconazole increased zoliflodacin $AUC_{0\text{-last}}$ and $AUC_{0\text{-}\infty}$ by 1.39- and 1.38-fold, respectively. Zoliflodacin C_{max} was nearly unchanged (1.03-fold increase).

No additional clinical studies were conducted with CYP3A4 inducers, as well as P-gp, BCRP, OATP1B1/3, OAT1/3, or MATEs substrates.

Methods

The Applicant submitted four PBPK reports (AZD0914-MS-002, SN-000914-2016-03, SN-000914-2016-04, PC0914-2024-0015). The final PBPK model and simulations are described in report PC0914-2024-0015 which is the focus of this review.

Model Development

The Applicant used Simcyp® PBPK Simulator Version 22 (Certara UK Ltd., Sheffield, UK) for all final PBPK modeling and simulation. The final model input parameters used for the simulations of zoliflodacin pharmacokinetics are presented in Table 118 and described briefly below.

Physicochemical and Binding Properties

Zoliflodacin is a diprotic acid. See Table 119 for the physicochemical and binding parameters.

Absorption

The advanced dissolution, absorption and metabolism model was used to describe zoliflodacin's absorption. The mechanistic P_{eff} (MechPeff) model was used to predict the effective human

jejunum I permeability ($P_{eff,man}$) value (7.75×10^{-4} cm/s). The diffusion layer model, specifically the particle (size) population balance model, was used to describe the intrinsic dissolution rate of zoliflodacin.

Distribution

The whole-body full PBPK model was used to describe zoliflodacin's distribution. Using Method 2, the initial predicted value of the volume of distribution at steady state (V_{ss}) was 0.3 L/kg, which was lower than the V_{ss} observed in all nonclinical species and resulted in the overprediction of C_{max} in healthy participants administered single 3 g oral dose of zoliflodacin (Study STI_Zoli003). Therefore, the predicted tissue-to-plasma partition coefficient (K_p) for adipose tissue was adjusted to recover the observed C_{max} and the final predicted V_{ss} value was 0.56 L/kg.

Elimination

The total human liver microsomes Cl_{int} was calculated from the apparent oral clearance (18.3 L/h) reported in Study STI_Zoli003 using the retrograde model. The proportion of human liver microsomes Cl_{int} mediated by CYP3A4 was determined based on the observed 1.4-fold increase in zoliflodacin $AUC_{0\text{-last}}$ due to itraconazole. The remaining clearance mediated by non-CYP3A pathways were grouped under a generic hepatic clearance pathway, accounting for approximately 68% of the total hepatic clearance (i.e., CYP and non-CYP mediated pathways). The Applicant estimated that non-CYP metabolism contributed to at least 30% of hepatic clearance (Study SN-000914-2016-02). Renal clearance was estimated to be 0.45 L/h based on Study D4930C00001.

Metabolism

The fm_{CYP3A} value (0.31) was estimated from one DDI study with itraconazole (Study STI_Zoli003).

Food Effect

For the simulations conducted under fed conditions, the following settings in the Substrate and Trial Design tabs were active:

"Substrate" (zoliflodacin): "Absorption" → "GI Tract" → "Permeability" → See "Fed" row for predicted advanced dissolution, absorption and metabolism model regional permeabilities

"Substrate" (zoliflodacin): "Absorption" → "GI Tract" → "Formulation" → "Transit Times" → See "Segregated Transit Times (Mean Residence Time [MRT]) (h)" for predicted MRT values in "Fine Particles"

Trial Design: Dosing → See "Fed" selected

Table 119. PBPK Model Input Parameters for Zoliflodacin

PARAMETER	Value	Reference
Physicochemical and Binding Parameters		
Molecular Weight (g/mol)	487.4	Report AZ13420914 KMN004
Log P	1.86	Report SN-000914-2016-03
Compound type	Diprotic Acid	Report SN-000914-2016-03
pKa	7.15, 11.9	Report SN-000914-2016-03
B:P	0.7	Clinical Study D4930C00003
fu	0.17	Report KPJ005
Main binding protein	HSA	Assumed
Absorption Model – ADAM Model		
fu_{gut}	1	Simcyp default
$P_{trans,0}$ ($x10^{-6}$ cm/s)	142.71	Predicted with Log $P_{o:w}$ method 1 in MechPeff model
Predicted P_{eff} (10^{-4} cm/s) in Jejunum 1	7.75	Predicted using Simcyp V22
Effective concentration at Epithelial Surface	Total Concentration (Bound + Unbound)	-
Use UBL Fluid volumes	Yes	-
Intrinsic Solubility (mg/mL)	0.2	Bradford *et al.*, 2020
Log $K_{m:w}$ Neutral	3.6664	Predicted using Simcyp V22
Log $K_{m:w}$ Ion	2.6664	Predicted using Simcyp V22
Use Segregated Transit Time Model	Yes	-
Use only the ascending colon transit time	Yes	-
Fine Particles: Ascending Colon MRT (h)	12	Changed to match the colon MRT from the default transit time model
Fine Particles: Ascending	30	Changed to match the colon MRT CV%
Colon MRT CV (%)		from the default transit time model
Fine Particles Stomach MRT under Fed state (h)	2.36	Adjusted from (b) (4) to 2.36 hours to capture the t_{max} observed in the fed state (Clinical Study STI_Zoli001)
Fine Particles Stomach MRT with High Fat Diet (h)	2.36	Assumed to be the same as that under fed state
Formulation type	Suspension	Selected based on relevant clinical dosage form
DLM Model options	Particle Population Balance Model	-
Distribution Model – Full PBPK Model		
V_{SS} (L/kg)	0.56	Predicted with Method 2 in Simcyp V22
Adipose Tissue K_p	1.257	Optimised using the observed geometric mean C_{max} in healthy subjects following a single oral dose of 3 g zoliflodacin (ZoliPa) (Clinical Study STI_Zoli003)
Elimination Parameters		
CYP3A4 CL_{int} (µL/min/mg)	5.17	Total HLM CL_{int} was calculated from the CL_{PO} (18.3 L/kg) reported in Clinical Study STI_Zoli003 using the retrograde model. The proportion of HLM CL_{int} mediated by CYP3A4 was determined based on the fold increase in zoliflodacin $AUC_{0\text{-last}}$ observed in the presence of itraconazole (1.3877), as reported in the same study.
Additional HLM CL_{int} (µL/min/mg) (represents clearance via other CYP enzymes, minor relative to CYP3A4, non-CYP metabolism, biliary excretion, and other clearance mechanisms)	10.99	
CL_R (L/h)	0.45	Clinical Study D4930C00001

Source: Report PC0914-2024-0015 Table 3
Abbreviations: ADAM, advance dissolution, absorption, and metabolism; CL, clearance; C_{max}, maximum plasma concentration; CV, coefficient of variation; CYP, cytochrome P 450; fu, free fraction of drug in plasma; HLM, human liver microsomes; HAD, human serum albumin; Log, logarithm; Km, Michaelis-Menten constant; Kp; partition coefficient; MRT, mean residence time; PBPK, physiologically based pharmacokinetic, UBL, ubiquitin-like protein; Vss, steady-state volume of distribution; ZoliPa, zoliflodacin granules for oral suspension manufactured by Patheon

Model Verification

The zoliflodacin PBPK model was verified against clinical PK data from the following studies:

Study D4930C00001: single oral doses of 800 mg, 1.6, 3, and 4 g zoliflodacin in healthy participants under fasted condition (part A), single oral doses of 1.5 and 3 g zoliflodacin in healthy participants under fasted and fed conditions (part B)

Studies DMID 16-0110 and DMID 16-0118 (NIAID): single oral doses of 2 and 4 g zoliflodacin in healthy participants under fasted condition

Trial STI_Zoli001: single oral dose of 3 g zoliflodacin in participants with gonorrhea

Study STI_Zoli002: single oral doses of 3 and 4 g zoliflodacin in healthy participants under fasted and fed conditions

Study STI_Zoli003 (Cohort 1): single oral dose of 3 g zoliflodacin in healthy participants under fasted and fed conditions

The contribution of CYP3A-mediated metabolism (fm_{CYP3A}) was verified using clinical DDI data from the following study:

Study STI_Zoli003 (Cohort 2): single oral dose of 3 g zoliflodacin in healthy participants on Days 1 and 9 under fasted conditions and strong CYP3A4 inhibitor itraconazole 400-mg loading dose on Day 4 followed by 200 mg once daily from Days 5 to 11 under fasted (when coadministered with zoliflodacin)

Model Application

The Applicant conducted the following simulations using drug and population PBPK models from Simcyp's library without modification:

Prediction of plasma zoliflodacin concentration in healthy participants following a single 3 g oral dose administered with or without ritonavir (strong CYP3A4 inhibitor), rifampin (strong CYP3A4 inducer), and efavirenz (moderate CYP3A4 inducer)

Prediction of plasma concentrations of transporter substrates – dabigatran (P-gp), metformin (MATE), methotrexate (OAT3), pravastatin (OATP1B), rosuvastatin (OATP1B/BCRP), and tenofovir (OAT1) – following coadministration with single oral dose of 3 g zoliflodacin under fasted and fed conditions

Prediction of plasma zoliflodacin concentration in healthy participants and patients with mild, moderate, and severe hepatic impairment following single 3 g dose.

Results

Q1. Can the Zoliflodacin PBPK Model Describe Zoliflodacin PK Under Fasted and Fed Conditions?

The base model of zoliflodacin initially overpredicted C_{max} (from Study STI_Zoli003) following a single oral dose of 3 g by 1.35-fold. To address this discrepancy, the Applicant conducted a manual sensitivity analysis to evaluate the impact of adipose K_p on the simulated C_{max}. An adipose K_p value of 1.257 (4-fold increase from default value) was selected as it recovered the observed C_{max}, with a predicted median T_{max} (2.95 hrs) within the observed range of 2.5 to 3 hrs

for a single oral dose of 3 g. The simulated geometric mean $AUC_{0-\infty}$, C_{max}, and T_{max} values were then within 0.8- to 1.25-fold of the observed data.

In the response to the IR received on July 2, 2025, the Applicant provided a rationale for adjusting the adipose tissue K_p parameter to address the overprediction of C_{max}. Since adipose tissue has one of the largest physiological volumes in the whole-body full PBPK model implemented in Simcyp® PBPK Simulator, it can be used as an empirical tool to influence the extent and rate of distribution. The Applicant stated "It is a common empirical approach in PBPK modelling to adjust the adipose tissue K_p value to improve the fit of plasma concentration-time profiles, particularly for parameters such as C_{max} and T_{max}. Another widely used approach is to adjust the global K_p scalar, which scales all tissue K_p values uniformly." Applicant stated that both approaches were assessed for zoliflodacin and adjustment of the adipose tissue K_p value to 1.257 provided a better recovery of both observed C_{max} and T_{max}.

The adipose K_p value was increased by 4-fold from the default predicted value, and it is possible that other factors may have contributed to the initial overpredicted C_{max}. Nevertheless, the predicted PK profile using the adjusted adipose K_p fit the observed pharmacokinetics adequately.

The final PBPK model reasonably describes the observed plasma concentration-time profiles of zoliflodacin following single oral doses of 800 mg up to 4 g in healthy participants under fasted conditions. The simulated PK parameters ($AUC_{0-\infty}$, C_{max}, T_{max}) were mostly within 1.5-fold of the observed data (Table 120). The observed and predicted plasma zoliflodacin concentration-time profiles following administration of a single oral 3-g dose under fasted condition is shown in Figure 10.

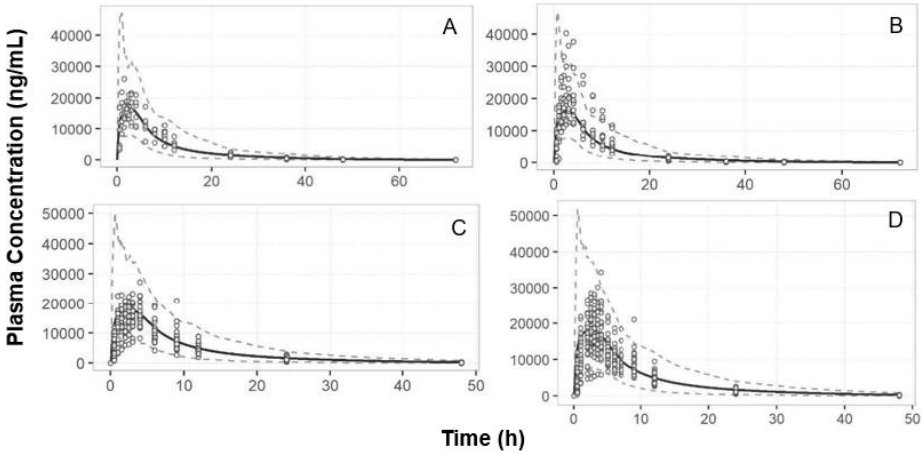
Table 120. Summary of Observed and Predicted PK Parameters of Zoliflodacin Following Single Oral Dose in Healthy Participants Under Fasted Conditions

Dose	Parameter	Predicted	Observed	Pred/Obs Ratio
800 mg[a]	$AUC_{0-\infty}$ (h*ng/mL)	72630 (68844-76624)	68300	1.06
	C_{max} (ng/mL)	11684 (11094-12306)	10000	1.17
	t_{max} (h)	1.91 (0.54-3.31)	2	0.95
1.5 g[b]	$AUC_{0-\infty}$ (h*ng/mL)	118781 (110393-127806)	109000	1.09
	C_{max} (ng/mL)	15096 (13914-16380)	15100	1.00
	t_{max} (h)	2.59 (0.54-4.90)	2.50	1.04
1.6 g[a]	$AUC_{0-\infty}$ (h*ng/mL)	120453 (113769-127530)	108000	1.12
	C_{max} (ng/mL)	15100 (14145-16120)	13400	1.13
	t_{max} (h)	2.52 (0.54-5.04)	1.5	1.68
2 g[c]	$AUC_{0-\infty}$ (h*ng/mL)	155588 (151527-159757)	103500	1.50
	C_{max} (ng/mL)	16537 (16016-17075)	11770	1.41
	t_{max} (h)	2.73 (0.35-5.98)	3.02	0.90

Dose	Parameter	Predicted	Observed	Pred/Obs Ratio
3 g[a,b,e,f]	$AUC_{0-\infty}$ (h*ng/mL)[a]	162403 (152357-173111)	183000	0.82
	C_{max} (ng/mL)[a]	17527 (16192-18971)	20200	0.74
	t_{max} (h)[a]	2.95 (0.54-5.83)	2.25	1.21
	$AUC_{0-\infty}$ (h*ng/mL)[b]	160263 (149356-171966)	195000	0.89
	C_{max} (ng/mL)[b]	16916 (15523-18434)	22900	0.87
	t_{max} (h)[b]	3.02 (0.54-5.83)	2.50	1.31
	$AUC_{0-\infty}$ (h*ng/mL)[e]	187691 (177326-198662)	171000	1.10
	C_{max} (ng/mL)[e]	18494 (17399-19657)	17600	1.05
	t_{max} (h)[e]	2.87 (0.43-5.91)	3.00	0.96
	$AUC_{0-\infty}$ (h*ng/mL)[f]	174234 (167015-181766)	165000	1.06
	C_{max} (ng/mL)[f]	18852 (17874-19883)	19400	0.97
	t_{max} (h)[f]	2.94 (0.43-5.83)	3.00	0.98
4 g[a,c,d,e]	$AUC_{0-\infty}$ (h*ng/mL)[a]	177338 (165438-190095)	209000	0.85
	C_{max} (ng/mL)[a]	18172 (16727-19742)	26100	0.70
	t_{max} (h)[a]	3.11 (0.54-6.12)	2.04	1.53
	$AUC_{0-\infty}$ (h*ng/mL)[d]	213521 (206597-220676)	174900	1.22
	C_{max} (ng/mL)[d]	19033 (18331-19762)	19360	0.98
	t_{max} (h)[d]	3.18 (0.36-6.95)	3.02	1.05
	$AUC_{0-\infty}$ (h*ng/mL)[d]	190393 (174688-207510)	213505	0.89
	C_{max} (ng/mL)[d]	18378 (16539-20421)	19857	0.93
	tmax (h)[d]	3.38 (0.54-6.23)	4.00	0.85
	$AUC_{0-\infty}$ (h*ng/mL)[e]	237469 (220723-255484)	224000	1.06
	C_{max} (ng/mL)[e]	19667 (18413-21007)	19900	0.99
	t_{max} (h)[e]	3.05 (0.46-6.24)	2.50	1.22

Source: Report PC0914-2024-0015 Table 7, Table 9, Table 10, Table 11, and Table 12
Data are presented as geometric mean and 90% confidence interval, except for t_{max} which is expressed as median (minimum, maximum).
[a] Study D4930C00001 Part A
[b] Study D4930C00001 Part B
[c] Study NIAID-16-0110
[d] Study NIAID-16-0118
[e] Study STI_Zoli002
[f] Study STI_Zoli003
Abbreviations: $AUC_{0-\infty}$, area under the concentration-time curve estimated to infinity; C_{max}, maximum plasma concentration; obs, observed; PK, pharmacokinetic; pred, predicted; t_{max}, time to maximum plasma concentration

Figure 9. Predicted and Observed PK Profile of Zoliflodacin After Single Oral Dose of 3 g in Healthy Participants Under Fasted Conditions



Source: Report PC0914-2024-0015 Figure 13 (panel C), Figure 16 (panel C), Figure 17 (panel A), Figure 18 (panel A)
Lines and circles represent simulated and observed data, respectively. Grey lines represent 5th and 95th percentiles and solid black line represents mean data for the simulated population.
Panel A: Study D4930C00001 Part A; panel B: Study D4930C00001 Part B; panel C: Study STI_Zoli002; panel D: Study STI_Zoli003 Cohort 1
Abbreviation: PK, pharmacokinetic

A model to describe zoliflodacin pharmacokinetics following single oral doses of 1.5, 3, and 4 g in healthy participants under fed conditions was also developed. The default fed ("high fat fed") model was used to simulate the food effect. The model initially underpredicted T_{max} using the default/predicted MRT of fine particles in the stomach; therefore, the stomach MRT for fine particles under a fed state was adjusted from 1.18 to 2.36 hours. The observed changes in $AUC_{0-\infty}$ and C_{max}, under high-fat fed condition, were approximately 1.5- to 2-fold and approximately 1.5-fold, respectively. The predicted food effects were approximately 1.5- and 1.3-fold for $AUC_{0-\infty}$ and C_{max}, respectively (See Table 121 and Table 122).

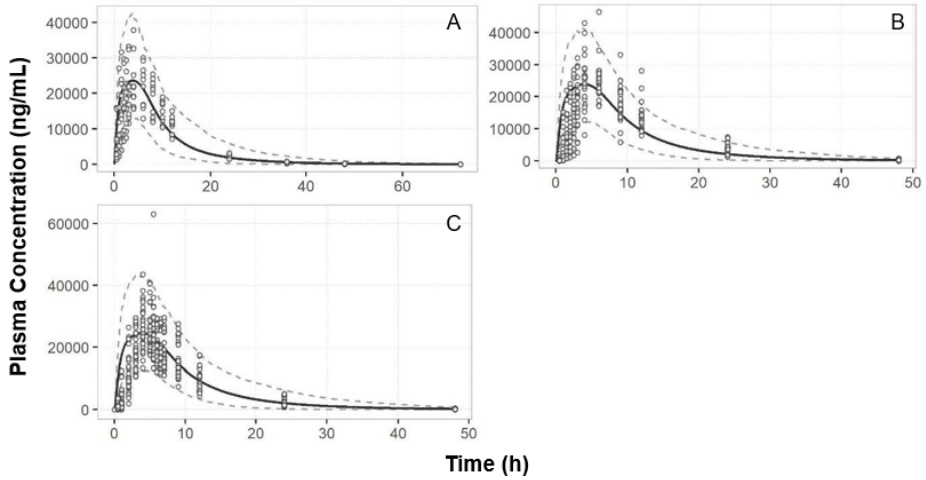
The resulting simulated PK parameters ($AUC_{0-\infty}$, C_{max}, T_{max}) were mostly within 1.25-fold of the observed data but slightly underestimated following 4 g (Table 122). The observed and predicted plasma zoliflodacin concentration-time profiles following administration of a single oral 3 g dose under fed conditions is shown in Figure 11.

Table 121. Summary of Observed and Predicted PK Parameters of Zoliflodacin Following Single Oral Dose in Healthy Participants Under Fed Conditions

Dose	Parameter	Predicted	Observed	Pred/Obs Ratio
1.5 g[a]	$AUC_{0-\infty}$ (h*ng/mL)	143285 (133327-153987)	129000	1.11
	C_{max} (ng/mL)	16426 (15502-17404)	12100	1.36
	t_{max} (h)	2.20 (0.97-6.05)	4.00	0.55
3 g[a,b,c]	$AUC_{0-\infty}$(h*ng/mL)[a]	257741 (242194-274286)	281000	0.92
	C_{max} (ng/mL)[a]	24002 (22603-25488)	24000	1.00
	t_{max} (h)[a]	4.18 (0.94-8.82)	4.00	1.04
	$AUC_{0-\infty}$ (h*ng/mL)[b]	271283 (259921-283141)	343000	0.79
	C_{max} (ng/mL)[b]	24095 (23143-25085)	27100	0.89
	t_{max} (h)[b]	4.28 (0.84-8.76)	6.00	0.71
	$AUC_{0-\infty}$ (h*ng/mL)[c]	270300 (260796-280150)	249000	1.09
	C_{max} (ng/mL)[c]	24702 (23858-25577)	28900	0.85
	t_{max} (h)[c]	4.39 (0.94-10.27)	4.50	0.98
4 g[b]	$AUC_{0-\infty}$ (h*ng/mL)	327937 (313472-343068)	486000	0.67
	C_{max} (ng/mL)	26698 (25570-27875)	37500	0.71
	t_{max} (h)	4.90 (0.82-9.79)	4.00	1.22

Source: Report PC0914-2024-0015 Table 7, Table 9, Table 10, Table 11, and Table 12
Note: Data are presented as geometric mean and 90% confidence interval, except for t_{max} which is expressed as median (minimum, maximum).
[a] Study D4930C00001 Part B
[b] Study STI_Zoli002
[c] Study STI_Zoli003 Cohort 1
Abbreviations: $AUC_{0-\infty}$, area under the concentration-time curve estimated to infinity; C_{max}, maximum plasma concentration; obs, observed; PK, pharmacokinetic; pred, predicted; t_{max}, time to maximum plasma concentration

Figure 10. Predicted and Observed PK Profile of Zoliflodacin After Single Oral Dose of 3 g in Healthy Participants Under Fed Conditions



Source: Report PC0914-2024-0015 Figure 16 (panel D), Figure 17 (panel B), Figure 18 (panel B)
Note: Lines and circles represent simulated and observed data, respectively. Grey lines represent 5th and 95th percentiles and solid black line represents mean data for the simulated population.
Panel A:Study D4930C00001 Part B; panel B: Study STI_Zoli002; panel C: Study STI_Zoli003 Cohort 1
Abbreviation: PK, pharmacokinetic

Q2. Can PBPK Analyses Predict the Effect of a CYP3A Inhibitor on Zoliflodacin Pharmacokinetics?

Yes, the PBPK model is adequate to simulate the effect of a CYP3A inhibitor on a single oral dose of zoliflodacin. The Applicant performed a manual sensitivity analysis to estimate the median fraction metabolized by CYP3A4 (fm_{CYP3A4}) required to recover the observed interaction

of zoliflodacin with strong CYP3A inhibitor itraconazole. The simulated trial design and participant demographics were based on Study STI_Zoli003 (Cohort 2), which included 18 participants (25 to 55 years of age, 16.7% female). Accordingly, 10 virtual trials of 18 healthy participants were simulated. The predicted median fm_{CYP3A4} value was 0.31. Non-CYP3A4 pathways contributing to potential DDIs were not evaluated but the individual-pathway risk is expected to be low, based on in vitro reaction phenotyping study results indicating lesser contributions of other CYP enzymes (e.g., CYP1A2, 2C9, 2C8, and 2C19) to the clearance of zoliflodacin compared to CYP3A4. The derived fm_{CYP3A4} value using the observed DDI effect with itraconazole is acceptable.

The simulated and observed $AUC_{0-\infty}$ and C_{max} values and corresponding ratios following a single oral dose of zoliflodacin 3 g in the absence and presence of itraconazole are listed in Table 122.

Table 122. Summary of Predicted and Observed Zoliflodacin PK Parameters and Ratios in the Absence and Presence of Itraconazole in Healthy Participants Under Fasted Condition

	Zoliflodacin		Zoliflodacin + Itraconazole		Ratio	
Parameter	**Simulated**	**Observed**	**Simulated**	**Observed**	**Simulated**	**Observed**
$AUC_{0-\infty}$ (h*ng/mL)	175599 (165813-185961)	164000	245141 (230532-260676)	226000	1.40 (1.38-1.41)	1.38
C_{max} (ng/mL)	18838 (17565-20203)	17600	22383 (20906-23964)	18100	1.19 (1.18-1.20)	1.03

Source: Report PC0914-2024-0015 Table 6
Data are presented as geometric mean and 90% confidence interval.
Simulation: Ten virtual trials of 18 healthy participants (16.7% female) aged 25 to 55 years receiving a single oral dose of zoliflodacin 3 g in the absence of itraconazole and on the sixth day of 8 days of itraconazole dosing (i.e., 400-mg loading dose with food followed by 200-mg once daily with food through Day 8). The default Simcyp compound library files for itraconazole and its metabolite hydroxy-itraconazole were used.
Abbreviations: $AUC_{0-\infty}$, area under the concentration-time curve estimated to infinity; C_{max}, maximum plasma concentration; PK, pharmacokinetic

The above clinical DDI was evaluated under fasting conditions in Study STI_Zoli003. Therefore, the Applicant was requested to estimate the potential change in AUC due to strong and moderate CYP3A inhibition under fed conditions using the fed model settings described above. In the response to the IR received on July 2, 2025, the Applicant provided the following simulation results. Table 123 shows the summary of simulation scenarios and Table 124 shows the simulated PK parameters and ratios for zoliflodacin in the absence and presence of strong (itraconazole and ritonavir) or moderate (fluconazole) CYP3A4 inhibitors.

Table 123. Summary of Simulation Scenarios

Zoliflodacin Dose	Inhibitor Dose Regimen	Prandial Condition
3 g single dose (Day 6)	Itraconazole 400 mg (Day 1) then 200 mg daily (Days 2 to 8)	Fasted: All doses with food except Day 6 Fed: All doses with food
3 g single dose (Day 14)	Ritonavir 100 mg twice daily (Day 1 to 16)	Fasted: All doses fasted Fed: All doses with food
3 g single dose (Day 14)	Fluconazole 200 mg daily (Day 1 to 16)	Fasted: all doses fasted Fed: All doses with food

Source: Applicant IR response received on July 2, 2025, Table 1

Table 124. Summary of Predicted Zoliflodacin PK Parameters and Ratios in the Absence and Presence of CYP3A4 Inhibitors in Healthy Participants Under Fasted and Fed Conditions

		Zoliflodacin (3 g Single Dose)		Zoliflodacin (3 g Single Dose) + CYP3A4 Inhibitor		Ratio	
Condition	CYP3A4 Inhibitor	$AUC_{0-\infty}$ (h*ng/mL)	C_{max} (ng/mL)	$AUC_{0-\infty}$ (h*ng/mL)	C_{max} (ng/mL)	$AUC_{0-\infty}$	C_{max}
Fasted	Itraconazole (strong)	175599	18838	245141	22383	1.40	1.19
	Ritonavir (strong)	176282	19227	260425	23029	1.48	1.20
	Fluconazole (moderate)	166413	16350	213289	18593	1.28	1.14
Fed	Itraconazole (strong)	273767	25456	380179	30951	1.39	1.22
	Ritonavir (strong)	265734^	24280	390449^	30134	1.47^	1.24
	Fluconazole (moderate)	264440	22941	337686	26583	1.28	1.16

Source: Applicant IR response received on July 2, 2025, Table 2
Note: Data presented as geometric mean
^ AUC_{0-last} reported for ritonavir DDI under fed conditions
Abbreviations: $AUC_{0-\infty}$, area under the concentration-time curve estimated to infinity; AUC_{last}, area under the concentration-time curve to the last measurable concentration; C_{max}, maximum plasma concentration; CYP, cytochrome P 450, PK, pharmacokinetic

The predicted change in zoliflodacin AUC was similar under fasted and fed conditions. In response to FDA's IR, the Applicant indicated that the predicted F_g and F_h values remain high (both approximately 0.9) under both fasted and fed conditions, suggesting low sensitivity of first-pass metabolism to food-induced changes. Thus, the predicted DDI with CYP3A4 inhibitors and inducers is expected to be comparable between the fasted and fed conditions. The reviewer noted that the Applicant's rationale is plausible; however, regulatory experience with the performance of food effect predictions remains limited. Regarding the exposure safety margin, based on clinical experience (e.g., Study STI_Zoli002), the predicted DDI magnitude is not expected to pose a safety concern. In Study STI_Zoli002, the observed exposures showed approximately a 40% increase in both $AUC_{0-\infty}$ and C_{max} following a 4 g dose compared to a 3 g dose. This exposure range was well tolerated under both fasted and fed conditions (Table 120 and Table 121). Therefore, the magnitude of the DDI effect ($AUC_{0-\infty}$ <1.5-fold, C_{max} 1.03-fold) is considered well tolerated. Given the mild food effect, reported safety margin, and single-dose administration of zoliflodacin, the food-DDI prediction is acceptable for this application.

Q3. Can PBPK Analyses Predict the Effect of a CYP3A Inducer on Zoliflodacin Pharmacokinetics?

Yes, the PBPK model is adequate to simulate the effect of repeat dosing of strong or moderate CYP3A inducer on a single oral dose of zoliflodacin 3 g under fasted condition. In the presence of a strong CYP3A inducer (rifampin), zoliflodacin $AUC_{0-\infty}$ and C_{max} are predicted to decrease by 56% and 38%, respectively. In the presence of a moderate CYP3A inducer (efavirenz), zoliflodacin $AUC_{0-\infty}$ and C_{max} are predicted to decrease by 41% and 27%, respectively. The simulated $AUC_{0-\infty}$ and C_{max} values and corresponding ratios following a single oral dose of zoliflodacin 3 g in the absence and presence of rifampin or efavirenz are listed in Table 125. The reviewer noted that the simulation focused on CYP3A-mediated clearance only. Other potential clearance pathways that could be influenced by CYP3A inducers were not fully characterized for zoliflodacin, which may contribute to some degree of uncertainty in the prediction.

Table 125. Summary of Predicted Zoliflodacin PK Parameters and Ratios in the Absence and Presence of Rifampin and Efavirenz in Healthy Participants Under Fasted Condition

Parameter	Zoliflodacin	Zoliflodacin + Rifampin	Ratio
$AUC_{0-\infty}$ (h*ng/mL)	168894 (156844-181869)	73675 (67327-80621)	0.44 (0.42-0.46)
C_{max} (ng/mL)	16606 (15191-18152)	10267 (9278-11361)	0.62 (0.60-0.64)
		Zoliflodacin + Efavirenz	**Ratio**
$AUC_{0-\infty}$ (h*ng/mL)	160585 (150358-171507)	94845 (88343-101825)	0.59 (0.57-0.61)
C_{max} (ng/mL)	16312 (14975-17768)	11963 (10939-13082)	0.73 (0.72-0.75)

Source: Report PC0914-2024-0015 Table 14 and Table 15
Data are presented as geometric mean and 90% confidence interval.
Simulation: Ten virtual trials of 10 healthy participants (50% female) aged 20 to 50 years receiving a single oral dose of zoliflodacin 3 g in the absence of rifampin or efavirenz and on the 14th day of 16 days of dosing of rifampin 600 mg once daily or efavirenz 600 mg once daily were generated. The default Simcyp compound library files for rifampin and efavirenz were used.
Abbreviations: $AUC_{0-\infty}$, area under the concentration-time curve estimated to infinity; C_{max}, maximum plasma concentration; PK, pharmacokinetic

Q4. Can PBPK Analyses Predict the Effect of Zoliflodacin on the pharmacokinetics of Transporter Substrates?

The Applicant simulated the pharmacokinetics of various transporter substrates – dabigatran (P-gp), metformin (MATEs), pravastatin (OATP1B1/1B3), rosuvastatin (BCRP), tenofovir (OAT1), methotrexate (OAT3) – in healthy participants following administration of a single 3 g dose of zoliflodacin under both fasted and fed conditions. The DDI assessment was based on zoliflodacin's in vitro enzyme competitive inhibition constant for unbound drug (Ki,u) (assumed to be equal to IC_{50}) for each transporter. Using the measured Ki,u values, the predicted geometric mean $AUC_{0-\infty}$ ratio in the absence or presence of zoliflodacin was <1.25 for dabigatran (P-gp), metformin (MATEs), tenofovir (OAT1), and methotrexate (OAT3) under fasted and fed states. For pravastatin (OATP1B1/1B3), the predicted geometric mean $AUC_{0-\infty}$ ratio was <2 under fasted and fed conditions. For rosuvastatin (BCRP), the simulated geometric mean $AUC_{0-\infty}$ change was <1.25-fold under fasted but within 1.25 to 2 under fed conditions.

The impact of uncertainty/variability associated with Ki,u on the predicted DDI magnitudes was evaluated by the Applicant by applying a 10-fold reduction (except for 15-fold reduction applied to P-gp's Ki,u). Using these reduced Ki,u values, the predicted geometric mean $AUC_{0-\infty}$ ratios were <1.25 for tenofovir (OAT1), within 1.25 to 2 for dabigatran (P-gp), rosuvastatin (BCRP), metformin (MATEs), and methotrexate (OAT3), and ~2.5 to ~3 for pravastatin (OATP1B1/1B3). The reviewer noted that the 10- and 15-fold uncertainty factors assigned for apical transporter substrates are insufficient to account for in vitro-in vivo extrapolation uncertainty based on the FDA's experience. The reviewer notes that a 15-fold uncertainty factor was used to evaluate the inhibitor potential of an in vitro OAT3 inhibitor in a previous NDA review (NDA 212887: cabotegravir).

However, due to zoliflodacin's single dose administration (and short $t_{1/2}$), the review team considers overall DDI risk is low, and no dose adjustments have been proposed when zoliflodacin is coadministered with P-gp, MATEs, OAT1/3, OATP1B1/3, or BCRP substrates.

Q5. Can PBPK Analyses Predict the Effect of Hepatic Impairment on Zoliflodacin Pharmacokinetics?

The Applicant simulated the pharmacokinetics of a single oral dose of zoliflodacin 3 g in healthy participants and participants with mild, moderate, and severe hepatic impairment. These

simulations were not reviewed due to limited relevancy; hepatic impairment is not considered a significant clinical risk for this single dose administration drug.

14.6. Pharmacogenetics

No pharmacogenetic assessments of zoliflodacin were conducted.

15. Study/Trial Design

Please refer to Section 6 for the study designs of the phase 3 trial (STI_Zoli001) and the phase 2 trial (DMID 14-0014) for treatment of uncomplicated urogenital gonorrhea.

Six completed phase 1 clinical studies provided additional supportive evidence of the safety of zoliflodacin in healthy adult participants. Please refer to section 17 for the clinical safety findings in those studies.

16. Efficacy

Trial DMID 14-0014 Primary Efficacy Endpoint Subgroup Analyses

Table 126. Proportion of Participants With Microbiological Cure at TOC, Micro-ITT Population, Trial DMID 14-0014

Demographic Parameters	Zoliflodacin 2000 mg N=57	Zoliflodacin 3000 mg N=56	Ceftriaxone 500 mg N=28
Overall	55/57 (96.5)	54/56 (96.4)	28 (100)
Sex, n (%)			
Female	0	7/8 (87.5)	1/1 (100)
Male	55/57 (96.5)	47/48 (97.9)	27/27 (100)
Race, n (%)			
Black or African American	34/36 (94.4)	34/35 (97.1)	18/18 (100)
White	18/18 (100)	15/15 (100)	8/8 (100)
Other	3/3 (100)	5/6 (83.3)	2/2 (100)
Ethnicity, n (%)			
Hispanic	4/4 (100)	2/3 (66.7)	3/3 (100)
Not-Hispanic	51/53 (96.2)	52/53 (98.1)	25/25 (100)

Source: FDA analysis; adsl.xpt
Abbreviations: Micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants in specified population or group; SD, standard deviation; TOC, test-of-cure

Trial STI_Zoli001 Baseline Demographics in the Primary Efficacy Analysis Population (Micro-ITT (Urogenital) Population)

Table 127. Baseline Demographics and Clinical Characteristics, Micro-ITT (Urogenital) Population, Trial STI_Zoli001

Characteristic	Zoliflodacin N=506	Ceftriaxone-Azithromycin N=238
Sex assigned at birth, n (%)		
Female	50 (9.9)	18 (7.6)
Male	456 (90.1)	220 (92.4)
Gender, n (%)		
Female	51 (10.1)	19 (8.0)
Male	455 (89.9)	219 (92.0)
Age, years		
Mean (SD)	30.3 (9.72)	29.2 (9.41)
Median	28.0	26.0
Min, max	16.0, 73.0	16.0, 67.0
Age group (years), n (%)		
<18	9 (1.8)	1 (<1)
≥18 to <65	2 (<1)	1 (<1)
≥65	495 (97.8)	236 (99.2)
Race, n (%)		
Black or African American	278 (54.9)	128 (53.8)
Asian	161 (31.8)	67 (28.2)
White	57 (11.3)	38 (16.0)
American Indian or Alaska Native Asian	6 (1.2)	1 (<1)
Native Hawaiian or Other Pacific Islander	1 (<1)	1 (<1)
Multiple	1 (<1)	2 (<1)
Other	2 (<1)	1 (<1)
Ethnicity, n (%)		
Hispanic	15 (3.0)	13 (5.5)
Not-Hispanic	491 (97.0)	225 (94.5)
Baseline Height (cm)		
Mean (SD)	172.7 (8.36)	172.4 (8.05)
Median	173.0	172.0
Min, max	150.0, 199.0	150.0, 196.5
Missing	1	0
Baseline weight (kg)		
Mean (SD)	70.5 (15.44)	68.8 (12.77)
Median	68.0	67.6
Min, max	36.5, 138.6	44.6, 119.0
Missing	2	0
BMI (kg/m^2)		
Mean (SD)	23.6 (4.56)	23.2 (4.24)
Median	22.8	22.5
Min, max	13.4, 46.0	15.0, 41.8
Missing	2	0
Region, n (%)		
Asia	149 (29.4)	64 (26.9)
Europe	45 (8.9)	17 (7.1)
South Africa	217 (42.9)	113 (47.5)
United States	95 (18.8)	44 (18.5)

Characteristic	Zoliflodacin N=506	Ceftriaxone-Azithromycin N=238
Country of participation, n (%)		
Belgium	3 (<1)	1 (<1)
Netherlands	42 (8.3)	16 (6.7)
Thailand	149 (29.4)	64 (26.9)
United States	95 (18.8)	44 (18.5)
South Africa	217 (42.9)	113 (47.5)
History of sexually transmitted infection(s), n (%)		
Yes	284 (56.1)	130 (54.6)
No	222 (43.9)	108 (45.4)
HIV status, n (%)		
Negative	27 (5.3)	6 (2.5)
Positive	374 (73.9)	185 (77.7)
Missing	105 (20.8)	47 (19.7)

Source: FDA Analysis; adsl.xpt; excluding participants from site 710-005
Abbreviations: Micro-ITT, microbiological intent-to-treat; N, number of participants in treatment arm; n, number of participants with characteristic; SD, standard deviation

17. Clinical Safety

17.1. Phase 1 Trials

A total of 250 (238 zoliflodacin, 12 placebo) participants were enrolled in six phase 1 clinical studies to assess the safety, tolerability and pharmacokinetic properties of zoliflodacin. Study participants were healthy adults between the ages of 18 to 55 years old. Oral dosages ranged from 200 mg to 4000 mg. Most adverse events observed during the phase 1 clinical studies were nervous system disorders. There were no SAEs and no deaths reported in the phase 1 clinical studies.

Table 128. Phase 1 Studies of Zoliflodacin in Healthy Humans

Study ID	Safety Population	Zoliflodacin Dosing Regimen(s)	Safety Results
D4930C00001	Healthy participants N=66 (Zoliflodacin =54, placebo =12)	Part A: Escalating PO dose 200 mg, 400 mg, 800 mg, 1600 mg, 3000 mg and 4000 mg orally QD Part B: Single PO dose of either 1500 mg or 3000 mg under fasted or fed conditions	Most common type of AEs reported were nervous system disorders (dysgeusia and headache). All AEs were of mild intensity. SAEs: No SAEs Deaths: No deaths Discontinuation: None
D4930C00003	Healthy participants N=6	Single dose of 3000 mg of powder for oral suspension.	There was one AE reported for back pain, contact dermatitis and vessel puncture site bruise. All AEs were of mild intensity. SAEs: No SAEs Deaths: No deaths Discontinuation: None
DMID 16-0110	Healthy participants N=72	Single dose of 2 g or 4 g PO	Most common type of AEs reported were cardiac and vascular disorders (heart rate decrease) and nervous system disorders (headache). Most AEs were mild or moderate intensity. Two participants had severe AEs (bacteriuria, QTc interval prolonged) SAEs: No SAEs Deaths: No deaths Discontinuation: None
DMID 16-0118	Healthy participants N=8	Single 4000 mg oral dose	Most common type of AEs reported were cardiac and vascular disorders (heart rate decreased and QT prolonged) Most AEs were of mild or moderate intensity. One AE was severe (decreased heart rate). SAEs: No SAEs Deaths: No deaths Discontinuation: None

Study ID	Safety Population	Zoliflodacin Dosing Regimen(s)	Safety Results
STI_Zoli002	Healthy participants N=48	Single oral 3000 mg dose or single oral 4000 mg dose administered as granules for oral suspension under fasted and fed conditions	Most common type of AEs reported were nervous system disorders (headache). All AEs were mild intensity SAEs: No SAEs TEAE: One participant had abnormally clinically significant 12-lead ECG findings Deaths: No deaths Discontinuation: One participant removed themselves from the study
STI_Zoli003	Healthy participants N=50	Single dose of 3000 mg of either granules for oral suspension or powder for oral suspension	Most common type of AEs reported were nervous system disorders (headache) and gastrointestinal disorders (nausea). All AEs were of mild or moderate intensity. SAEs: No SAEs Deaths: No deaths Discontinuation: One participant with rash

Source: Reviewer table
Abbreviations: AE, adverse event; ECG, electrocardiogram; N, number of participants in safety population; SAE, serious adverse event; PO, by mouth; QTc, corrected QT interval; TEAE, treatment-emergent adverse event

Participants who Discontinued Treatment or Had TEAEs

Study STI_Zoli002

- Participant (b) (6) was a 36-year-old White Hispanic male who withdrew consent prior to the second dosing period (i.e., was not dosed in the fed state). No explanation was provided by the participant as to why he removed his consent. Study treatment was discontinued.
- Participant (b) (6) was a 23-year-old White, Hispanic male who had an abnormal and clinically significant 12-lead ECG finding at 3 hours post zoliflodacin dose in the second dosing period, which was reported as a TEAE of Wolff-Parkinson-White (WPW) syndrome. This condition was not detected at screening and the participant was asymptomatic. WPW syndrome is a congenital condition that involves an accessary electrical pathway in the heart and leads to a characteristic ECG pattern. The pattern on ECG can be intermittent and may not have been captured during the screening ECG. The Applicant considered the finding resolved and assessed it as related to study treatment as it was not detected at screening. This reviewer does not agree that this finding was related to the study drug as WPW is a congenital condition with intermittent presentation which may not have been manifest on the ECG at screening but which would be highly unlikely to develop after a single exposure to zoliflodacin.

Study STI_Zoli003

Participant (b) (6) was a 27-year-old female with no reported medical history, except a mild COVID-19 infection 3 months prior to screening, who was discontinued due to mild treatment associated adverse event of rash on trunk and arms. This rash began following the administration of study treatment on Day 1. The participant received treatment with oral paracetamol and transdermal dimetindene maleate (antihistamine). On Day 2, laboratory results showed an increase in leukocyte and neutrophil counts. Eosinophilic granulocytes were normal. An infection was suspected; however, no source was identified. There was significant improvement of the rash on Day 3 and the participant was discontinued from the study treatment. The rash resolved on Day 5. There was a follow-up visit on Day 8 and laboratory studies were checked which showed normalization of leukocyte and neutrophil counts.

Trial STI_Zoli001

Table 129 lists all TEAEs that occurred by SOC and Preferred Term in Trial STI_Zoli001.

Table 129. Participants With Adverse Events by System Organ Class and Preferred Term, Safety Population, Trial STI_Zoli001

System Organ Class Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone-Azithromycin N=308 n (%)
Any AE	286 (46.2)	143 (46.4)
Blood and lymphatic system disorders (SOC)	49 (7.9)	27 (8.8)
Neutropenia	42 (6.8)	24 (7.8)
Leukopenia	24 (3.9)	7 (2.3)
Lymphadenopathy	2 (0.3)	1 (0.3)
Eosinophilia	1 (0.2)	1 (0.3)
Thrombocytopenia	1 (0.2)	1 (0.3)
Lymphopenia	0	3 (1.0)
Cardiac disorders (SOC)	1 (0.2)	0
Palpitations	1 (0.2)	0
Endocrine disorders (SOC)	1 (0.2)	0
Hyperthyroidism	1 (0.2)	0
Eye disorders (SOC)	1 (0.2)	0
Swelling of eyelid	1 (0.2)	0
Gastrointestinal disorders (SOC)	54 (8.7)	39 (12.7)
Nausea	16 (2.6)	12 (3.9)
Diarrhoea	15 (2.4)	22 (7.1)
Abdominal pain	6 (1.0)	1 (0.3)
Anal pruritus	3 (0.5)	1 (0.3)
Flatulence	3 (0.5)	3 (1.0)
Toothache	3 (0.5)	0
Vomiting	3 (0.5)	1 (0.3)
Abdominal pain lower	2 (0.3)	0
Haemorrhoids	2 (0.3)	0
Abdominal distension	1 (0.2)	0
Abdominal pain upper	1 (0.2)	2 (0.6)
Constipation	1 (0.2)	2 (0.6)
Dental caries	1 (0.2)	0
Dyschezia	1 (0.2)	0

System Organ Class Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone-Azithromycin N=308 n (%)
Gastritis	1 (0.2)	0
Odynophagia	1 (0.2)	0
Proctalgia	1 (0.2)	0
Anal rash	0	1 (0.3)
Anorectal discomfort	0	1 (0.3)
Aphthous ulcer	0	1 (0.3)
Dyspepsia	0	1 (0.3)
General disorders and administration site conditions (SOC)	29 (4.7)	49 (15.9)
Malaise	8 (1.3)	5 (1.6)
Pyrexia	6 (1.0)	4 (1.3)
Fatigue	5 (0.8)	4 (1.3)
Injection site pain	5 (0.8)	38 (12.3)
Vaccination site pain	4 (0.6)	0
Asthenia	1 (0.2)	0
Catheter site pain	1 (0.2)	0
Chills	1 (0.2)	0
Feeling hot	1 (0.2)	1 (0.3)
Influenza like illness	0	2 (0.6)
Pain	0	2 (0.6)
Hepatobiliary disorders (SOC)	1 (0.2)	1 (0.3)
Alcoholic liver disease	1 (0.2)	0
Hyperbilirubinaemia	0	1 (0.3)
Infections and infestations (SOC)	48 (7.8)	12 (3.9)
COVID-19	4 (0.6)	0
Upper respiratory tract infection	4 (0.6)	0
Vulvovaginal candidiasis	4 (0.6)	1 (0.3)
Urethritis chlamydial	3 (0.5)	0
Chlamydial infection	2 (0.3)	0
Conjunctivitis	2 (0.3)	0
Gonorrhoea	2 (0.3)	0
Nasopharyngitis	2 (0.3)	2 (0.6)
Urethral discharge syndrome	2 (0.3)	0
Urethritis gonococcal	2 (0.3)	0
Bacterial vaginosis	1 (0.2)	1 (0.3)
Balanitis candida	1 (0.2)	0
Bartholin's abscess	1 (0.2)	0
Candida infection	1 (0.2)	0
Cellulitis	1 (0.2)	0
Ear infection	1 (0.2)	0
Fungal skin infection	1 (0.2)	0
Genital herpes	1 (0.2)	0
Genital infection fungal	1 (0.2)	0
Genital ulcer syndrome	1 (0.2)	0
Genitourinary tract gonococcal infection	1 (0.2)	0

System Organ Class Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone-Azithromycin N=308 n (%)
Hepatitis C	1 (0.2)	0
Herpes simplex	1 (0.2)	0
HIV infection	1 (0.2)	0
Molluscum contagiosum	1 (0.2)	1 (0.3)
Monkeypox	1 (0.2)	0
Oropharyngeal gonococcal infection	1 (0.2)	0
Penile abscess	1 (0.2)	0
Pharyngitis	1 (0.2)	0
Pharyngotonsillitis	1 (0.2)	0
Proctitis gonococcal	1 (0.2)	0
Syphilis	1 (0.2)	0
Tinea cruris	1 (0.2)	0
Tinea infection	1 (0.2)	0
Tinea versicolour	1 (0.2)	0
Urethritis trichomonal	1 (0.2)	0
Urinary tract infection	1 (0.2)	0
Body tinea	0	1 (0.3)
Fungal infection	0	1 (0.3)
Gastroenteritis	0	1 (0.3)
Influenza	0	1 (0.3)
Orchitis	0	1 (0.3)
Otitis externa	0	1 (0.3)
Tonsillitis	0	1 (0.3)
Injury, poisoning and procedural complications (SOC)	9 (1.5)	6 (1.9)
Immunisation reaction	2 (0.3)	1 (0.3)
Animal bite	1 (0.2)	0
Arthropod bite	1 (0.2)	0
Contusion	1 (0.2)	0
Muscle strain	1 (0.2)	0
Penis injury	1 (0.2)	1 (0.3)
Road traffic accident	1 (0.2)	0
Skin injury	1 (0.2)	0
Soft tissue injury	1 (0.2)	0
Adverse event following immunisation	0	1 (0.3)
Head injury	0	1 (0.3)
Skin abrasion	0	1 (0.3)
Skin laceration	0	1 (0.3)

System Organ Class Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone-Azithromycin N=308 n (%)
Investigations (SOC)	57 (9.2)	25 (8.1)
Neutrophil count decreased	21 (3.4)	15 (4.9)
Alanine aminotransferase increased	12 (1.9)	5 (1.6)
Blood bilirubin increased	6 (1.0)	1 (0.3)
White blood cell count decreased	6 (1.0)	2 (0.6)
Blood creatinine increased	5 (0.8)	3 (1.0)
Glomerular filtration rate decreased	5 (0.8)	1 (0.3)
Haemoglobin decreased	3 (0.5)	0
Lymphocyte count decreased	3 (0.5)	0
Bilirubin conjugated increased	2 (0.3)	0
Platelet count decreased	2 (0.3)	1 (0.3)
Blood pressure systolic increased	1 (0.2)	1 (0.3)
Creatinine renal clearance decreased	1 (0.2)	0
Hepatic enzyme increased	1 (0.2)	0
Monocyte count decreased	1 (0.2)	0
Platelet count increased	1 (0.2)	0
Blood pressure increased	0	1 (0.3)
Metabolism and nutrition disorders (SOC)	0	5 (1.6)
Abnormal loss of weight	0	3 (1.0)
Decreased appetite	0	1 (0.3)
Hypocalcaemia	0	1 (0.3)
Musculoskeletal and connective tissue disorders (SOC)	11 (1.8)	5 (1.6)
Back pain	3 (0.5)	2 (0.6)
Arthralgia	2 (0.3)	0
Myalgia	2 (0.3)	1 (0.3)
Pain in extremity	2 (0.3)	1 (0.3)
Groin pain	1 (0.2)	1 (0.3)
Neck pain	1 (0.2)	0
Neoplasms benign, malignant and unspecified (incl cysts and polyps) (SOC)	2 (0.3)	0
Anogenital warts	2 (0.3)	0
Nervous system disorders (SOC)	80 (12.9)	23 (7.5)
Headache	61 (9.9)	14 (4.5)
Dizziness	21 (3.4)	5 (1.6)
Hypersomnia	2 (0.3)	1 (0.3)
Hypoaesthesia	1 (0.2)	0
Presyncope	1 (0.2)	1 (0.3)
Somnolence	1 (0.2)	1 (0.3)
Tension headache	0	1 (0.3)
Psychiatric disorders (SOC)	1 (0.2)	6 (1.9)
Insomnia	1 (0.2)	4 (1.3)
Borderline personality disorder	0	1 (0.3)
Depression	0	1 (0.3)
Intentional self-injury	0	1 (0.3)
Major depression	0	1 (0.3)
Panic attack	0	1 (0.3)

System Organ Class Preferred Term	Zoliflodacin N=619 n (%)	Ceftriaxone-Azithromycin N=308 n (%)
Renal and urinary disorders (SOC)	9 (1.5)	2 (0.6)
Haematuria	2 (0.3)	0
Urethral discharge	2 (0.3)	0
Dysuria	1 (0.2)	0
Renal impairment	1 (0.2)	0
Urethral pain	1 (0.2)	1 (0.3)
Urethritis noninfective	1 (0.2)	1 (0.3)
Urinary incontinence	1 (0.2)	0
Reproductive system and breast disorders (SOC)	8 (1.3)	3 (1.0)
Pruritus genital	2 (0.3)	0
Vaginal haemorrhage	2 (0.3)	0
Balanoposthitis	1 (0.2)	0
Menstruation irregular	1 (0.2)	0
Scrotal pain	1 (0.2)	0
Testicular pain	1 (0.2)	0
Dysmenorrhoea	0	1 (0.3)
Penile erythema	0	1 (0.3)
Vulvovaginal pruritus	0	1 (0.3)
Respiratory, thoracic and mediastinal disorders (SOC)	14 (2.3)	9 (2.9)
Oropharyngeal pain	6 (1.0)	1 (0.3)
Nasal congestion	4 (0.6)	1 (0.3)
Rhinorrhoea	3 (0.5)	3 (1.0)
Cough	1 (0.2)	3 (1.0)
Sneezing	0	1 (0.3)
Tonsillar hypertrophy	0	2 (0.6)
Skin and subcutaneous tissue disorders (SOC)	10 (1.6)	2 (0.6)
Pruritus	3 (0.5)	1 (0.3)
Alopecia	1 (0.2)	0
Dermatitis allergic	1 (0.2)	0
Dry skin	1 (0.2)	0
Macule	1 (0.2)	0
Night sweats	1 (0.2)	1 (0.3)
Post inflammatory pigmentation change	1 (0.2)	0
Rash	1 (0.2)	0
Social circumstances (SOC)	0	1 (0.3)
Victim of crime	0	1 (0.3)
Surgical and medical procedures (SOC)	0	1 (0.3)
Dental operation	0	1 (0.3)
Vascular disorders (SOC)	1 (0.2)	2 (0.6)
Hot flush	1 (0.2)	0
Hypertension	0	1 (0.3)
Hypotension	0	1 (0.3)

Source: adae.xpt; Software: R
Treatment-emergent adverse events defined as an AE that has a start date on or after the first dose date of study drug, or it has a start date before the date of the first dose date of study drug but worsens in severity on or after the first dose date of Duration of treatment is 1 day (single dose). Study duration is approximately 30 days including follow-up.
Abbreviations: AE, adverse event; N, number of participants in treatment arm; n, number of participants with adverse event; SOC, system organ class

18. Clinical Virology

Not applicable.

19. Clinical Microbiology

19.1. Nonclinical Microbiology

19.1.1. Antimicrobial Spectrum of Activity

The Applicant is seeking labeling and susceptibility interpretive criteria for *N. gonorrhoeae*. This review will focus on the activity of zoliflodacin against *N. gonorrhoeae*.

The in vitro activity of zoliflodacin was determined against clinical isolates of *N. gonorrhoeae* collected in the United States between 2020 and 2023,[36] in previous years [2012 to 2013][37] and in global surveillance studies.[38,39,40,41,42,43] Isolates were collected mostly from males presenting with urethral discharge at sexually transmitted infection clinics and from their partners across the United States as well as global centers. The MIC was determined by agar dilution method following Clinical Laboratory and Standards Institute (CLSI) guidelines.

Figure 12 and Table 130 show the in vitro activity of zoliflodacin against the following *N. gonorrhoeae* clinical isolates:

[36] Studies PC0914-2024-0002, PC0914-2024-0003, PC0914-2024-0010, PC0914-2024-0011, PC0914-2024-0014

[37] Papp, JR, K Lawrence, S Sharpe, J Mueller, and RD Kirkcaldy, 2016, In vitro growth of multidrug-resistant *Neisseria gonorrhoeae* isolates is inhibited by ETX0914, a novel spiropyrimidinetrione, Int J Antimicrob Agents, 48(3):328-330, https://www.ncbi.nlm.nih.gov/pubmed/27499432.

[38] Jacobsson, S, D Golparian, RA Alm, M Huband, J Mueller, JS Jensen, M Ohnishi, and M Unemo, 2014, High in vitro activity of the novel spiropyrimidinetrione AZD0914, a DNA gyrase inhibitor, against multidrug-resistant *Neisseria gonorrhoeae* isolates suggests a new effective option for oral treatment of gonorrhea, Antimicrob Agents Chemother, 58(9):5585-5588, https://www.ncbi.nlm.nih.gov/pubmed/24982070.

[39] Unemo, M, J Ringlander, C Wiggins, H Fredlund, S Jacobsson, and M Cole, 2015, High in vitro susceptibility to the novel spiropyrimidinetrione ETX0914 (AZD0914) among 873 contemporary clinical *Neisseria gonorrhoeae* isolates from 21 European countries from 2012 to 2014, Antimicrob Agents Chemother, 59(9):5220-5225, https://www.ncbi.nlm.nih.gov/pubmed/26077246.

[40] Su, XH, BX Wang, WJ Le, YR Liu, C Wan, S Li, RA Alm, JP Mueller, and PA Rice, 2016, Multidrug-Resistant *Neisseria gonorrhoeae* Isolates from Nanjing, China, Are Sensitive to Killing by a Novel DNA Gyrase Inhibitor, ETX0914 (AZD0914), Antimicrob Agents Chemother, 60(1):621-623, https://www.ncbi.nlm.nih.gov/pubmed/26482313.

[41] Unemo, M, J Ahlstrand, L Sanchez-Buso, M Day, D Aanensen, D Golparian, S Jacobsson, MJ Cole, and European Collaborative Group, 2021, High susceptibility to zoliflodacin and conserved target (GyrB) for zoliflodacin among 1209 consecutive clinical *Neisseria gonorrhoeae* isolates from 25 European countries, 2018, J Antimicrob Chemother, 76(5):1221-1228, https://www.ncbi.nlm.nih.gov/pubmed/33564854.

[42] Le, W, X Su, X Lou, X Li, X Gong, B Wang, CA Genco, JP Mueller, and PA Rice, 2021, Susceptibility Trends of Zoliflodacin against Multidrug-Resistant *Neisseria gonorrhoeae* Clinical Isolates in Nanjing, China, 2014 to 2018, Antimicrob Agents Chemother, 65(3), https://www.ncbi.nlm.nih.gov/pubmed/33318010.

[43] Study AZD0914-M2-021

The zoliflodacin MIC values against *N. gonorrhoeae* ranged from ≤0.002 to 0.25 mg/L with $MIC_{50/90}$ values of 0.06 mg/L and 0.12 mg/L, respectively. Zoliflodacin inhibited 97.0% of isolates at a concentration of 0.12 mg/L and 100% of isolates at a concentration of 0.5 mg/L.

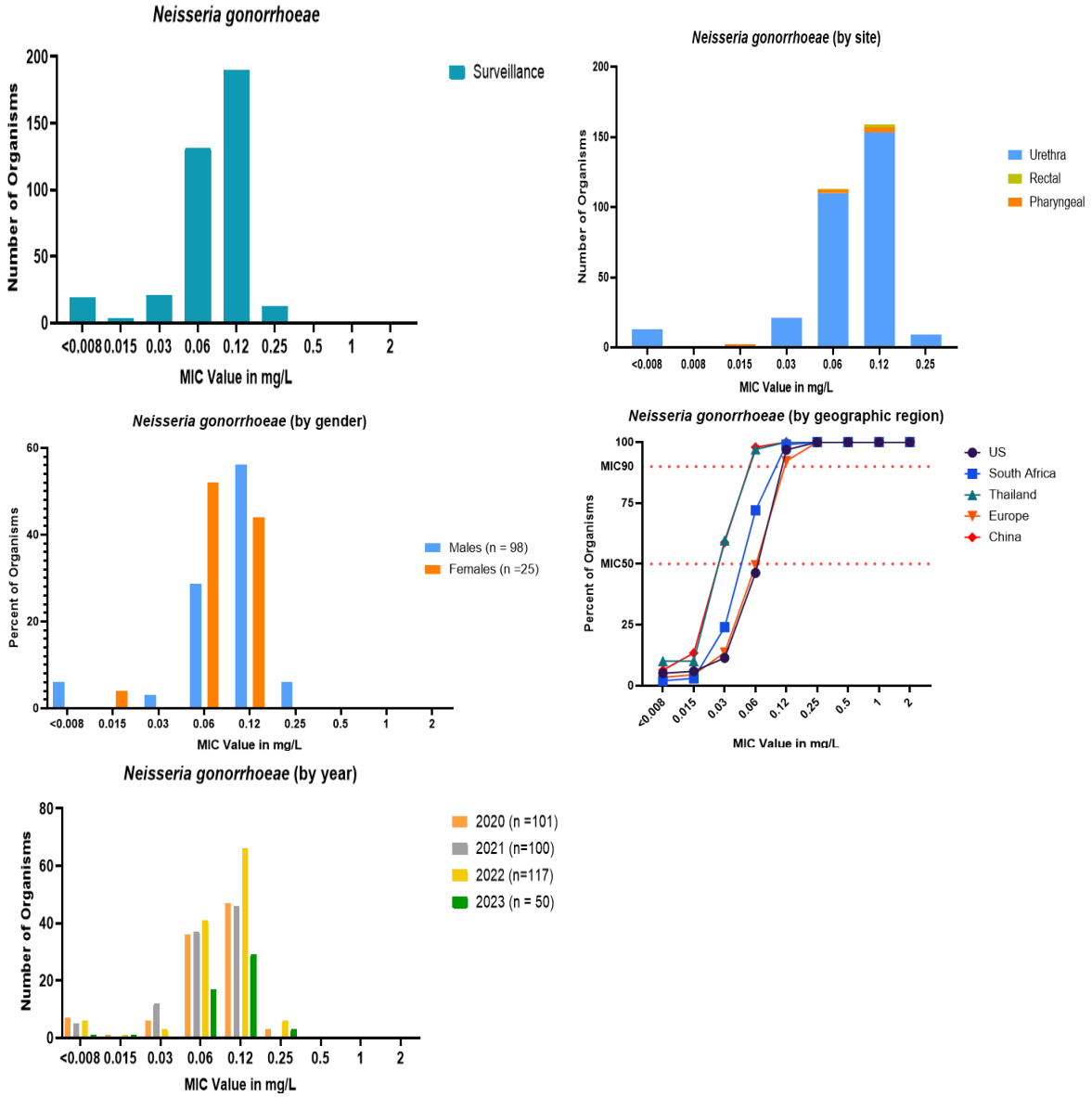
There were no differences in zoliflodacin MIC values in isolates obtained from male or female participants, from participants of different ages or from different anatomic sites of infection.

Zoliflodacin MIC values did not vary by geographic region showing MIC values ≤0.25 mg/L in >99% of isolates. The zoliflodacin MIC_{90} values ranged from 0.125 mg/L (Asia and in 71% of European countries) and 0.25 mg/L (29% of European countries). The highest zoliflodacin MIC result from these studies was 0.5 mg/L which was only seen in two studies from Europe. Isolates obtained from the United States showed similar zoliflodacin MIC_{90} values as global studies (0.125 mg/L) with MICs ranging from ≤0.008 to 0.25 mg/L.

Zoliflodacin MIC_{90} values (0.125 mg/L) remained stable globally over time for the period 2020 through 2023 with 97% of the isolates inhibited at zoliflodacin MICs ≤0.25 mg/L. These results were consistent with surveillance studies from previous years (2012 to 2013) and in recent years (2020 to 2023).

Many of the studies included isolates that were resistant to penicillin, cefixime, ceftriaxone, azithromycin, tetracyclines and fluoroquinolones. Resistance to these antibacterials did not appear to alter the activity of zoliflodacin.

Figure 11. Activity of Zoliflodacin Against *Neisseria gonorrhoeae* Clinical Isolates in Global Surveillance Studies



Source: PC0914-2024-0002, PC0914-2024-0003, PC0914-2024-0010, PC0914-2024-0011, PC0914-2024-0014
Abbreviations: MIC, minimum inhibitory concentration, n, number of participants

Table 130. Activity of Zoliflodacin Against *Neisseria gonorrhoeae* in Global Surveillance Studies

		Minimum Inhibitory Concentration (MIC in mg/L)			
Neisseria Gonorrhoeae	**Number Tested**	**MIC_{50}**	**MIC_{90}**	**Range**	**Reference**
Publications/region					
Global	250	0.125	0.25	0.004-0.25	Jacobsson et al., 2014[44]
Europe	873	0.06-0.125	0.125-0.25	≤0.002-0.25	Unemo et al., 2015[45]
China	187	0.03	0.06	≤0.002-0.125	Su et al., 2016[46]
USA	100	0.06	0.125	0.008-0.25	Papp et al., 2016[47]
Thailand & South Africa	199	0.06	0.125	0.004-0.25	Jacobsson et al., 2019[48]
Europe	1209	0.125	0.125	≤0.004-0.5	Unemo et al., 2021[49]
China	986	0.06	0.125	≤0.002-0.25	Le et al., 2021[50]
United States	100	0.03	0.06	≤0.004-0.06	AZD0914-M2-021
By year (United States)					
2020	100	0.06	0.12	≤0.008-0.25	PC0914-2024-0010
2021	100	0.06	0.12	≤0.008-0.25	PC0914-2024-0011
2022	123	0.12	0.12	≤0.008-0.25	PC0914-2024-0014
2023	53	0.12	0.12	0.008-0.25	PC0914-2024-0002
Gender					
Male	98	0.12	0.12	≤0.008-0.25	PC0914-2024-0014
Female	25	0.12	0.12	0.016-0.125	PC0914-2024-0014

Source: Reviewer-generated table
Abbreviation: MIC, minimum inhibitory concentration

Time-kill studies showed a dose-dependent bactericidal killing (≥3 $\log_{10}$ killing) at 8× to 16× MIC against *N. gonorrhoeae* isolates, with slower rates of killing observed at 2× and 4× MIC. Against first step zoliflodacin *N. gonorrhoeae gyrB* mutants, bactericidal killing at 16× MIC was observed, albeit at a slower rate compared to nonmutants.[51]

19.1.2. Interaction With Other Antimicrobials

No significant in vitro antimicrobial interactions were observed between zoliflodacin and ceftriaxone, cefixime, spectinomycin, tetracycline or gentamicin against wild-type and first-step mutants *N. gonorrhoeae* isolates. The fractional inhibitory concentration index values ranged

[44] See Footnote 24

[45] See Footnote 25

[46] See Footnote 26

[47] See Footnote 23

[48] Jacobsson, S, R Kularatne, R Kittiyaowamarn, V Maseko, P Paopang, P Sangprasert, P Sirivongrangson, L Piddock, T Wi, E Alirol, and M Unemo, 2019, High in vitro susceptibility to the first-in-class spiropyrimidinetrione zoliflodacin among consecutive clinical *Neisseria gonorrhoeae* isolates from Thailand (2018) and South Africa (2015-2017), Antimicrob Agents Chemother, 63(12), https://www.ncbi.nlm.nih.gov/pubmed/31548184.

[49] See Footnote 27

[50] See Footnote 28

[51] Foerster, S, D Golparian, S Jacobsson, LJ Hathaway, N Low, WM Shafer, CL Althaus, and M Unemo, 2015, Genetic Resistance Determinants, In Vitro Time-Kill Curve Analysis and Pharmacodynamic Functions for the Novel Topoisomerase II Inhibitor ETX0914 (AZD0914) in *Neisseria gonorrhoeae*, Front Microbiol, 6:1377, https://www.ncbi.nlm.nih.gov/pubmed/26696986.

from >0.5 to 4 indicating indifference. There were two *N. gonorrhoeae* isolates which showed synergy of zoliflodacin with ciprofloxacin.[52,53]

19.1.3. Susceptibility Testing Methods

The MIC values of zoliflodacin were determined by the agar dilution method in accordance with CLSI guidelines (M07-A11) [2018]. A multicenter Tier 2 QC study was conducted in accordance with CLSI guidelines (M23) and performed at eight independent laboratories against the *N. gonorrhoeae* QC strain ATCC 49226, which showed that the MICs fell within the range of 0.06 to 0.5 mg/L.[54,55] These criteria have been published by CLSI in M100-Ed30 (2020).

19.1.4. Animal Models of Infection

There were no animal studies that evaluated the activity of zoliflodacin against *N. gonorrhoeae* isolates. *Neisseria gonorrhoeae* is a human specific pathogen that grows poorly in nonhumans. As such, animal models (mouse, rabbit, guinea pig, hamster, chimpanzees etc.) are unreliable models of *N. gonorrhoeae* infection.

19.1.5. Pharmacokinetics and Pharmacodynamics

Based on studies conducted in the in vitro HFIM, the Applicant proposed that the PK-PD driver best associated with antibacterial activity is the ratio of the area under concentration curve to the MIC (AUC/MIC) for zoliflodacin. Dose ranging and dose fractionation studies in HFIM showed that single doses ranging from 2 g to 8 g successfully eradicated *N. gonorrhoeae* reference strains WHO-F and WHO-X. Strain WHO-F is susceptible to all relevant antimicrobials with zoliflodacin MIC of 0.06 mg/L. Strain WHO-X is extensively drug-resistant, including resistance to ceftriaxone with zoliflodacin MIC of 0.12 mg/L. Maximal killing and resistance suppression were observed at the *f*$AUC_{0-\infty}$/MIC ratio of 70.6.[56] The PK-PD target attainment analyses showed that against *N. gonorrhoeae* isolates with zoliflodacin MIC values ≤0.25 mg/L, plasma *f*$AUC_{0-\infty}$/MIC ratio target of 70.6 will be achieved in ≥90% participants weighing ≥50 kg receiving a 3 g oral zoliflodacin dose under fed conditions and in participants weighing ≥35 kg to <50 kg receiving a 3 g oral zoliflodacin dose under fasted conditions (for further information see Section 5).

[52] See Footnote 27

[53] Foerster, S, G Drusano, D Golparian, M Neely, LJV Piddock, E Alirol, and M Unemo, 2019, In vitro antimicrobial combination testing of and evolution of resistance to the first-in-class spiropyrimidinetrione zoliflodacin combined with six therapeutically relevant antimicrobials for *Neisseria gonorrhoeae*, J Antimicrob Chemother, 74(12):3521-3529, https://www.ncbi.nlm.nih.gov/pubmed/31730160.

[54] Miller, AA, MM Traczewski, MD Huband, PA Bradford, and JP Mueller, 2019, Determination of MIC Quality Control Ranges for the Novel Gyrase Inhibitor Zoliflodacin, J Clin Microbiol, 57(9), https://www.ncbi.nlm.nih.gov/pubmed/31315953.

[55] Study AZD0914-M2-032

[56] Study PC0914-20254-0001

19.2. Microbiology Data From Clinical Studies

The phase 3 clinical trial (STI_Zoli001) evaluated the efficacy and safety of a single 3 g dose of zoliflodacin compared to a single IM 500 mg dose of ceftriaxone in combination with a single oral 1 g dose of azithromycin in ≥12-year-old adult and pediatric participants with clinical evidence or clinical suspicion of uncomplicated urogenital gonorrhea. The phase 3 trial design and results are described elsewhere in the review (see Section 6). Of the 930 participants randomized to study treatment, 744 participants were included in the microbiological modified intent-to-treat (micro-MITT) population. The micro-MITT population included all participants who had a positive *N. gonorrhoeae* culture from the relevant anatomical site (i.e., urogenital, pharyngeal and rectal body sites) at Baseline and whose baseline susceptibility test result showed no pre-existing resistance to ceftriaxone or azithromycin (i.e., participants with *N. gonorrhoeae* isolates that had ceftriaxone MIC >0.25 mg/L and azithromycin MIC >1 mg/L were excluded from the population). The following summarizes the clinical microbiology assessments (Table 131):

In the micro-MITT population, 744 participants had central laboratory confirmed, urogenital cultures that were positive for *N. gonorrhoeae* at Baseline (506 participants in the zoliflodacin arm and 238 participants in the ceftriaxone-azithromycin arm). A total of 81 participants (8.7%) had positive pharyngeal cultures (53 participants in the zoliflodacin arm and 28 participants in the ceftriaxone-azithromycin arm) and 114 participants (12.3%) had positive rectal cultures (79 participants in the zoliflodacin arm and 35 participants in the ceftriaxone-azithromycin arm).

The primary efficacy outcome defined as microbiological cure as determined by culture at urethral or cervical sites at TOC (Day 6±2) in the micro-MITT population was 90.4% (460/506 participants) for the zoliflodacin arm and 96.2% (229/238 participants) in the ceftriaxone-azithromycin arm. The microbiological failure rate was higher in the zoliflodacin arm (9.1%) compared to the ceftriaxone-azithromycin arm (3.8%). Most participants who had microbiological failure had nonassessable microbiological outcome at TOC either due to a missed TOC Visit, unobtainable specimen, or participant visit that was out-of-window. There were 15 participants in the zoliflodacin arm and no participants in the ceftriaxone-azithromycin arm that continued to test positive for *N. gonorrhoeae* at the urogenital site at TOC (see genomic analyses studies below).

The microbiological cure by culture at the pharyngeal site at TOC was 79.2% (42/53 participants) for the zoliflodacin arm compared to 78.6% (22/28 participants) in the ceftriaxone-azithromycin arm. The microbiological cure by culture at the rectal site at TOC was 87.3% (69/79 participants) in the zoliflodacin arm compared to 88.6% (31/35 participants) in the ceftriaxone-azithromycin arm. The proportions of participants per arm with microbiological cure were similar in the clinically-evaluable and per-protocol populations.

The proportion of males with microbiological cure in the ceftriaxone-azithromycin arm were higher compared to zoliflodacin arm, and the proportion of females with microbiological cure in the zoliflodacin arm were higher compared to ceftriaxone-azithromycin arm. There

were numerical differences in treatment response between the male and female subgroups, but the comparisons overlapped, indicating similarity of treatment effect.

The clinical cure at TOC of symptomatic gonorrhea infection in participants assigned male at birth who had at least one sign or symptom of urethral gonorrhea at Baseline was similar in both treatment arms.

Table 131. Microbiological Cure for *Neisseria gonorrhoeae* by Anatomic Site in Participants of Phase 3 Clinical Trial STI_Zoli001 at TOC Visit (Micro-MITT Population)

Parameter	Zoliflodacin n/N (%)	Ceftriaxone-Azithromycin n/N (%)	Difference (95% CI)
Urogenital site at TOC (primary endpoint)			
Microbiological cure	460/506 (90.9)	229/238 (96.2)	−5.36 (−8.6, −1.4)
Microbiological failure	46/506 (9.1)	9/238 (3.8)	
Positive for NG	15/506 (3.0)	0/238 (0.0)	
Nonassessable	31/506 (6.1)	9/238 (3.8)	
Missed TOC visit	15/506 (3.0)	6/238 (2.5)	
TOC OOW	12/506 (2.4)	2/238 (0.8)	
Unobtainable specimen	4/506 (0.8)	1/238 (0.4)	
Secondary endpoints			
Pharyngeal site at TOC			
Micro-MITT	42/53 (79.2)	22/28 (78.6)	0.7 (−20.8, 16.8)
Clinical evaluable	42/46 (91.3)	22/23 (95.7)	−4.4 (−16.5, 13.1)
Per-protocol	39/46 (84.8)	20/23 (87.0)	−2.2 (−17.7, 18.4)
Rectal site at TOC			
Micro-MITT	69/79 (87.3)	31/35 (88.6)	−1.2 (−12.6, 14.4)
Clinical evaluable	69/72 (95.8)	31/31 (100.0)	−4.2 (−11.6, 7.2)
Per-protocol	61/64 (95.3)	30/31 (96.8)	−1.5 (−10.5, 11.9)
Microbiological cure by gender at urogenital site (micro-ITT)			
Male	412/456 (90.4)	213/220 (96.8)	−6.4 (−9.93, 2.45)
Female	48/50 (96.0)	16/18 (88.9)	7.11 (5.28, −28.9)
Clinical cure in biological males at urogenital site at TOC (micro-ITT)			
Micro-MITT	375/460 (81.5)	194/220 (88.2)	−6.6 (−11.91, 0.73)
Per-protocol	345/410 (84.1)	183/201 (91.0)	−6.9 (−11.91, 1.61)

Source: Study Report# STI_Zoli001
Abbreviations: Micro-MITT, microbiological modified Intent-to-treat; NG, *Neisseria gonorrhoeae*; N, number of participants in treatment arm; n, number of participants with outcome; OOW, out-of-window assessment; TOC, test-of-cure

Zoliflodacin MIC values ranged from ≤0.008 to 0.5 mg/L with $MIC_{50/90}$ values of 0.06 mg/L and 0.12 mg/L, respectively (Table 132). There were no major differences in baseline zoliflodacin susceptibility by anatomic sites (i.e., urogenital, pharyngeal and rectal). There was no observable trend towards higher zoliflodacin MIC values among participants who had microbiological failure compared to participants who had successful microbiological outcomes.

Approximately 75% of urogenital isolates were nonsusceptible to ciprofloxacin at Baseline. All isolates that were nonsusceptible to ciprofloxacin had zoliflodacin MIC values ≤0.25 mg/L, suggesting no cross-resistance between ciprofloxacin and zoliflodacin despite both agents targeting DNA gyrase. Tetracycline resistance was >60% for urogenital isolates, which was the same between treatment arms. The percent of resistant isolates varied by location with ranges of 0.7% to 18.7% for azithromycin and 36.8% to 97.5% for ciprofloxacin (based on CLSI criteria).

The overall eradication in the zoliflodacin arm varied between 50.0% to 100% depending on the anatomic site (Table 6). When categorized by MIC at the urogenital site, a favorable microbiological response was observed in 85.0% at MICs ≤0.008 mg/L, 84.1% at ≤0.0015 mg/L, 90.2% at ≤0.03 mg/L, 91.4% at ≤0.06 mg/L, 91.4% at ≤0.12 mg/L, 90.9% at ≤0.25 mg/L.

Table 132. Microbiological Cure at TOC by Anatomic Site and Baseline *Neisseria gonorrhoeae* Zoliflodacin MIC (Micro-MITT), Trial STI_Zoli001

Zoliflodacin MIC (mg/L)	Urogenital N=506	Pharyngeal N=53	Rectal N=79
≤0.008	51/60 (85.0)	5/6 (83.3)	9/12 (75.0)
0.015	7/9 (77.8)	1/1 (100.0)	2/2 (100.0)
0.03	96/104 (94.2)	10/10 (100.0)	118/126 (93.7)
0.06	189/207 (91.3)	12/13 (92.3)	26/31 (83.9)
0.12	102/109 (93.6)	9/13 (69.2)	19/19 (100.0)
0.25	11/15 (73.3)	4/8 (50.0)	3/3 (100.0)
0.5	1/1 (100.0)	1/2 (50.0)	0/0

Source: Study Report# STI_Zoli001
Abbreviations: MIC, minimum inhibitory concentration; Micro-MITT, microbiological modified Intent-to-treat; N, number of participants assessed per anatomical site; TOC, test-of-cure

Three participants had zoliflodacin MICs of 0.5 mg/L (2 pharyngeal and 1 urogenital site). All three participants had infection at an additional anatomic site which yielded *N. gonorrhoeae* isolates with zoliflodacin MICs ≤0.25 mg/L; all were eradicated at TOC (Table 133).

In a post hoc susceptibility retesting of the isolates, the zoliflodacin MIC of 0.5 mg/L was confirmed for both pharyngeal isolates; however, the urogenital isolate when retested showed zoliflodacin MIC of 0.12 mg/L. This participant had a microbiological cure at TOC.

Of the two participants with infection at the pharyngeal site, one participant (Participant (b) (6)) had a microbiological cure while the other participant (Participant (b) (6)) had microbiological failure.

Whole genome sequencing showed that none of the 3 isolates with MIC values of 0.5 mg/L had mutations known to be associated with an increase in zoliflodacin MIC (i.e., *GyrB* D429, K450 or S467N mutations, see also Section 20.2). It is possible that isolates with zoliflodacin MICs of 0.5 mg/L may represent the high end of the wildtype distribution. None of the isolates were tested for the efflux overproduction or expression.

Table 133. Whole Genome Sequencing of Isolates With Zoliflodacin MICs of 0.5 mg/L

Patient ID	Isolate ID[a]	Visit	Anatomical Site	Microbiological Outcome at TOC	Zoliflodacin MIC (mg/L)	MLST ST	NG-STAR ST
(b) (6)	(b) (6) -1AU	Baseline	URO	Cure	0.5	17441	3434
	-1AP	Baseline	PH	Cure	0.5	11422	193
	-1AP	Baseline	PH	Failure	0.5	11422	193

Source: Study Report PC0914-2024-0008 Table 4
Abbreviations: ID, identification; MIC, minimum inhibitory concentration, SNPs, single nucleotide polymorphisms; MLST, multi-locus sequence typing; ST, sequence type; NG-STAR, *Neisseria gonorrhoeae* sequence typing for antimicrobial resistance; URO, urogenital; REC, Rectal; PH, pharyngeal

There were 19 participants who had microbiological failures in the zoliflodacin arm. Of the 19 participants, 15 participants had microbiological failures at the urogenital site, five participants at the pharyngeal site and three participants at the rectal site. There were no notable shifts in zoliflodacin MICs from Baseline to TOC. Molecular and phenotypic characterization of the baseline and TOC *N. gonorrhoeae* isolates were performed on isolates cultured from participants in the zoliflodacin arm. Whole genome sequencing revealed no emergence of resistance to zoliflodacin, as TOC isolates lacked zoliflodacin resistance mutations associated with an increase in zoliflodacin (i.e., *GyrB* D429, K450 or S467N mutations). Detailed whole genome sequencing of the 19 participants with microbiological failure showed that:

Nine participants had microbiological failure at the urogenital site only, of which 7 were documented as persistence of the baseline isolate by identical multi-locus sequence typing (MLST) and NG-sequence typing for antimicrobial resistance (STAR) sequence types (Table 134). These 7 participants included Participants (b) (6) The remaining two participants had a different isolate at TOC than that obtained at Baseline. Participant (b) (6) had MLST/NG-STAR sequence type 11422/193 at Baseline and sequence type 10314/2148 at TOC. Participant (b) (6) had sequence type 1587/427 at Baseline and sequence type 1588/1632 at TOC. It is possible that the baseline isolate was eradicated. The TOC isolate appears to be the result of a new infection indicating the possibility of re-infection with a novel strain.

Table 134. Whole Genome Sequencing of Isolates in Participants With Microbiological Failures (Urogenital Only), Trial STI_Zoli001

Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
Urogenital only												
(b) (6)	Netherlands	40	M	Neg	Visit 1/Day 1	Urogenital	0.25	2	0.015	11422	193	--
					Visit 4/Day 6	Urogenital	0.03	0.12	≤0.002	10314	2148	Microbiological failure. Strain differs from baseline urogenital isolate
	South Africa	34	M	Pos	Visit 1/Day 1	Urogenital	≤0.008	≤0.06	≤0.002	1587	427	--
					Visit 4/Day 6	Urogenital	≤0.008	≤0.06	≤0.002	1587	427	Microbiological failure. Persistence of baseline urogenital isolate
	South Africa	26	M	Pos	Visit 1/Day 1	Urogenital	0.06	0.12	0.015	14794	5643	--
					Visit 4/Day 6	Urogenital	0.06	0.12	0.015	14794	5643	Microbiological failure. Persistence of baseline urogenital isolate
	South Africa	21	M	Missing	Visit 1/Day 1	Urogenital	0.06	0.12	0.015	14794	5643	--
					Visit 4/Day 6	Urogenital	0.06	0.12	0.015	14794	5643	Microbiological failure. Persistence of baseline urogenital isolate
	South Africa	45	M	Neg	Visit 1/Day 1	Urogenital	0.015	≤0.06	0.004	18033	1496	--
					Visit 4/Day 6	Urogenital	0.015	≤0.06	0.004	18033	1496	Microbiological failure. Persistence of baseline urogenital isolate
	South Africa	22	F	Neg	Visit 1/Day 1	Urogenital	≤0.008	≤0.06	≤0.002	1587	427	--
					Visit 4/Day 6	Urogenital	0.015	≤0.06	0.004	1588	1632	Microbiological failure. Strain differs from baseline urogenital isolate
	South Africa	30	M	Neg	Visit 1/Day 1	Urogenital	0.03	0.12	0.015	13942	1623	--
					Visit 4/Day 6	Urogenital	0.03	≤0.06	0.008	13942	1623	Microbiological failure. Persistence of baseline urogenital isolate
	Thailand	43	M	Neg	Visit 1/Day 1	Urogenital	0.06	0.12	0.008	1587	719	--
					Visit 4/Day 6	Urogenital	0.06	0.12	0.008	1587	719	Microbiological failure. Persistence of baseline urogenital isolate

Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
(b) (6)	United States	28	M	Neg	Visit 1/Day 1	Urogenital	0.06	0.25	0.008	15228	2978	--
					Visit 4/Day 6	Urogenital	0.06	0.25	0.008	15228	2978	Microbiological failure. Persistence of baseline urogenital isolate

Source: Study Report PC0914-2024-0008 Table 2
Abbreviations: AZI, azithromycin; CRO, ceftriaxone; F, female; ID, identification; M, male; MIC, minimum inhibitory concentration, SNPs, single nucleotide polymorphisms; MLST, multi-locus sequence typing; ST, sequence type; NG-STAR, *Neisseria gonorrhoeae* sequence typing for antimicrobial resistance; URO, urogenital; REC, Rectal; PH, pharyngeal; ZFD, zoliflodacin

Four participants had infections at both the urogenital and pharyngeal sites (Table 135). In two of these participants ((b) (6) and (b) (6)), the baseline pathogen at the urogenital site was eradicated; however, there was documented persistence of the pharyngeal pathogen (i.e., identical MLST and NG-STAR sequence types at Baseline and TOC). Participant (b) (6) had documented persistence of *N. gonorrhoeae* at both the urogenital and pharyngeal sites. Participant (b) (6) only had a urogenital isolate at Baseline which was eradicated by TOC; however, a pharyngeal isolate of the same sequence type was obtained at TOC and end-of-trial. This participant did not meet the definition of microbiological failure as there was no pharyngeal isolate at Baseline; however, the participant was included in the whole genome sequencing analyses due to the presence of a postbaseline isolate.

Table 135. Whole Genome Sequencing of Isolates in Participants With Microbiological Failures (Urogenital and Pharyngeal Sites), Trial STI_Zoli001

Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
Urogenital + Pharyngeal												
(b) (6)	Netherlands	37	M	Neg	Visit 1/Day 1	Urogenital	0.25	2	0.015	11422	193	Microbiological cure
					Visit 1/Day 1	Pharyngeal	0.5	4	0.03	11422	193	--
					Visit 4/Day 6	Pharyngeal	0.25	4	0.015	11422	193	Microbiological failure. Persistence of baseline pharyngeal isolate
	Netherlands	28	M	Neg	Visit 1/Day 1	Urogenital	0.06	1	0.015	9362	4463	Microbiological cure
					Visit 4/Day 6	Pharyngeal	0.06	2	0.015	9362	4463	No pharyngeal isolate at baseline (same strain type as urogenital cure)
					Visit 5/Day 30	Pharyngeal	0.06	1	0.015	9362	4463	No pharyngeal isolate at baseline. *N. gonorrhoeae* isolated from pharynx at Visit 4 and 5 (same strain type as urogenital cure)
	United States	30	M	Neg	Visit 1/Day 1	Urogenital	0.06	0.12	0.004	8154	3908	Microbiological cure
					Visit 1/Day 1	Pharyngeal	0.25	0.12	0.008	8154	3908	--
					Visit 4/Day 6	Pharyngeal	0.12	0.12	0.008	8154	3908	Microbiological failure. Persistence of baseline pharyngeal isolate
	United States	22	M	Neg	Visit 1/Day 1	Urogenital	0.25	2	0.008	9363	3966	--
					Visit 4/Day 6	Urogenital	0.25	2	0.015	9363	3966	Microbiological failure. Persistence of baseline urogenital isolate
					Visit 1/Day 1	Pharyngeal	0.25	2	0.015	9363	3966	--
					Visit 4/Day 6	Pharyngeal	0.25	4	0.015	9363	3966	Microbiological failure. Persistence of baseline pharyngeal isolate

Source: Study Report PC0914-2024-0008 Table 2
Abbreviations: AZI, azithromycin; CRO, ceftriaxone; F, female; ID, identification; M, male; MIC, minimum inhibitory concentration, SNPs, single nucleotide polymorphisms; MLST, multi-locus sequence typing; ST, sequence type; NG-STAR, *Neisseria gonorrhoeae* sequence typing for antimicrobial resistance; URO, urogenital; REC, Rectal; PH, pharyngeal; ZFD, zoliflodacin

Five participants had *N. gonorrhoeae* at both urogenital and rectal sites (Table 136). In two of these participants (Participants (b) (6) and (b) (6)), the baseline pathogen at the rectal site was eradicated; however, there was documented persistence of the urogenital pathogen. Participant (b) (6) had different sequence types at the urogenital and rectal sites (sequence type 1587/427 and 1587/5642, respectively); however, the rectal site was eradicated with documented persistence at the urogenital site. Participant (b) (6) had different sequence types at the urogenital site (sequence type 11368/5664) and the rectal site (sequence

type 1587/427) and documented persistence at both anatomical sites. Participant (b) (6) had different sequence types isolated from the urogenital (sequence type 1583/5641) and the rectal (sequence type 14601/5204) sites at Baseline. At TOC, the urogenital isolate was eradicated, and a new sequence type was isolated at the rectal site (sequence type 1587/new). This scenario could represent an eradication of baseline infections at both sites and a possible new infection at the rectal site at TOC.

Table 136. Whole Genome Sequencing of Isolates in Participants With Microbiological Failures (Urogenital and Rectal Sites), Trial STI_Zoli001

Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
Urogenital + Rectal												
(b) (6)	South Africa	29	M	Neg	Visit 1/Day 1	Urogenital	0.12	0.12	0.015	11368	5664	--
					Visit 4/Day 6	Urogenital	0.12	0.12	0.015	11368	5664	Microbiological failure. Persistence of baseline urogenital isolate
					Visit 1/Day 1	Rectal	≤0.008	≤0.06	0.015	1587	427	--
					Visit 4/Day 6	Rectal	≤0.008	≤0.06	0.004	1587	427	Microbiological failure. Persistence of baseline rectal isolate. Note: baseline urogenital and rectal isolates are different strain types
	South Africa	27	M	Neg	Visit 1/Day 1	Urogenital	0.015	≤0.06	0.004	1583	5641	Microbiological cure
					Visit 1/Day 1	Rectal	0.06	≤0.06	0.004	14601	5204	Note: baseline urogenital and rectal isolates are different strain types.
					Visit 4/Day 6	Rectal	≤0.008	≤0.06	0.004	1587	New[b]	Microbiological failure. Strain differs from baseline rectal isolate and both are different from baseline urogenital isolate.
	South Africa	29	M	Neg	Visit 1/Day 1	Urogenital	≤0.008	≤0.06	0.004	1587	427	--
					Visit 4/Day 6	Urogenital	≤0.008	≤0.06	0.004	1587	427	Microbiological failure. Persistence of baseline urogenital isolate
					Visit 1/Day 1	Rectal	0.03	0.12	0.008	1587	5642	Microbiological cure. Note: baseline urogenital and rectal isolates are different strain types.
	South Africa	27	M	Neg	Visit 1/Day 1	Urogenital	≤0.008	≤0.06	0.004	1587	427	--
					Visit 4/Day 6	Urogenital	≤0.008	≤0.06	0.004	1587	427	Microbiological failure. Persistence of baseline urogenital isolate.
					Visit 1/Day 1	Rectal	≤0.008	≤0.06	0.004	1587	427	Microbiological cure
(b) (6)	South Africa	55	M	Pos	Visit 1/Day 1	Urogenital	0.03	0.12	0.008	14794	5643	--
					Visit 4/Day 6	Urogenital	0.06	0.12	0.008	14794	5643	Microbiological failure. Persistence of baseline urogenital isolate
					Visit 1/Day 1	Rectal	0.06	0.12	0.008	14794	5643	Microbiological cure

Source: Study Report PC0914-2024-0008 Table 2
Abbreviations: ID, identification; MIC, minimum inhibitory concentration, SNPs, single nucleotide polymorphisms; MLST, multi-locus sequence typing; ST, sequence type; NG-STAR, *Neisseria gonorrhoeae* sequence typing for antimicrobial resistance; URO, urogenital; REC, Rectal; PH, pharyngeal

Two participants had *N. gonorrhoeae* isolated from all three anatomic sites (urogenital, pharyngeal and rectal, Table 137). Participant (b) (6) had the same sequence type isolated from the urogenital and pharyngeal sites (sequence type 17948/3549) and a different sequence type isolated from the rectum (15331/2482) at Baseline. The baseline isolates from the urogenital and rectal sites were eradicated; however, the isolate at the pharyngeal site was documented as persistence. Participant (b) (6) had the same sequence type isolated from the pharyngeal and rectal sites at Baseline (sequence type 1587/427). The pharyngeal isolate was eradicated; however, the rectal isolate persisted. The baseline isolate from the urogenital tract was a different sequence type than the pharyngeal and rectal isolates (sequence type 13942/1623). A third sequence type was isolated from the urogenital tract at TOC (sequence type 11368/5664)

suggesting that the baseline urogenital isolate was eradicated, and the TOC isolate represented a possible new infection.

Table 137. Whole Genome Sequencing of Isolates in Participants With Microbiological Failures (Urogenital, Pharyngeal and Rectal Sites), Trial STI_Zoli001

Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
Urogenital + Pharyngeal + Rectal												
(b) (6)	South Africa	29	M	Neg	Visit 1/Day 1	Urogenital	0.03	≤0.06	0.008	13942	1623	--
					Visit 4/Day 6	Urogenital	0.12	≤0.06	0.008	11368	5664	Microbiological failure. Strain differs from baseline urogenital isolate
					Visit 1/Day 1	Pharyngeal	≤0.008	≤0.06	0.004	1587	427	Microbiological cure. Note: baseline urogenital and pharyngeal isolates are different strain types
					Visit 1/Day 1	Rectal	≤0.008	≤0.06	0.004	1587	427	Note: baseline urogenital and rectal strains are different strain types but rectal and pharyngeal strain types are the same
					Visit 4/Day 6	Rectal	≤0.008	0.12	0.004	1587	427	Microbiological failure. Strain differs from baseline rectal isolate by 28 genetic events, although baseline and TOC isolates are the same MLST type.
Patient ID	Country	Age	Sex at birth	HIV status	Visit	Site of infection	MIC (µg/mL)			MLST	NG STAR	Microbiological Outcome
							ZFD	AZM	CRO			
(b) (6)	Thailand	29	M	Neg	Visit 1/Day 1	Urogenital	0.06	1	0.008	17948	3549	Microbiological cure
					Visit 1/Day 1	Pharyngeal	0.12	2	0.008	17948	3549	--
					Visit 4/Day 6	Pharyngeal	0.12	2	0.008	17948	3549	Microbiological failure. Persistence of baseline isolate
					Visit 1/Day 1	Rectal	0.12	0.5	0.25	15331	2482	Microbiological cure. Note: baseline urogenital and pharyngeal isolate strain types are different from that of rectal baseline isolate strain type

Source: Study Report PC0914-2024-0008 Table 2
Abbreviations: ID, identification; MIC, minimum inhibitory concentration, SNPs, single nucleotide polymorphisms; MLST, multi-locus sequence typing; ST, sequence type; NG-STAR, *Neisseria gonorrhoeae* sequence typing for antimicrobial resistance; URO, urogenital; REC, Rectal; PH, pharyngeal

Overall, four participants were found to have *N. gonorrhoeae* isolated at TOC that were different from the isolates at Baseline (Participants (b) (6)). It is possible that these participants may have been infected with more than one isolate at the time of enrollment but only one strain was isolated at Baseline and the other strain was isolated at TOC. It is also possible that the strain isolated at Baseline was eradicated and the participant was newly infected with a second isolate in the interval between enrollment and the TOC Visit. As per protocol, each participant was specifically asked at each visit if they had unprotected sexual intercourse since the last visit. Three of the four participants (Participants (b) (6)) indicated "No" at TOC, therefore it is unclear which of the above scenarios, if any, applied to these participants. However, Participant (b) (6) answered "yes" to this question, therefore the isolate at the TOC most likely represented a new infection.

Most of the participants were infected with a unique sequence type. There were a few cases where the sequence type was isolated from multiple participants, with 1587/427 sequence type

observed in 6 participants (Participants (b) (6)). Sequence type 14794/5643 was observed in three participants (Participants (b) (6)). Sequence type 13942/1623 was observed in two participants (Participants (b) (6)). Participant (b) (6) was infected with two of the common sequence types. It is important to note that all participants infected with the common sequence types were enrolled in South Africa. It is unknown if the isolates were particularly common in South Africa or if the isolates contain virulence factors that cause these strains to be associated with microbiological failure.

The phase 2 supportive clinical trial (DMID 14-0014) evaluated the efficacy and safety of a single oral 2 g dose of zoliflodacin or 3 g dose of zoliflodacin compared with a single IM 500 mg dose of ceftriaxone in adult participants ages 18 to 55 years with uncomplicated urogenital gonorrhea. The phase 2 trial design and results are described elsewhere in the review (See Section 6). Of the 180 eligible participants, 179 participants were included in the micro-MITT population. A total of 141 had a positive urethral/cervical culture for *N. gonorrhoeae* (56 participants in the 3 g zoliflodacin arm, 57 participants in the 2 g zoliflodacin arm, and 28 participants in the ceftriaxone arm). There were 23 participants who had positive pharyngeal cultures (11 participants in the 3 g zoliflodacin arm, eight participants in the 2 g zoliflodacin arm, and four participants in the ceftriaxone arm) and 15 participants had positive rectal cultures (seven participants in the 3 g zoliflodacin arm, five participants in the 2 g zoliflodacin arm, and three participants in the ceftriaxone arm).

In participants treated with the 3 g zoliflodacin dose, zoliflodacin MIC values range from 0.008 to 0.25 mg/L with $MIC_{50/90}$ values of 0.06 mg/L and 0.12 mg/L, respectively (Table 138). No participants had zoliflodacin MICs of 0.5 mg/L or higher. These MIC results were similar to what was observed in the phase 3 trial.

There were three participants who had microbiological failure, one participant with infection at the urogenital site and two participants with infection at the pharyngeal site. No participants had failures at the rectal site.

Participant (b) (6) had positive cultures at the urogenital and rectal sites at Baseline, but only the urogenital site isolate was tested for MIC which was 0.06 mg/L. At TOC, the rectal site was culture negative for *N. gonorrhoeae* but urogenital infection was persistent. The zoliflodacin MIC was 0.06 mg/L.

Participant (b) (6) had positive cultures at the urogenital and pharyngeal sites at Baseline; both isolates had similar zoliflodacin MICs of 0.125 mg/L. At TOC, the urogenital site was culture negative for *N. gonorrhoeae* but infection at the pharyngeal site was persistent (zoliflodacin MIC was 0.125 mg/L).

Participant (b) (6) had positive cultures at the urogenital and pharyngeal sites at Baseline; both isolates had similar zoliflodacin MICs of 0.06 mg/L. The urogenital site was culture negative for *N. gonorrhoeae* at TOC, but infection at the pharyngeal site was persistent (zoliflodacin MIC was 0.06 mg/L).

Table 138. Microbiological Cure at TOC by Anatomic Site and Baseline *Neisseria gonorrhoeae* Zoliflodacin MIC (Micro-MITT), Trial DMID 14-0014

Zoliflodacin MIC (mg/L)	Urogenital (n=506)	Pharyngeal (n=53)	Rectal (n=79)
0.008	1/1 (100.0)	1/1 (100.0)	0/0
0.015	0/0	0/0	0/0
0.03	5/5 (100.0)	0/0	0/0
0.06	21/22 (95.5)	2/3 (66.7)	3/3 (100.0)
0.12	22/22 (100.0)	5/6 (83.3)	2/2 (100.0)
0.25	5/5 (100.0)	1/1 (100.0)	1/1 (100.0)
0.5	0/0	0/0	0/0

Source: Study Report# STI_DMID 14-0014
Abbreviations: Micro-MITT, microbiological modified Intent-to-treat; MIC, minimum inhibitory concentration; n, number of participants assessed; TOC, test-of-cure

19.3. Susceptibility Testing Interpretive Criteria

The following data and analyses supported the recommended susceptibility interpretive criteria for zoliflodacin:

Surveillance studies and the clinical trials (phase 2 and 3) showed that the upper bound of the wild-type population of zoliflodacin MIC is 0.5 mg/L. Isolates from different geographical regions globally and U.S. surveillance studies (2012 to 2023) showed similar zoliflodacin activity that remained consistent with MIC_{90} values ranging from 0.125 to 0.25 mg/L. The highest zoliflodacin MIC result from these studies was 0.5 mg/L which was only seen in two studies from Europe. In the phase 3 clinical trial (STI_Zoli001), there were three participants who had zoliflodacin MICs at 0.5 mg/L.

The PK-PD target attainment analyses showed that for isolates with zoliflodacin MIC values ≤0.25 mg/L, plasma free drug exposure $AUC_{0-\infty}$/MIC ratio target of 70.6 will be achieved in ≥90% of participants receiving a 3 g oral zoliflodacin dose under fasted conditions for patients weighing ≥35 kg to < 50 kg or under fed conditions for patients weighing ≥50 kg.

Clinical evidence shows high microbiological eradication against *N. gonorrhoeae* isolates at the urogenital site with zoliflodacin MIC values ≤0.25 mg/L. There were three participants who had zoliflodacin MICs of 0.5 mg/L; two of those participants had microbiological cures and one participant had microbiological failure. Overall, the proportion of participants with microbiological favorable response at zoliflodacin MICs of ≤0.25 mg/L was 90.9%.

There are no disk correlates for the zoliflodacin antimicrobial susceptibility testing.

The recommended susceptibility interpretive criteria are summarized in Table 139 below.

Table 139. Recommended Susceptibility Interpretive Criteria for Zoliflodacin

	Agar Dilution (mg/L)		
Pathogen	Susceptible (S)	Intermediate (I)	Resistant (R)
Neisseria gonorrhoeae	≤0.25	-	-

Source: Reviewer-generated table

20. Mechanism of Action/Drug Resistance

20.1. Mechanism of Action

Zoliflodacin is a bacterial type II topoisomerase inhibitor; like the fluroquinolones, the primary targets are DNA gyrase and DNA topoisomerase IV. These bacterial type II topoisomerases catalyze topological changes in DNA during replication by first unwinding the supercoiling in DNA and then introducing transient single- or double-stranded breaks in DNA followed by re-ligation. DNA gyrase catalyzes the ATP-dependent introduction of negative supercoils ahead of replication fork and transcription complexes to relieve torsional strain with DNA. DNA topoisomerase IV catalyzes decatenation which is essential for separating linked catenanes of two DNA molecules and can relax positive and negative supercoils behind replication forks, unlinking daughter chromosomes during cell division and removing knots formed during recombination events.[57]

Both DNA gyrase and DNA topoisomerase IV are heterotetrameric enzymes composed of two subunits with A_2B_2 structure. DNA gyrase subunits are *GyrA* and *GyrB* where *GyrA* is responsible for DNA cleavage and religation while *GyrB* is involved in the binding and hydrolysis of ATP to drive the catalytic reaction. DNA topoisomerase IV subunits are *GyrIA* and *GyrIB* (in gram-positive isolates) and *ParC* and *ParE* (in gram-negative isolates). The *ParC/GyrIA* is responsible for separating linked catenates of two DNA molecules during replication.[58,59,60]

Like fluoroquinolones, zoliflodacin inhibits DNA gyrase-catalyzed supercoiling and topoisomerase IV catalyzed decatenation activity in gram-positive and gram-negative isolates. In vitro assays with purified type II topoisomerase showed zoliflodacin inhibits DNA gyrase-catalyzed supercoiling and topoisomerase IV-catalyzed decatenation resulting in stabilization of the enzyme-DNA cleaved complex.[61,62,63,64,65]

The supercoiling assay primarily measures DNA gyrase activity by tracking migration or relaxed versus supercoiled DNA through agarose gels in the presence of inhibitors. Against *N.*

---

[57] Bradford, PA, AA Miller, J O'Donnell, and JP Mueller, 2020, Zoliflodacin: An Oral Spiropyrimidinetrione Antibiotic for the Treatment of *Neisseria gonorrhoeae*, Including Multi-Drug-Resistant Isolates, ACS Infect Dis, 6(6):1332-1345, https://www.ncbi.nlm.nih.gov/pubmed/32329999.

[58] Ibid.

[59] Kern, G, T Palmer, DE Ehmann, AB Shapiro, B Andrews, GS Basarab, P Doig, J Fan, N Gao, SD Mills, J Mueller, S Sriram, J Thresher, and GK Walkup, 2015, Inhibition of *Neisseria gonorrhoeae* Type II Topoisomerases by the Novel Spiropyrimidinetrione AZD0914, J Biol Chem, 290(34):20984-20994, https://www.ncbi.nlm.nih.gov/pubmed/26149691.

[60] Collins, JA, AA Oviatt, PF Chan, and N Osheroff, 2024, Target-Mediated Fluoroquinolone Resistance in *Neisseria gonorrhoeae*: Actions of Ciprofloxacin against Gyrase and Topoisomerase IV, ACS Infect Dis, 10(4):1351-1360, https://www.ncbi.nlm.nih.gov/pubmed/38606464.

[61] Study AZD0914-M2-005

[62] Study AZD0914-M2-016

[63] Study AZD0914-M2-017

[64] Study AZD0914-M2-030

[65] See Footnote 24

gonorrhoeae DNA gyrase, the supercoiling inhibition IC_{50} values for zoliflodacin and ciprofloxacin were 6.3µM and 1.1µM, respectively.

The decatenation assay primarily measures bacterial DNA topoisomerase IV activity by monitoring unlinking of catenated DNA substrates with linear double-strand breaks accumulating when topoisomerase IV is in a complex due to addition of an inhibitor. Against *N. gonorrhoeae* DNA topoisomerase IV, the decatenation IC_{50} values for zoliflodacin and ciprofloxacin were 19.1µM and 6.7µM, respectively.

Cleaved complex assays track the migration of single-stranded nicked DNA, double-stranded cleaved DNA, and intact DNA through agarose gels. Against *N. gonorrhoeae* the cleaved complex IC_{50} value of zoliflodacin and ciprofloxacin for DNA gyrase were 1.7µM and 1.9µM, respectively, and for DNA topoisomerase IV were 0.5µM and 0.063µM, respectively.

Zoliflodacin exhibited little inhibition of human topoisomerase IIα (IC_{50}>400µM versus 110µM for ciprofloxacin) and weak inhibition of human topoisomerase IIβ (IC_{50}=79µM) which appears comparable with ciprofloxacin (111µM).[66,67,68,69]

20.2. Drug Resistance

The primary target of zoliflodacin is the *gyrB* subunit of DNA gyrase in *N. gonorrhoeae* isolates. The frequency of first step spontaneous mutations was considered low with mutation frequencies that ranged from $<7.9\times10^{-10}$ to 1.5×10^{-8} at 4× MIC. Resistant *gyrB* mutants were reported to have a 16- to 32-fold increase in zoliflodacin MICs (1 to 2 mg/L). No spontaneous resistant mutants were detected at 8x MIC.[70,71,72] The frequency of second-step spontaneous mutations was not evident at 4x or 8x MIC with mutation frequencies below the limit of detection ranging from $<1.1\times10^{-9}$ to $<7.6\times10^{-10}$ after 24- and 48-hours incubation. After additional incubation (up to 96 hours), several colonies emerged which resulted in a further 4- to 8-fold increase in zoliflodacin MICs.[73,74]

---

[66] See Footnote 24

[67] Study AZD0914-M2-018

[68] Study AZD0914-M2-029

[69] Study AZD0914-M2-023

[70] Study AZD0914-M2-022

[71] Foerster, S, D Golparian, S Jacobsson, LJ Hathaway, N Low, WM Shafer, CL Althaus, and M Unemo, 2015, Genetic Resistance Determinants, In Vitro Time-Kill Curve Analysis and Pharmacodynamic Functions for the Novel Topoisomerase II Inhibitor ETX0914 (AZD0914) in *Neisseria gonorrhoeae*, Front Microbiol, 6:1377, https://www.ncbi.nlm.nih.gov/pubmed/26696986.

[72] Foerster, S, G Drusano, D Golparian, M Neely, LJV Piddock, E Alirol, and M Unemo, 2019, In vitro antimicrobial combination testing of and evolution of resistance to the first-in-class spiropyrimidinetrione zoliflodacin combined with six therapeutically relevant antimicrobials for *Neisseria gonorrhoeae*, J Antimicrob Chemother, 74(12):3521-3529, https://www.ncbi.nlm.nih.gov/pubmed/31730160.

[73] Study AZD0914-M2-034

[74] Alm, RA, SD Lahiri, A Kutschke, LG Otterson, RE McLaughlin, JD Whiteaker, LA Lewis, X Su, MD Huband, H Gardner, and JP Mueller, 2015, Characterization of the novel DNA gyrase inhibitor AZD0914: low resistance potential and lack of cross-resistance in *Neisseria gonorrhoeae*, Antimicrob Agents Chemother, 59(3):1478-1486, https://www.ncbi.nlm.nih.gov/pubmed/25534723.

Resistance studies of isolates with zoliflodacin MICs ≥0.5 mg/L showed that:

Mutations in the *GyrB* gene were the predominant mechanism of zoliflodacin resistance against *N. gonorrhoeae*. Whole genome sequencing of first-step zoliflodacin mutants showed two key mutations at the C-terminus of *GyrB* (Asp429Asn [D429N] or Lys450Thr [K450T] or Lys450Asn [K450N]) with zoliflodacin MICs of 1 to 2 mg/L. Second step zoliflodacin mutants developed additional *GyrB* mutations (S467N) that further increased MIC values by 4- to 8-fold.[75,76,77,78]

Against laboratory-derived *N. gonorrhoeae* that overproduced the efflux pumps *MtrCDE, MacAB* and *NorM*, the up-regulated efflux pumps were associated with zoliflodacin MIC increases from 0.125 mg/L in the wild-type parental strain to 1 to 4 mg/L in resistant strains. Inactivation of the *MtrCDE* efflux pump resulted in full restoration of susceptibility to zoliflodacin (MIC =0.125 mg/L). However, inactivation of the *MacAB* or *NorM* efflux pump did not appear to have a significant effect on zoliflodacin MICs in three of four strains tested.[79]

No cross resistance was observed for zoliflodacin against clinical isolates possessing frequently encountered mutations in either *GyrA, GyrB, parC* or *parE*, which are known to cause resistance to fluoroquinolones or other type II topoisomerases (novobiocin or Coumermycin A1). The zoliflodacin MICs ranged from 0.125 to 0.25 mg/L against these bacterial isolates. Conversely, zoliflodacin mutants carrying mutations in the *GyrB* region (D429N, K450T or K450N) did not significantly affect the MICs of ciprofloxacin, azithromycin, ceftriaxone or tetracycline.[80,81]

21. Other Drug Development Considerations

Not applicable.

22. Data Integrity–Related Consults (Office of Scientific Investigations, Other Inspections)

Please refer to Section 10 for a summary of findings from the clinical site inspection. Details of the three clinical site inspections and one Applicant site inspection completed by the Office of Scientific Investigations for Trial STI_Zoli001 are presented here. Based on the results of these inspections, Trial STI_Zoli001 appears to be conducted adequately, and the clinical data generated by these clinical sites appear to be reliable in support of this NDA.

[75] See Footnote 57
[76] See Footnote 59
[77] Study AZD0914-M2-022
[78] Study AZD0914-M2-034
[79] See Footnote 56
[80] Study AZD0914-M2-001
[81] See Footnote 57

The clinical sites were chosen using a risk-based approach primarily based on numbers of enrolled participants and treatment effect. The three clinical sites were Dr. Lindley Barbee (site 840-102), Dr. Sinead Delany-Maretlwe (site 710-003) and Dr. Nittaya Phanuphak Pungpapong (site 764-001). The Applicant office in Waltham, Massachusetts was also inspected.

The inspection results from a Clinical Inspection (CI) Summary by Dr. John Lee are included below:

Lindley Barbee, M.D.

908 Jefferson Street

Seattle, Washington 98104

Inspection Dates: August 18 to 26, 2025

Protocol STI_Zoli001, Site 840-102

70 participants were screened, 70 were enrolled, and 57 completed the study. Case records were completely reviewed for a random sampling of 19 enrolled participants.

Study conduct at this CI site was audited for: adherence to the study protocol, IRB oversight, site monitoring, staff training, study medication disposition and accountability, and CI financial disclosure.

Case records review covered: informed consent, participant eligibility, participant enrollment and randomization, efficacy endpoint assessment, and adverse event monitoring.

Data verification (against source data) covered: treatment assignment, major efficacy endpoints, adverse events, protocol deviations, and concomitant medication use.

The inspectional findings were noteworthy for four participants not excluded for: limited use of nonstudy antibiotics and CYP3A4 inhibitors (one participant), self-reported history of current substance abuse (two participants), and inadequate contraception (one participant).

These findings appeared isolated, minor, and unlikely to significantly impact the study outcome (4 of 930 participants, <0.5%). For each specific protocol deviation (nature), the frequency decreases further: 0.1% (antibiotic / CYP3A4 inhibitor), 0.2% (substance abuse), and 0.1% (inadequate contraception). For each protocol deviation, an appreciable impact on the relevant efficacy endpoints is not expected. Deviation-related adverse events (including pregnancy) were not observed.

Significant GCP deficiencies or regulatory violations were otherwise not observed. The Applicant's monitoring appeared adequate. The study records showed complete CI financial disclosure, adequate reporting of adverse events and protocol deviations, and acceptable drug accountability. The major efficacy and adverse event data were verifiable.

Sinead Delany-Moretlwe, M.D.

Hillbrow Health Precinct 22

Esselen Street Hillbrow

Johannesburg, South Africa

Inspection Dates: September 1 to 5, 2025

Protocol STI_Zoli001, Site 710-003

194 participants were screened, 149 participants were enrolled, and 135 completed the study. Case records were reviewed in detail for a random sampling of 40 enrolled participants.

Study conduct at this CI site was audited for: adherence to the study protocol, IRB oversight, site monitoring, staff training, study medication disposition and accountability, and CI financial disclosure.

Case records review covered: informed consent, participant eligibility, participant enrollment and randomization, efficacy endpoint assessment, and adverse event monitoring.

Data verification (against source data) covered: treatment assignment, major efficacy endpoints, adverse events, protocol deviations, and concomitant medication use.

Significant GCP deficiencies or regulatory violations were not observed. The Applicant's monitoring appeared adequate. Informed consent forms were obtained for all participants. The study records showed complete CI financial disclosure, adequate reporting of adverse events and protocol deviations, and acceptable drug accountability. The major efficacy and adverse event data were verifiable.

Nittaya Phanuphak Pungpapong, M.D.

104 Ratchadamri Road

Lumpini Pathumwan

Bangkok, Thailand

Inspection Dates: August 25 to 29, 2025

Protocol STI_Zoli001, Site 764-001

108 participants were screened, 105 participants were enrolled, and 103 completed the study. All participant case records were reviewed, including detailed review for a random sample of 20 enrolled participants.

Study conduct at this CI site was audited for: adherence to the study protocol, IRB oversight, site monitoring, staff training, study medication disposition and accountability, and CI financial disclosure.

Case records review covered: informed consent, participant eligibility, participant enrollment and randomization, efficacy endpoint assessment, and adverse event monitoring.

Data verification (against source data) covered: treatment assignment, major efficacy endpoints, adverse events, protocol deviations, and concomitant medication use.

The inspectional findings were noteworthy for seven participants under 18 years of age without either signed parental consent or documented IRB waiver. The protocol allowed enrollment of minors of age 12 years and older. If unable to obtain signed parental consent, minors could be enrolled with IRB approval. For the 7 minors without signed parental consent, verbal IRB approval (by phone call) was documented in the study records but was not followed by a written IRB approval.

Significant GCP deficiencies or regulatory violations were otherwise not observed. The Applicant's monitoring appeared adequate. The study records showed complete CI financial

disclosure, adequate reporting of adverse events and protocol deviations, and acceptable drug accountability. The major efficacy and adverse event data were verifiable.

Entasis Therapeutics, Inc.

35 Gatehouse Drive

Waltham, MA 02451

Inspection Dates: July 18 to 24, 2025

Protocol STI_Zoli001

This Bioresearch Monitoring Operations review-based Applicant inspection consisted of a general records review, to evaluate compliance with the GCP principles and regulations applicable to the Applicant, including oversight of CI sites.

The records reviewed included: product accountability records, monitoring correspondence, vendor contracts, documentation of data management procedures, safety reporting records, staff qualification and training, and clearance records for electronic data systems. The CI sites selected to evaluate the adequacy of the Applicant's oversight included those inspected separately, Sites 840-102, 710-003, and 764-001.

No significant regulatory violations were observed. The Applicant's oversight of the study including CI site monitoring appears to have been adequate to assure participant safety and study data quality.

For further details, please refer to the clinical inspection summary written by Dr. John Lee from the Office of Scientific Investigations, dated October 16, 2025.

23. Labeling: Key Changes

This PI review includes a high-level summary of the rationale for major changes to the finalized PI as compared to the Applicant's draft PI submitted on April 15, 2025 (see Table 140). The PI was reviewed to ensure that the PI meets regulatory/statutory requirements, is consistent (if appropriate) with labeling guidance, conveys clinically meaningful and scientifically accurate information needed for the safe and effective use of the drug, and provides clear and concise information for the healthcare practitioner.

The established pharmacologic class (EPC) for NUZOLVENCE in the Indications and Usage section of the Highlights section of the PI was modified to "spiropyrimidinetrione bacterial type II topoisomerase inhibitor" instead of (b) (4) that was proposed by the Applicant. The modified EPC provides a more complete EPC that includes the chemical structure and the mode of action of zoliflodacin that is scientifically valid and clinically meaningful. The word (b) (4) proposed in the Applicant's EPC was removed because the (b) (4) properties are evident with the inclusion of the mode of action for zoliflodacin of bacterial type II topoisomerase inhibitor in the modified EPC. This EPC modification was based on the labeling recommendations in the FDA guidance for industry *Labeling for Human Prescription Drug and Biological Products-Determining Established Pharmacological Class for*

Use in the Highlights of Prescribing Information.[82] Refer to Sections 5 and 20.1 for additional details.

Table 140. Key Labeling Changes and Considerations

(b) (4)

[82] FDA guidance for industry Labeling for Human Prescription Drug and Biological Products — Determining Established Pharmacologic Class for Use in the Highlights of Prescribing Information (October 2009).

2 Pages of Draft Labeling have been Withheld in Full as b4 (CCI/TS) immediately following this page

(b) (4)

[redacted]

Medication Guide and Instructions for Use

To mitigate the potential safety risks based on nonclinical findings, three Warnings were added to the PI as detailed above, i.e., Embryo-Fetal Toxicity: Potential Risk for Pregnant Females, Embryo-Fetal Toxicity: Potential Risk Related to Males with Female Partners of Reproductive Potential, and Testicular Toxicity and Risks to Male Fertility. Given these additions, a Medication Guide was required. An Instructions for Use was also developed based on the PI.

Please refer to the Division of Medical Policy Programs review (in DARRTS dated 11/04/2025) and the Office of Prescription Drug Promotion review (in DARRTS dated 11/06/2025) for additional information.

Carton and Container Labeling

The carton and container labeling were revised to reflect changes made to the PI. Please refer to the Division of Medication Error Prevention and Analysis review (in DARRTS dated July 31, 2025, November 5, 2025, and December 8, 2025) and Integrated Quality Review (in DARRTS dated October 14, 2025) for additional information.

23.1. Approved Labeling Types

Upon approval of this application, the following labeling documents will be FDA-approved:

1. Prescribing Information
2. Medication Guide
3. Instructions for Use
4. Container Labeling
5. Carton Labeling

24. Postmarketing Requirements and Commitments

The following postmarketing requirements have been requested by the FDA and agreed to by the Applicant. Please refer to the approval letter for the final PMR language and timelines.

PMRs under 505(o):

1. Conduct a worldwide descriptive study that collects prospective and retrospective data in women exposed to NUZOLVENCE (zoliflodacin) during pregnancy to assess risk of maternal complications, and adverse effects on the developing fetus, neonate, and infant during the first year of life.

The milestone dates are as follows:

Draft Protocol Submission: 07/2026
Final Protocol Submission: 01/2027
Interim Report Submission: 10/2030
Study Completion: 07/2036
Final Report Submission: 01/2037

2. Perform a lactation study (milk only or mother-infant pair study) in lactating women who have received NUZOLVENCE (zoliflodacin) to measure concentrations of zoliflodacin in

breast milk using a validated assay. Assess the effects on the breastfed infant, if available, based on the study population.

The milestone dates are as follows:

Draft Protocol Submission: 07/2026
Final Protocol Submission: 10/2026
Study Completion: 12/2027
Final Report Submission: 06/2028

3. Conduct a U.S. surveillance study over a 5-year period after the introduction of NUZOLVENCE (zoliflodacin) to the market to determine if resistance or decreased susceptibility to NUZOLVENCE is occurring in the target population of bacteria that are in the approved NUZOLVENCE label.

The milestone dates are as follows:

Draft Protocol Submission: 10/2026
Final Protocol Submission: 12/2026
Interim report #1 submission: 10/2028
Interim report #2 submission: 10/2029
Interim report #3 submission: 10/2030
Interim report #4 submission: 10/2031
Interim report #5 submission: 10/2032
Study Completion: 10/2032
Final Report Submission: 06/2033

4. Conduct a clinical trial to evaluate the effect of NUZOLVENCE (zoliflodacin) on human testicular function.

The milestones dates are as follows:

Draft Protocol Submission: 03/2026
Final Protocol Submission: 06/2026
Study Completion: 12/2027
Final Report Submission: 06/2028

25. Financial Disclosure

Table 141. Clinical Investigator Financial Disclosure for [STI_Zoli001, DMID 14-0014, STI_Zoli002, STI_Zoli003, DMID 16-0110, DMID 16-0118, D4930C00001, D4930C00003]

Was a list of clinical investigators provided:	Yes ☒	No ☐ (Request list from Applicant)
Total number of investigators identified: 196		
Number of investigators who are Sponsor employees (including both full-time and part-time employees): 0		
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): 0		
If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c), and (f)): Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: 0 Significant payments of other sorts: 0 Proprietary interest in the product tested held by investigator: 0 Significant equity interest held by investigator: 0 Sponsor of covered study: 0		
Is an attachment provided with details of the disclosable financial interests/arrangements:	Yes ☒	No ☐ (Request details from Applicant)
Is a description of the steps taken to minimize potential bias provided:	Yes ☒	No ☐ (Request information from Applicant)
Number of investigators with certification of due diligence (Form FDA 3454, box 3): 15		
Is an attachment provided with the reason:	Yes ☒	No ☐ (Request explanation from Applicant)

Abbreviation: FDA, Food and Drug Administration

Out of 196 investigators, a total of 15 investigators did not have financial disclosure forms. One person retired during the study and a financial disclosure form was not obtained prior to retirement. One investigator was inadvertently added to the study site but was not delegated any investigator tasks. The Applicant was not able to locate the financial disclosure forms for six investigators. There were seven investigators who left their sites prior to the start of the study and did not perform any study-related activities.

26. References

Alm, RA, SD Lahiri, A Kutschke, LG Otterson, RE McLaughlin, JD Whiteaker, LA Lewis, X Su, MD Huband, H Gardner, and JP Mueller, 2015, Characterization of the novel DNA gyrase inhibitor AZD0914: low resistance potential and lack of cross-resistance in *Neisseria gonorrhoeae*, Antimicrob Agents Chemother, 59(3):1478-1486, https://www.ncbi.nlm.nih.gov/pubmed/25534723.

Atallah-Yunes, SA, A Ready, and PE Newburger, 2019, Benign ethnic neutropenia, Blood Rev, 37:100586, https://www.ncbi.nlm.nih.gov/pubmed/31255364.

Bradford, PA, AA Miller, J O'Donnell, and JP Mueller, 2020, Zoliflodacin: an oral spiropyrimidinetrione antibiotic for the treatment of *Neisseria gonorrheae*, including multi-drug-resistant isolates, ACS Infect Dis, 6(6):1332-1345, https://www.ncbi.nlm.nih.gov/pubmed/32329999.

CDC, 2021, Gonococcal infections among adolescents and adults, https://www.cdc.gov/std/treatment-guidelines/gonorrhea-adults.htm.

CDC, 2023, Sexually transmitted infections surveillance 2023, https://www.cdc.gov/sti-statistics/media/pdfs/2025/09/2023_STI_Surveillance_Report_FINAL_508.pdf.

CDC, 2024, Drug-resistant gonorrhea, https://www.cdc.gov/gonorrhea/hcp/drug-resistant/index.html.

(b) (4)

Collins, JA, AA Oviatt, PF Chan, and N Osheroff, 2024, Target-mediated fluoroquinolone resistance in *Neisseria gonorrhoeae*: actions of Ciprofloxacin against Gyrase and Topoisomerase IV, ACS Infect Dis, 10(4):1351-1360, https://www.ncbi.nlm.nih.gov/pubmed/38606464.

CTEP, Common Terminology Criteria for Adverse Events (CTCAE), Version 5.0, Place Published: Cancer Therapy Evaluation Program. https://dctd.cancer.gov/research/ctep-trials/for-sites/adverse-events/ctcae-v5-5x7.pdf.

Daniels, G, 2005, The molecular genetics of blood group polymorphism, Transpl Immunol, 14(3-4):143-153, https://www.ncbi.nlm.nih.gov/pubmed/15982556.

FDA draft guidance for industry *Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products* (December 2019).

FDA draft guidance for industry *Demonstrating Substantial Evidence of Effectiveness With One Adequate and Well-Controlled Clinical Investigation and Confirmatory Evidence* (September 2023).

FDA guidance for industry *Labeling for Human Prescription Drug and Biological Products — Determining Established Pharmacologic Class for Use in the Highlights of Prescribing Information* (October 2009).

FDA guidance for industry *Providing Clinical Evidence of Effectiveness for Human Drug and Biological Products* (May 1998).

FDA guidance for industry *Uncomplicated Gonorrhea: Developing Drugs for Treatment* (August 2015).

Foerster, S, G Drusano, D Golparian, M Neely, LJV Piddock, E Alirol, and M Unemo, 2019, In vitro antimicrobial combination testing of and evolution of resistance to the first-in-class spiropyrimidinetrione zoliflodacin combined with six therapeutically relevant antimicrobials for *Neisseria gonorrhoeae*, J Antimicrob Chemother, 74(12):3521-3529, https://www.ncbi.nlm.nih.gov/pubmed/31730160.

Foerster, S, D Golparian, S Jacobsson, LJ Hathaway, N Low, WM Shafer, CL Althaus, and M Unemo, 2015, Genetic resistance determinants, in vitro time-kill curve analysis and pharmacodynamic functions for the novel Topoisomerase II inhibitor ETX0914 (AZD0914) in *Neisseria gonorrhoeae*, Front Microbiol, 6:1377, https://www.ncbi.nlm.nih.gov/pubmed/26696986.

Hiruy, H, S Bala, JM Byrne, KG Roche, SH Jang, P Kim, S Nambiar, D Rubin, Y Yasinskaya, LH Bachmann, K Bernstein, R Botgros, S Cammarata, RL Chaves, CD Deal, GL Drusano, EM

Duffy, AE Eakin, S Gelone, T Hiltke, EW Hook Iii, AE Jerse, CJ McNeil, L Newman, S O'Brien, C Perry, HEL Reno, RA Romaguera, J Sato, M Unemo, TEC Wi, K Workowski, GA O'May, SJ Shukla, and JJ Farley, 2024, FDA, CDC, and NIH Co-sponsored public workshop summary-development considerations of antimicrobial drugs for the treatment of gonorrhea, Clin Infect Dis, https://www.ncbi.nlm.nih.gov/pubmed/39045871.

ICH guidance for industry *M12 Drug Interaction Studies* (August 2024), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/m12-drug-interaction-studies.

ICH guidance for industry *M10 Bioanalytical Method Validation and Study Sample Analysis* (November 2022), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/m10-bioanalytical-method-validation-and-study-sample-analysis.

ICH guidance for industry *E14 Clinical Evaluation of QT/QTc Interval Prolongation and Proarrhythmic Potential for Non-Antiarrhythmic Drugs* (October 2005), https://www.fda.gov/regulatory-information/search-fda-guidance-documents/e14-clinical-evaluation-qtqtc-interval-prolongation-and-proarrhythmic-potential-non-antiarrhythmic-0.

Jacobsson, S, D Golparian, RA Alm, M Huband, J Mueller, JS Jensen, M Ohnishi, and M Unemo, 2014, High in vitro activity of the novel spiropyrimidinetrione AZD0914, a DNA gyrase inhibitor, against multidrug-resistant *Neisseria gonorrhoeae* isolates suggests a new effective option for oral treatment of gonorrhea, Antimicrob Agents Chemother, 58(9):5585-5588, https://www.ncbi.nlm.nih.gov/pubmed/24982070.

Jacobsson, S, R Kularatne, R Kittiyaowamarn, V Maseko, P Paopang, P Sangprasert, P Sirivongrangson, L Piddock, T Wi, E Alirol, and M Unemo, 2019, High in vitro susceptibility to the first-in-class spiropyrimidinetrione zoliflodacin among consecutive clinical *Neisseria gonorrhoeae* isolates from Thailand (2018) and South Africa (2015-2017), Antimicrob Agents Chemother, 63(12), https://www.ncbi.nlm.nih.gov/pubmed/31548184.

Kern, G, T Palmer, DE Ehmann, AB Shapiro, B Andrews, GS Basarab, P Doig, J Fan, N Gao, SD Mills, J Mueller, S Sriram, J Thresher, and GK Walkup, 2015, Inhibition of *Neisseria gonorrhoeae* type II Topoisomerases by the novel spiropyrimidinetrione AZD0914, J Biol Chem, 290(34):20984-20994, https://www.ncbi.nlm.nih.gov/pubmed/26149691.

Le, W, X Su, X Lou, X Li, X Gong, B Wang, CA Genco, JP Mueller, and PA Rice, 2021, Susceptibility trends of Zoliflodacin against multidrug-resistant *Neisseria gonorrhoeae* clinical isolates in Nanjing, China, 2014 to 2018, Antimicrob Agents Chemother, 65(3), https://www.ncbi.nlm.nih.gov/pubmed/33318010.

Miller, AA, MM Traczewski, MD Huband, PA Bradford, and JP Mueller, 2019, Determination of MIC quality control ranges for the novel gyrase inhibitor Zoliflodacin, J Clin Microbiol, 57(9), https://www.ncbi.nlm.nih.gov/pubmed/31315953.

Papp, JR, K Lawrence, S Sharpe, J Mueller, and RD Kirkcaldy, 2016, In vitro growth of multidrug-resistant *Neisseria gonorrhoeae* isolates is inhibited by ETX0914, a novel spiropyrimidinetrione, Int J Antimicrob Agents, 48(3):328-330, https://www.ncbi.nlm.nih.gov/pubmed/27499432.

Quilter, LAS, SB St Cyr, and LA Barbee, 2024, The management of gonorrhea in the era of emerging antimicrobial resistance: what primary care clinicians should know, Med Clin North Am, 108(2):279-296, https://www.ncbi.nlm.nih.gov/pubmed/38331480.

Raccagni, AR, M Ranzenigo, E Bruzzesi, C Maci, A Castagna, and S Nozza, 2023, *Neisseria gonorrhoeae* antimicrobial resistance: the future of antibiotic therapy, J Clin Med, 12(24), https://www.ncbi.nlm.nih.gov/pubmed/38137836.

Su, XH, BX Wang, WJ Le, YR Liu, C Wan, S Li, RA Alm, JP Mueller, and PA Rice, 2016, Multidrug-resistant *Neisseria gonorrhoeae* isolates from Nanjing, China, are sensitive to killing by a novel DNA gyrase inhibitor, ETX0914 (AZD0914), Antimicrob Agents Chemother, 60(1):621-623, https://www.ncbi.nlm.nih.gov/pubmed/26482313.

Unemo, M, J Ahlstrand, L Sanchez-Buso, M Day, D Aanensen, D Golparian, S Jacobsson, MJ Cole, and European Collaborative Group, 2021, High susceptibility to zoliflodacin and conserved target (GyrB) for zoliflodacin among 1209 consecutive clinical *Neisseria gonorrhoeae* isolates from 25 European countries, 2018, J Antimicrob Chemother, 76(5):1221-1228, https://www.ncbi.nlm.nih.gov/pubmed/33564854.

Unemo, M, J Ringlander, C Wiggins, H Fredlund, S Jacobsson, and M Cole, 2015, High in vitro susceptibility to the novel spiropyrimidinetrione ETX0914 (AZD0914) among 873 contemporary clinical *Neisseria gonorrhoeae* isolates from 21 European countries from 2012 to 2014, Antimicrob Agents Chemother, 59(9):5220-5225, https://www.ncbi.nlm.nih.gov/pubmed/26077246.

WHO, 2025, Gonorrhoea (*Neisseria gonorrhoeae* infection), https://www.who.int/news-room/fact-sheets/detail/gonorrhoea-(neisseria-gonorrhoeae-infection).

Workowski, KA, LH Bachmann, PA Chan, CM Johnston, CA Muzny, I Park, H Reno, JM Zenilman, and GA Bolan, 2021, Sexually transmitted infections treatment guidelines, 2021, MMWR Recomm Rep, 70(4):1-187, https://www.ncbi.nlm.nih.gov/pubmed/34292926.

Yang, KJ, N Kojima, CC Bristow, and JD Klausner, 2023, Effectiveness of Cefixime for the treatment of *Neisseria gonorrhoeae infection* at 3 anatomic sites: A Systematic Review and Meta-Analysis, Sex Transm Dis, 50(3):131-137, https://www.ncbi.nlm.nih.gov/pubmed/36729626.

27. Review Team

Table 142. Reviewers of Integrated Assessment

Role	Name(s)
Regulatory project manager	Joseph Nguyen
Chief, regulatory project management staff(s)	Gregory DiBernardo; Trang Tran
Nonclinical reviewer	Leah Rosenfeld
Nonclinical team leader	Amy Nostrandt
OCP reviewer(s)	Tajhia Whigham; Jihye Ahn; Christina Won
OCP team leader(s)	Abhay Joshi; Jiang Liu; Yuching Yang
Clinical reviewer	Shayna Dooling
Clinical secondary reviewer	Jae Ho Hong
Biometrics reviewer	Jie Cong
Biometrics secondary reviewer	Daniel Rubin
Microbiology reviewer	Simone Shurland
Microbiology team leader	Avery Goodwin
Cross-discipline team leader; Clinical team leader	Ramya Gopinath
Deputy Division director (pharm/tox)	Terry Miller
Deputy Division director (OCP)	Zhixia Danielsen
Tertiary reviewer (OB)	Scott Komo
Associate Director for Labeling	Abimbola Adebowale
Director regulatory project management staff (DRO-ID)	Maureen Dillon Parker
Deputy division director (clinical)	Dmitri Iarikov
Division director (clinical)	Peter Kim
Office director (or designated signatory authority)	Adam Sherwat

Abbreviations: OCP, Office of Clinical Pharmacology; OB, Office of Biostatistics

Table 143. Additional Reviewers of Application

Office or Discipline	Name(s)
OPQ	Saki Golafale; Dorota Matecka (TL)
DPMH (Maternal Health Review)	Carrie Ceresa; Miriam Dinatale (TL)
OPDP	Qumerunnisa Syed; Sam Skariah (TL)
DMPP	Mary Carroll; Marcia Williams (TL)
OSI	John Lee; Phillip Kronstein (TL)
OSE/DEPI	Ikponmwosa Osaghae; Yan Li (TL)
OSE/DMEPA	Deborah Myers; Valerie Vaughan (TL)
OSE/DRISK	Brad Moriyama; Naomi Boston (TL)
OSE/HF	Tianyi Zhang; Oluwamurewa Oguntimein (TL)

Abbreviations: OPQ, Office of Pharmaceutical Quality; OPDP, Office of Prescription Drug Promotion; OSI, Office of Scientific Investigations; OSE, Office of Surveillance and Epidemiology; DEPI, Division of Epidemiology; DMEPA, Division of Medication Error Prevention and Analysis; DRISK, Division of Risk Management; DMPP, Division of Medical Policy Programs; DPMH, Division of Pediatric and Maternal Health; TL, Team Leader; HF, Human Factor

27.1. Reviewer Signatures

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Pharmacology Discipline Secondary Reviewer	Abhay Joshi OCP DIDP	Sections: 5, 6.1, 8.1, 8.2, 14.1, 14.2, 14.3	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Abhay Joshi

Digitally signed by Abhay Joshi

Date: 12/10/2025 5:41 PM EST
GUID: 20251210224125

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Discipline Tertiary Reviewer	Ramya Gopinath OID DAI	Sections: 1, 2, 3, 4, 6.3, 7, 8.3, 8.4, 10, 11, 12, 15, 17, 21, 22, 23, 24, 25, 26, 27	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Ramya Gopinath

Digitally signed by Ramya Gopinath

Date: 12/10/2025 5:51 PM EST
GUID: 20251210225159

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Discipline Primary Reviewer	Shayna Dooling OID DAI	Sections: 1, 2, 3, 4, 6, 7, 8, 10, 11, 15, 16, 17, 21, 22, 23, 24, 25	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Shayna Dooling

Digitally signed by Shayna Dooling

Date: 12/10/2025 6:09 PM EST
GUID: 2025121023920

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Microbiology Discipline Secondary Reviewer	Avery Goodwin OID DAI	Sections: 19, 20, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Avery Goodwin

Digitally signed by Avery Goodwin

Date: 12/11/2025 5:53 AM EST
GUID: 20251211105352

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Microbiology Discipline Primary Reviewer	Simone Shurland OID DAI	Sections: 19, 20, 23	Based on my assessment of the application: ☐ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ✓ Not applicable.	

Signature: Simone Shurland

Digitally signed by Simone Shurland

Date: 12/11/2025 6:30 AM EST
GUID: 2025121111304

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Regulatory Project Manager Discipline Primary Reviewer	Joseph Nguyen ORO DROID	Sections: 12	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Joseph Nguyen

Digitally signed by Joseph Nguyen

Date: 12/11/2025 8:02 AM EST
GUID: 2025121113254

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Pharm-tox/Non-clinical Discipline Tertiary Reviewer	Terry Miller OID DPTID	Sections: 3, 7, 8, 13, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: Terry Miller			**Digitally signed by Terry Miller** **Date: 12/11/2025 8:16 AM EST** **GUID: 20251211131613**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Pharmacology Discipline Primary Reviewer	Tajhia Whigham OCP DIDP	Sections: 5.1, 5.2, 6.1, 8.1, 8.2, 14.1, 14.2, 14.3	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: Tajhia Whigham			**Digitally signed by Tajhia Whigham** **Date: 12/11/2025 8:54 AM EST** **GUID: 20251211135421**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Biostatistics Discipline Primary Reviewer	Jie Cong OB DBIV	Sections: 2, 3, 6, 16, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: Jie Cong			**Digitally signed by Jie Cong** **Date: 12/11/2025 9:31 AM EST** **GUID: 20251211143125**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
PBPK Reviewer Discipline Primary Reviewer	Christina Won OCP DIDP	Sections: 14.5.2	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: Christina Won			**Digitally signed by Christina Won** **Date: 12/11/2025 9:41 AM EST** **GUID: 20251211144131**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Associate Director for Labeling Discipline Secondary Reviewer	Abimbola Adebowale OID DAI	Sections: 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: **Abimbola Adebowale**			**Digitally signed by Abimbola Adebowale** **Date: 12/11/2025 10:25 AM EST** **GUID: 20251211152538**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Associate Director for Labeling Discipline Primary Reviewer	Abimbola Adebowale OID DAI	Sections: 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: **Abimbola Adebowale**			**Digitally signed by Abimbola Adebowale** **Date: 12/11/2025 10:25 AM EST** **GUID: 20251211152538**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Associate Director for Labeling Discipline Tertiary Reviewer	Abimbola Adebowale OID DAI	Sections: 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: **Abimbola Adebowale**			**Digitally signed by Abimbola Adebowale** **Date: 12/11/2025 10:26 AM EST** **GUID: 2025121115269**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Pharmacometrics Reviewer Discipline Primary Reviewer	Jihye Ahn OCP DPM	Sections: 5, 6, 8, 14	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: **Jihye Ahn**			**Digitally signed by Jihye Ahn** **Date: 12/11/2025 10:33 AM EST** **GUID: 20251211153322**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Biostatistics Discipline Tertiary Reviewer	Scott Komo OB DBIV	Sections: 6, 15, 16, 23 - 26	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Scott Komo

Digitally signed by Scott Komo

Date: 12/11/2025 11:01 AM EST
GUID: 2025121116115

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Biostatistics Discipline Secondary Reviewer	Scott Komo OB DBIV	Sections: 6, 15, 16, 23-26	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Scott Komo

Digitally signed by Scott Komo
Sign on behalf of Signing for Daniel Rubin
Date: 12/11/2025 11:04 AM EST
GUID: 202512111647

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Regulatory Project Manager Discipline Secondary Reviewer	Gregory Dibernardo ORO DROID	Sections: 12	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Gregory Dibernardo

Digitally signed by Gregory Dibernardo

Date: 12/11/2025 11:10 AM EST
GUID: 20251211161044

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Pharm-tox/Non-clinical Discipline Primary Reviewer	Leah Rosenfeld OID DPTID	Sections: 2.2, 3, 7.1, 7.7, 8.4, 13, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Leah Rosenfeld

Digitally signed by Leah Rosenfeld

Date: 12/11/2025 11:32 AM EST
GUID: 20251211163220

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Pharmacology Discipline Tertiary Reviewer	Zhixia Danielsen OCP DIDP	Sections: 5, 6.1, 8.1, 8.2, 14	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: Zhixia Danielsen			**Digitally signed by Zhixia Danielsen** **Date: 12/11/2025 11:50 AM EST** **GUID: 20251211165044**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
CMC (OPQ/ONDP) Discipline Primary Reviewer	Dorota Matecka OPQAI DPQAIII	Sections: 9, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	Signing on behalf of the OPQ team
Signature: Dorota Matecka			**Digitally signed by Dorota Matecka** **Date: 12/11/2025 12:09 PM EST** **GUID: 20251211179 53**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
CMC (OPQ/ONDP) Discipline Secondary Reviewer	Dorota Matecka OPQAI DPQAIII	Sections: 9, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	Signing on behalf of the OPQ team

Signature: Dorota Matecka

Digitally signed by Dorota Matecka

Date: 12/11/2025 12:10 PM EST
GUID: 20251211171050

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Pharm-tox/Non-clinical Discipline Secondary Reviewer	Amy Nostrandt OID DPTID	Sections: 2.2, 3.1.2, 7.1, 7.7, 8.4, 13, 23	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Amy Nostrandt

Digitally signed by Amy Nostrandt

Date: 12/11/2025 12:47 PM EST
GUID: 20251211174755

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
PBPK Reviewer Discipline Secondary Reviewer	Yuching Yang OCP DPM	Sections: 8, 14	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Yuching Yang

Digitally signed by Yuching Yang

Date: 12/11/2025 1:10 PM EST
GUID: 20251211181010

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Pharmacometrics Reviewer Discipline Deputy Director (acting)	Jiang Liu OCP DPM	Sections: 5, 6, 7 , 8, 14	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	

Signature: Jiang Liu

Digitally signed by Jiang Liu

Date: 12/11/2025 1:43 PM EST
GUID: 20251211184349

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Clinical Discipline Secondary Reviewer	Jae Hong OID DAI	Sections: 1, 2, 3, 4, 6, 7 8, 10, 11, 15, 16, 17, 21, 22, 23, 24, 25	Based on my assessment of the application: ✓ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ☐ Not applicable.	
Signature: **Jae Hong**			**Digitally signed by Jae Hong** **Date: 12/11/2025 2:01 PM EST** **GUID: 2025121119127**	

Discipline and Role	Reviewer Name, Office/Center, and Division	Sections Authored in Full or in Part	Recommendation to Signatory	Comments on Recommendation to Signatory
Regulatory Project Manager Discipline Tertiary Reviewer	Maureen Dillon Parker ORO DRO-ID	Sections: 12	Based on my assessment of the application: ☐ No deficiencies preclude approval. ☐ Deficiencies preclude approval. ✓ Not applicable.	
Signature: **Maureen Dillon Parker**			**Digitally signed by Maureen Dillon Parker** **Date: 12/11/2025 2:57 PM EST** **GUID: 20251211195754**	

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

RAMYA GOPINATH
12/11/2025 02:33:15 PM

PETER W KIM
12/11/2025 03:06:58 PM

ADAM I SHERWAT
12/11/2025 05:13:43 PM