Tolerability and Pharmacokinetics of Contezolid at Therapeutic and Supratherapeutic Doses in Healthy Chinese Subjects, and Assessment of Contezolid Dosing Regimens Based on Pharmacokinetic/Pharmacodynamic Analysis

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ABSTRACT

Purpose: This study assessed the tolerability and pharmacokinetic (PK) properties of a new-generation oxazolidinone, contezolid (MRX-I), and its major inactive metabolite, M2, after single oral administrations of 800, 1200, and 1600 mg in the fed state, and compared the efficacy of 3 dosing regimens in the treatment of methicillin-resistant *Staphylococcus aureus* (MRSA) infection based on PK/pharmacodynamic (PD) analysis.

Methods: A Phase I study at a single study center was conducted with 2 parts. In the first part, 20 healthy subjects received a single oral dose of 1200 or 1600 mg of contezolid or placebo in the fed state in a double-blind, placebo-controlled, dose-escalation tolerance study. In the second part of the study, 52 subjects received a single oral dose of 800 mg of contezolid in the fed state in a single-center, randomized, blinded, 4-period, crossover, thorough QT study. Noncompartmental analyses were used to evaluate the PK properties of contezolid and M2. Steady-state concentrations of contezolid following the 3 dosing regimens (800, 1200, and 1600 mg q12h) were simulated by employing a newly developed 2-compartmental PK model. The minimum inhibitory concentration (MIC) distributions of contezolid were analyzed in 178 *Staphylococcus*, *Enterococcus*, and *Streptococcus* clinical isolates. Monte Carlo simulations were conducted to predict the efficacy of the 3 dosing regimens to obtain probability of target attainment and cumulative fraction of response.

Findings: Single-dose oral administrations of 800, 1200, and 1600 mg of contezolid were well tolerated in healthy subjects in the fed state, and nonlinear PK was observed. The mean plasma exposures to M2 exceeded 17.3% of contezolid exposure in the 3 groups. Both MIC_{50} and MIC_{90} (MICs that inhibit the growth of 50% and 90% of microorganisms, respectively) of contezolid against MRSA were 1 mg/L with clinical isolates from China. PK/PD analysis and Monte Carlo simulations predicted that 800 mg q12h of oral contezolid would be efficacious against MRSA infection, with a MIC of $\leq$4 mg/L (probability of target attainment, >90%; cumulative fraction of response, >90%).

Implications: Contezolid is a well-tolerated treatment option for MRSA infection, including at

Accepted for publication April 15, 2019
https://doi.org/10.1016/j.clinthera.2019.04.025
0149-2918/$ - see front matter

supratherapeutic doses up to 1600 mg. The regimen of 800 mg q12h could achieve efficacy in treating bacterial infection with MRSA. To our knowledge, this is the first PK study to predict that a dosing regimen of 800 mg q12h of oral contezolid is sufficient for treating MRSA infection, with a MIC of ≤4 mg/L. A Phase III study of this suggested dosing regimen is being conducted. Chinadrugtrials.org.cn identifier: CTR20161074. (*Clin Ther*. 2019;41:1164–1174) © 2019 Published by Elsevier Inc.

Keywords: contezolid, MRX-I, pharmacokinetic-pharmacodynamic, pharmacokinetics, supra-therapeutic doses, tolerability.

INTRODUCTION

Since methicillin-resistant *Staphylococcus aureus* (MRSA) infection was first reported in the 1960s, it has presented a major threat to human health worldwide.[1] Similar to methicillin-susceptible *S aureus* (MSSA), MRSA can affect the skin, soft tissues, respiratory system, bones, joints, endovascular system, and bloodstream.[2,3] According to the World Health Organization's Antimicrobial Resistance section of the Global Report on Surveillance in 2014, the prevalence of MRSA among *S aureus* exceeds 20%, and even surpasses 80% in some regions.[4] In contrast to the recent decline in hospital-associated MRSA, community-associated MRSA infection has emerged all over the world.[2] Furthermore, infection with MRSA causes significant economic burden and extends hospital stays.[5]

Vancomycin and linezolid are widely used for the treatment of MRSA infection.[6] However, the emergence of vancomycin-intermediate and -resistant *S aureus* have threatened current therapeutic options available for MRSA infection.[7] Linezolid, although effective in treating MRSA infection, has been associated with myelosuppression, which restricts use of the drug.[8] Therefore, there is an urgency to develop new, effective and-well tolerated antibacterial agents.

Contezolid (MRX-I), (*S*)-5-([isoxazol-3-ylamino] methyl)-3-(2,3,5-trifluoro-4-[4-oxo-3,4-dihydropyridin-1(2H)-yl]phenyl)oxazolidin-2-one, is a new antibacterial oxazolidinone with activity against gram-positive bacteria, such as staphylococci, streptococci, and enterococci, including MRSA, methicillin-resistant *Streptococcus epidermidis (MRSE)*, penicillin-resistant *Streptococcus pneumoniae (PRSE)*, and vancomycin-resistant enterococci (VRE).[9] Similar to that of linezolid, the antibacterial effect of contezolid is mediated by binding to a distinct region of 23S RNA adjacent to the peptidyl transferase center of the 50S ribosomal subunit. In a study in a murine model of systemic and neutropenic infection of the thigh, contezolid showed similar or better efficacy than that of linezolid against MRSA, methicillin-resistant *Streptococcus epidermidis*, penicillin-resistant *Streptococcus pneumoniae*, and VRE.[10] A major potential advantage of contezolid over linezolid is a significantly improved safety profile, with minimal myelosuppression and monoamine oxidase inhibition.[9] The pharmacokinetic (PK) properties of oral contezolid 200 to 1800 mg in the fasting state have been previously investigated in healthy Chinese subjects[11]; dose response from 300 to 900 mg was not linear in fasted subjects, and about 2% of MRX-I was excreted via kidneys in an unchanged form. The principal metabolic pathway of MRX-I in humans involves opening of the 2,3-dihydropyridin-4-one ring and generating metabolites in the plasma and urine.[12] Recently, 2 Phase II studies of contezolid (ClinicalTrials.gov identifier: NCT02269319; Chinadrugtrials.org.cn identifier: CTR20140056) demonstrated that the clinical efficacy of contezolid 800 mg q12h was similar to that of linezolid in treating severe skin and skin structure infections. The tolerability and efficacy of contezolid in treating complicated skin and soft tissue infections (cSSTIs) are being evaluated in an ongoing Phase III trial in China (Chinadrugtrials.org.cn identifier: CTR20150855).

Although food has a significant effect on the exposure to contezolid,[11,13] the tolerability and PK properties of oral contezolid 1200 and 1600 mg in the fed state have not been determined. Furthermore, 13 metabolites of contezolid have been detected in plasma, urine, and feces,[13] although it is believed that they have no antibacterial activity (personal communication in October, 2018, MicuRx). Among these, M2 is the main metabolite, and the plasma exposures to M2 were 24.0% of the contezolid exposures.[12] According to the US Food and Drug Administration's guidance on the tolerability testing of drug metabolites,[14] the tolerability and PK properties of M2 in humans should be evaluated.

As a key component of developing new antibacterial agents, optimization of PK/

pharmacodynamic (PD) properties is crucial in selecting the appropriate therapeutic dose to achieve clinical efficacy and to minimize the emergence of resistance.[15] The AUC/MIC ratio was identified as the most predicted PK/PD index of contezolid against *S aureus*.[13] Hence, it is vital to evaluate the efficacy of contezolid in different dose regimens during development.

In the present study, we investigated the safety and PK profiles of contezolid and its major metabolite M2 in healthy Chinese subjects in the fed state after oral administration of therapeutic and supratherapeutic dosage regimens. Furthermore, PK/PD analysis was conducted to evaluate the probability of target attainment (PTA) and cumulative fraction of response (CFR) with 3 contezolid dosage regimens against MRSA. Finally, the standard dosage regimen (800 mg q12h) was proposed for clinical application.

SUBJECTS AND METHODS

Study Design

In the first part of the Phase I study conducted at a single study center, 1200 and 1600 mg cohorts of contezolid* were evaluated in a double-blind, placebo-controlled manner. Both cohorts enrolled 10 subjects (8 contezolid, 2 matching placebo) and were balanced with respect to sex. In the second part of the study, 52 healthy subjects (26 women and 26 men) were randomized into a 4-period, crossover, thorough QT evaluation in which each subject received 800 and 1600 mg of contezolid and a matching placebo in a blinded manner, and 400 mg of open-label moxifloxacin (study drug dosing was separated by 7-day washout periods). All of the participants were provided a standardized breakfast before study drug administration. The study protocol was approved by the ethics committee of Huashan Hospital, Fudan University, and conducted according to the Good Clinical Practice standards. All of the subjects provided written informed consent before participating in the study.

* Contezolid 400-mg tablets were manufactured by Zhejiang Huahai Pharmaceutical Co Ltd (Zhejiang, China) and distributed by MicuRx Pharmaceuticals Inc (catalog number 20161223; Shanghai, China).

Subjects

Healthy women and men aged 18 to 45 years, with a body mass index that ranged from 18 to 26 kg/m^2, were recruited. For female subjects, serum pregnancy tests were performed at screening and also on the day prior to study drug dosing. Subjects were excluded if they had significant cardiac, pulmonary, neurologic, hepatic, renal, gastrointestinal, hematologic, and/or psychiatric disease as assessed by medical history, physical examination, and laboratory tests. Subjects who abused alcohol, nicotine, or illicit drugs were excluded. Also excluded were potential subjects who participated in any other investigational drug trial within the previous 1 month, or donated blood (>500 mL) within the previous 3 months, and those who ate grapefruit or drank grapefruit juice within the previous 7 days. If the ECG screens of potential subjects showed a QTcF of >450 ms, a QRS interval of >110 ms, a PR interval of >210 ms, or second- or third-degree atrioventricular block, they were also excluded.

LC-MS/MS Assays

Plasma concentrations of contezolid and M2 were determined by a validated ultra-performance LC-MS/MS assay using Acquity UPLC BEH C_8 (2.1 × 100 mm, 1.7 μm internal diameter; Waters, Milford, MA). Gradient elution was performed to separate drug using acetonitrile and water as the mobile phase at a flow rate of 0.4 mL/min. Internal standards were D5-MRX-I and linezolid for contezolid and M2, respectively. There were no matrix effects, and the recovery of both contezolid and M2 exceeded 90%. Contezolid and M2 were stable for 24 hours at room temperature, and were also stable at −20°C for 5 months and at −40°C for 10 months.

Noncompartmental and Compartmental Analysis of PK Properties

Plasma samples for PK analysis were collected at 2 hours before dosing, and at 0.25, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 16, and 23.5 hours after dosing. Noncompartmental analysis was used to evaluate the PK properties of contezolid and M2, using WinNonlin software version 6.3 (Pharsight Corporation, Mountain View, CA).

Dose proportionalities of AUC and C_{max} were evaluated using a power model. The model was calculated using the following equation[16,17]:

$$\ln(PK) = \alpha + \beta \cdot \ln(\text{Dose}) \quad \text{(Equation 1)}$$

The linear reference interval was calculated as:

$$1 + \ln(Q_L)/\ln(r), 1 + \ln(Q_U)/\ln(r), \quad \text{(Equation 2)}$$

where $Q_L = 0.8$ and $Q_U = 1.25$; r was the ratio of the highest dose and lowest dose. In the present study, $r = 2$. The properties were in proportion to dose if the 95% CI of regression coefficient β was completely within the reference interval.

In addition, a 2-compartment model with first-order elimination and 4 absorption transit compartments by WinNonlin was employed to analyze drug-concentration data, and 3 doses (800, 1200, and 1600 mg) using the same model were simulated with different initial values. The steady-state concentrations of 3 dosing regimens were simulated by the PK modeling.

Susceptibility Study

A total of 178 clinical isolates of staphylococci, enterococci, and streptococci were collected from 30 hospitals in different provinces of China in 2015−2017. Contezolid had a purity of 99.5%. Following the recommendations of the Clinical and Laboratory Standards Institute, the minimum inhibitory concentration (MIC) of contezolid was determined by the microbroth dilution method, and linezolid and vancomycin were used as reference agents. Clinical isolates were grown on cation adjusted-Mueller Hinton broth (catalog number 6117994; Becton, Dickinson, and Company, Franklin Lakes, NJ). Hemolytic streptococci were grown on cation adjusted-Mueller Hinton broth containing 10% lysed horse blood (catalog number QL13, RA2903G; Wenzhou Kangtai Biotech Co Ltd, Zhejiang, China). The results were evaluated by current Clinical and Laboratory Standards Institute interpretive criteria. *S aureus* ATCC 29213, *Enterococcus faecalis* ATCC 29212, and *Streptococcus pneumoniae* ATCC 49619 were used as control strains.

Monte Carlo Simulation

A Monte Carlo simulation was performed with 5000 subjects using MATLAB software version 7.0.1 (MathWorks Inc, Natick, MA) to obtain the PTA and CFR. The steady-state PK properties were calculated by the 2-compartment model mentioned earlier. The simulated dosages of contezolid were 800, 1200, and 1600 mg q12h. As introduced previously, free drug (f)-AUC_{0-24h}/MIC was the PD index that best correlated with efficacy in mouse models of thigh infection, and an $fAUC_{0-24h}$/MIC of 2.3 was required for the static effect (personal communication in O2018, MicuRx). Generally, a dosing regimen with a PTA or CFR of >90% is regarded as optimal for treating infections caused by a specific microorganism.

Statistical Analysis

Results are expressed as mean (SD). The t test and ANOVA were used to compare the PK properties of different cohorts. $P < 0.05$ was considered statistically significant.

RESULTS

Tolerability and Tolerability

Demographic data of the study participants are presented in Table I, and no significant differences in any aspects between the different treatment groups were observed. All of the participants completed the study and no serious adverse events were reported (Table II). All of the adverse events were mild and most were gastrointestinal disorders. After a single oral dose of 800 mg of contezolid, 7 mild drug-related adverse events were reported in 9.6% (5/52) of the subjects. In the 1200 mg cohort, neutrophil counts were decreased mildly in 12.5% (1/8) of the subjects, while in the 1600 mg cohort, nausea and epigastric discomfort occurred in 12.5% (1/8) of subjects. All of the adverse events resolved quickly and without intervention.

There were no QT or QTcF intervals that exceeded 500 ms or increases in QT or QTcF intervals of >60 ms compared with baseline after single oral administrations of contezolid and placebo in either of the cohorts. Moreover, no morphologic changes considered clinically significant were observed in the placebo or contezolid groups.

PK Properties

Figure 1 shows the plasma concentration−time profile of contezolid and its inactive metabolite M2 after single oral doses of 800, 1200, and 1600 mg of contezolid in the fed state. The PK properties of contezolid and M2 in each cohort are presented in Table III. $t_{1/2}$ of contezolid after a single dose was

Table I. Summary of demographics.

Parameter	800 mg (n = 52)	1200 mg (n = 8)	1600 mg (n = 8)	Placebo (n = 4)
Sex, no. (%)				
Female	26 (50)	4 (50)	4 (50)	2 (50)
Male	26 (50)	4 (50)	4 (50)	2 (50)
Age, mean (SD), y	26.1 (4.74)	26.4 (5.22)	24.1 (1.36)	29.0 (4.06)
Height, mean (SD), cm	164.9 (7.73)	163.6 (5.91)	163.1 (6.06)	166.2 (7.22)
Weight, mean (SD), kg	59.6 (7.64)	56.2 (8.64)	57.8 (5.07)	60.5 (9.49)
BMI, mean (SD), kg/m^2	21.9 (1.80)	20.9 (1.94)	21.7 (1.60)	21.8 (1.67)

BMI = body mass index.

Table II. Drug-related treatment-emergent adverse events (AEs) after single-dose administration of oral contezolid in healthy Chinese subjects.

Variable	800 mg (n = 52)	1200 mg (n = 8)	1600 mg (n = 8)	Placebo (n = 4)
Subjects with at least 1 drug-related AE	5 (9.6)	1 (12.5)	1 (12.5)	0
Clinical	3 (5.8)	0	1 (12.5)	—
Nausea	2 (3.8)	—	1 (12.5)	—
Epigastric discomfort	1 (1.9)	—	1 (12.5)	—
Diarrhea	1 (1.9)	—	—	—
Vomiting	1 (1.9)	—	—	—
Laboratory	2 (3.8)	1 (12.5)	0	—
Elevated blood uric acid	2 (3.8)	0	—	—
Depressed neutrophil count	—	1 (12.5)	—	—

AEs = adverse events.

slightly increased with the increased amounts of contezolid administered ($P < 0.05$). The mean (SD) plasma concentrations at 12 hours after dosing with 800, 1200, and 1600 mg were 0.59 (0.50), 1.69 (1.70) and 1.1 (0.49) mg/L, respectively. The mean plasma exposures to M2 following single oral administrations of 800, 1200, and 1600 mg of contezolid were 23.5%, 17.3%, and 22.0% of contezolid plasma exposures, respectively. Linearity analysis showed that the 95% CI of slope β for $AUC_{0-\infty}$ of contezolid was 1.168 to 1.691, while the linear reference interval was 0.678 to 1.322. The 95% CI of slope β for the C_{max} of contezolid was 0.960 to 1.405. As the 95% CI of the slope for C_{max} spanned the reference interval, dose proportionality was considered to be inconclusive.[16] Hence, additional data are needed to address the PK proportionality of contezolid in healthy Chinese subjects after oral administration.

MIC Distribution of Contezolid Against Staphylococci, Enterococci, and Streptococci

Contezolid displayed excellent *in vitro* activity against clinical isolates of staphylococci, enterococci, and streptococci; the MIC distributions are shown in Table IV. The MIC_{50} and MIC_{90} (MICs to inhibits the growth of 50% and 90% of organisms, respectively) of contezolid, linezolid, and vancomycin against staphylococci, enterococci, and streptococci are displayed in Table V. The MIC_{50} and MIC_{90} of contezolid against MRSA were both 1 mg/L. The MIC_{90}s of contezolid against MSSA and MRSA (both, 1 mg/L) were half of those of linezolid (both, 2 mg/L). Figure 2 shows the MIC

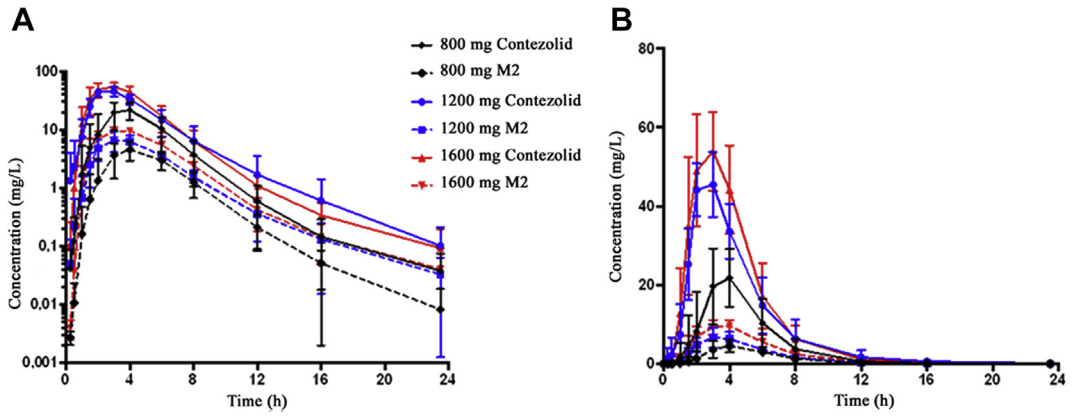

Figure 1. Mean (SD) plasma concentration–time profiles of contezolid and its major inactive metabolite, M2, in semi-log (A) and linear (B) scales for the *y* axis.

Table III. Pharmacokinetic properties of mean contezolid and metabolite M2 in healthy Chinese subjects. Data are given as mean (SD).

Compound/ Parameter	800 mg (n = 52)	1200 mg (n = 8)	1600 mg (n = 8)
MRX-I			
C_{max}, μg/mL	26.5 (6.06)	47.1 (6.96)	56.3 (6.98)
$AUC_{0\text{-}23.5}$, h · μg/mL	96.8 (26.3)	205.7 (56.5)	238.5 (49.1)
$AUC_{0-\infty}$, h · μg/mL	96.9 (26.2)	206.1 (56.8)	238.9 (49.1)
$t_{1/2}$, h	1.91 (0.77)	2.46 (0.97)	2.68 (0.92)
T_{max}, h	3.71 (1.10)	2.50 (0.53)	2.5 (0.71)
Vz/F, L	24.5 (13.1)	22.8 (15.1)	26.9 (10.7)
CL/F, L/h	8.83 (2.29)	6.19 (1.62)	6.98 (1.57)
M2			
C_{max}, μg/mL	5.13 (1.42)	6.83 (2.26)	10.2 (1.46)
$AUC_{0-23.5}$, h · μg/mL	22.8 (4.79)	35.5 (6.65)	52.4 (10.3)
$AUC_{0-\infty}$, h · μg/mL	22.8 (4.78)	35.7 (6.59)	52.7 (10.4)
$t_{1/2}$, h	2.16 (0.60)	2.90 (1.42)	3.05 (1.20)
T_{max}, h	4.14 (1.05)	3.38 (0.52)	3.38 (0.74)

distributions of contezolid and linezolid against staphylococci, which indicate that contezolid exhibits more potent antibacterial activity than does linezolid. The MIC_{50} and MIC_{90} of contezolid against methicillin-susceptible and -resistant coagulase-negative staphylococci were 1 mg/L. The MIC_{50} of contezolid against enterococci was 1 mg/L and MIC_{90s} were 1 or 2 mg/L; these values were similar to or less than those of linezolid. Moreover, contezolid exhibited good antibacterial activity against streptococci, with MIC_{50} and MIC_{90} similar to or less than those of linezolid.

PK/PD Properties

A 2-compartment model with first-order elimination and 4 absorption transit compartments well described the PK properties of contezolid at all 3 doses (see Supplemental Figure 1 and Supplemental Table 1 in

Table IV. Activity of contezolid against staphylococci, enterococci, and streptococci isolated in China, 2015–2017. Data are given as the percentage of strains.

Organism	MIC							
	≤0.015 mg/L	0.03 mg/L	0.06 mg/L	0.125 mg/L	0.25 mg/L	0.5 mg/L	1 mg/L	2 mg/L
Staphylococci								
MRSA (n = 45)	0	0	0	0	2.2	33.4	64.4	0
MSSA (n = 35)	0	0	0	0	0	31.4	68.6	0
MRCNS (n = 16)	0	0	0	0	0	31.3	68.7	0
MSCNS (n = 13)	0	0	0	0	0	46.2	46.2	7.6
Enterococci								
E faecalis (n = 15)	0	0	0	0	0	0	73.3	26.7
E faecium (n = 15)	0	0	0	0	6.7	6.7	80.0	6.6
Streptococci								
S pyogenes (n = 10)	10.0	0	0	0	0	70.0	20.0	0
S agalactiae (n = 10)	0	0	0	0	0	0	100.0	0
S dysgalactiae (n = 9)	0	0	0	0	0	55.6	44.4	0
S anginosus (n = 10)	0	0	0	0	10.0	40.0	40.0	10.0

MIC = minimal inhibitory concentration; MRCNS = methicillin-resistant coagulase-negative staphylococci; MRSA = methicillin-resistant *Staphylococcus aureus*; MSCNS = methicillin-susceptible coagulase-negative staphylococci; MSSA = methicillin-susceptible *Staphylococcus aureus*.

Table V. MIC_{50} and MIC_{90} of contezolid, linezolid and vancomycin against staphylococci, enterococci, and streptococci. Data are given as milligrams per liter.

Organism (n)	MIC_{50}			MIC_{90}		
	MRX-I	Linezolid	Vancomycin	MRX-I	Linezolid	Vancomycin
Staphylococci						
MRSA (n = 45)	1	1	1	1	2	1
MSSA (n = 35)	1	2	1	1	2	1
MRCNS (n = 16)	1	1	1	1	2	2
MSCNS (n = 13)	1	1	1	1	1	2
Enterococci						
E faecalis (n = 15)	1	1	1	2	2	2
E faecium (n = 15)	1	1	1	1	2	2
Streptococci						
S pyogenes (n = 10)	0.5	0.5	0.25	1	1	0.5
S agalactiae (n = 10)	1	1	0.5	1	1	0.5
S dysgalactiae (n = 9)	0.5	1	0.25	1	1	0.25
S anginosus (n = 10)	0.5	0.5	0.5	1	1	0.5

MIC = minimal inhibitory concentration; MRCNS = methicillin-resistant coagulase-negative staphylococci; MRSA = methicillin-resistant *Staphylococcus aureus*; MSCNS = methicillin-susceptible coagulase-negative staphylococci; MSSA = methicillin-susceptible *Staphylococcus aureus*.

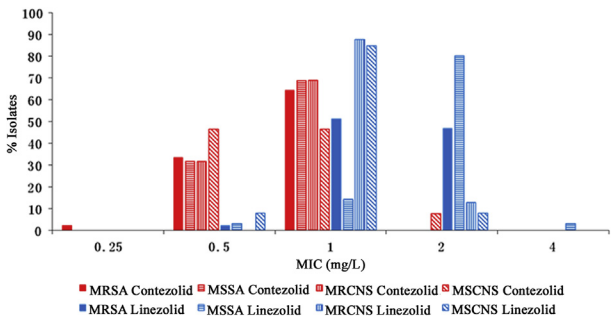

Figure 2. Contezolid and linezolid minimum inhibitory concentration (MIC) distributions for a collection of 109 staphylococci isolates in China. MRSA, methicillin-resistant *Staphylococcus aureus*; MSSA, methicillin-susceptible *Staphylococcus aureus*; MRCNS, methicillin-resistant coagulase-negative staphylococci; MSCNS, methicillin-susceptible coagulase-negative staphylococci.

the online version at https://doi.org/10.1016/j.clinthera.2019.04.025). Observed contezolid plasma concentrations versus predicted contezolid concentrations showed an adequate fit of the 2-compartment model (see Supplemental Figure 2 in the online version at https://doi.org/10.1016/j.clinthera.2019.04.025). Steady-state exposures (AUC_{0-24h}) of contezolid were simulated using the developed model for different dosage regimens (800, 1200, and 1600 mg q12h); concentration–time profiles are shown in Figure 3.

Monte Carlo simulations were conducted to assess the PTAs of 3 contezolid dosage regimens. Figure 4 shows that 800 mg q12h of contezolid should be efficacious against infections caused by MRSA, with a MIC of ≤4 mg/L (PTA, >90%). As the MIC_{90s} for MRSA, MSSA, and methicillin-susceptible and -resistant coagulase-negative staphylococci were 1 mg/L, estimated CFR values were >99.9% with all 3 dosage regimens (800, 1200, and 1600 mg q12h) against MRSA.

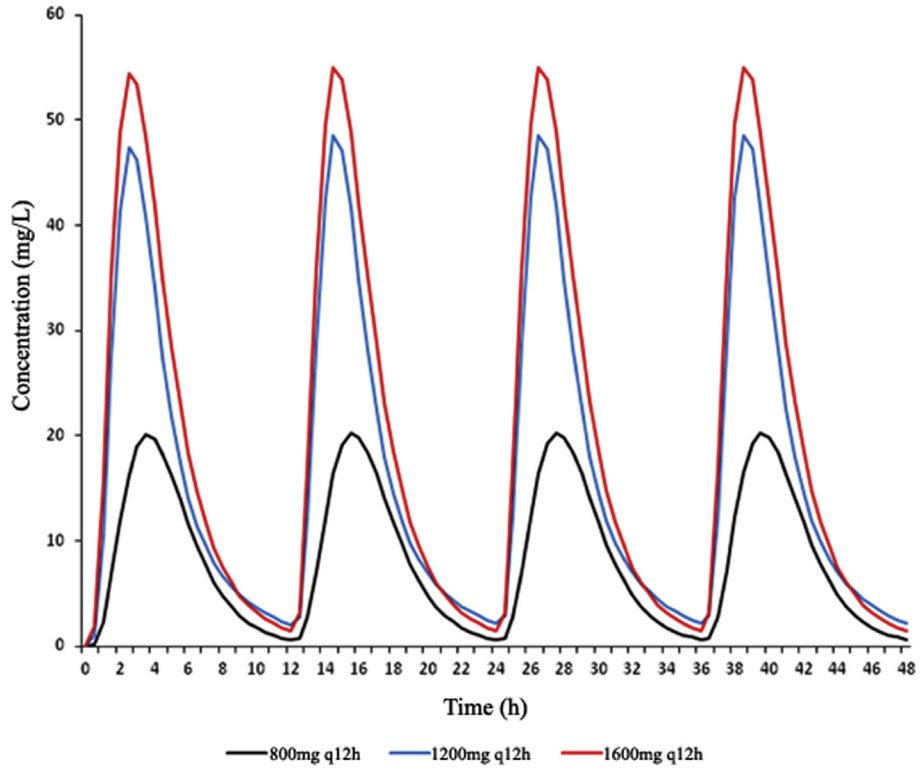

Figure 3. Predictive steady-state plasma concentrations of contezolid at different dosing regimens.

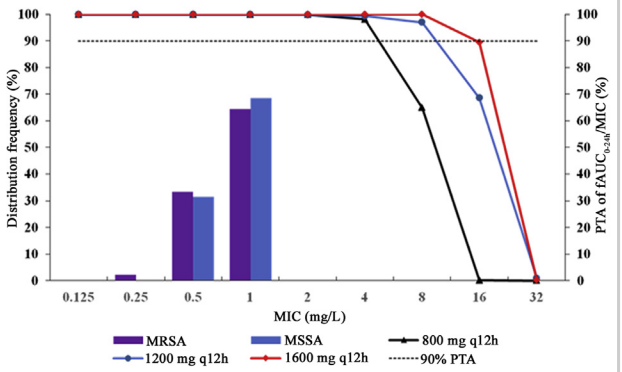

Figure 4. Probability of target attainment (PTA) of *f*AUC$_{0-24}$/minimum inhibitory concentration (MIC) for contezolid following oral administration of 800, 1200, and 1600 mg q12h. Target fAUC$_{0-24}$/MIC was 2.3. MRSA, methicillin-resistant *Staphylococcus aureus*; MSSA, methicillin-susceptible *Staphylococcus aureus*.

DISCUSSION

This is the first study demonstrating the favorable safety and PK profiles of contezolid, a new-generation oxazolidinone, and its major metabolite, M2, at supratherapeutic doses in the fed state in healthy Chinese subjects. Single-dose oral administrations of contezolid of 800, 1200, and 1600 mg were well tolerated, and the most common adverse events were gastrointestinal disorders; these findings are consistent with those from previous Phase I studies (MRX-I-01,[11] MRX-I-02[13]). Drug-related adverse events were mild, and no serious adverse events or deaths were reported. Both MIC$_{50}$ and MIC$_{90}$ of contezolid against MRSA were 1 mg/L. PK/PD analysis showed that contezolid 800 mg q12h can treat infection with MRSA with a MIC of ≤4 mg/L.

The results of a previous Phase I clinical trial showed that contezolid was absorbed rapidly from the intestines, and that a fat-rich diet could significantly increase exposure to the drug.[11] Nonlinear kinetics were observed when dosing >800 mg in the fasting state.[11] Linear kinetics of contezolid in the fed state, however, had not been determined in the previous study. In the present study, we found that the increase in AUC$_{0-\infty}$ was not proportional in the dose range from 800 to 1600 mg in the fed state. The C$_{max}$ and AUC$_{0-\infty}$ observed in the 800 mg cohort in the present study were not statistically different from those reported in the previous Phase I multiple-dose study of 800 mg q12h.[11]

Contezolid is extensively metabolized in the body, with 9 metabolites identified in humans.[12] M2 is the most prominent metabolite in plasma and urine, is also found in feces,[12] and is inactive *in vitro*. The biotransformation of contezolid to M2 is dependent on the oxidative ring opening of 2,3-dihydropyridin-4-one in humans and is related to the Fenna-Matthews-Olson 5 enzyme.[12] Urinary clearance of unchanged contezolid in humans was <3% of the administered dose,[11] while M2 was the main metabolite detected in urine.[12] In addition, the ratio of the plasma exposure of M2 to that of contezolid was similar, ranging from 800 to 1600 mg, which demonstrated that the transformation of contezolid to M2 was independent of the dose of contezolid.

In order to evaluate the steady-state exposures of oral 800, 1200, or 1600 mg of contezolid, a 2-compartment PK model was conducted to predict the plasma contezolid concentrations following 800, 1200, and 1600 mg q12h dosing regimens. As food has a significant effect on the absorption of contezolid, we constructed a 4-transit absorption compartment to fit the absorption phase, which provided a more favorable fit than did the lag-compartment model (see Supplemental Figure 3 in the online version at https://doi.org/10.1016/j.clinthera.2019.04.025). Minimal drug accumulation was observed with different dosing regimens.

A study in the United States with 606 *S aureus* isolates showed that the MIC$_{50}$ and MIC$_{90}$ of contezolid against *S aureus* were 0.5 and 1 mg/L, respectively.[18] In the present study using clinical isolates in China, contezolid MICs were similar to those in the US study, and contezolid (MIC$_{90}$, 1 mg/L) was slightly more potent than linezolid (MIC$_{90}$, 2 mg/L) against MRSA, MSSA, and methicillin-resistant coagulase-negative staphylococci. The MIC$_{50}$ and MIC$_{90}$ of contezolid and linezolid against hemolytic streptococci were similar, although vancomycin MIC$_{50}$ and MIC$_{90}$ were lower. In addition to staphylococci and streptococci, enterococci have also been identified as an important cause of cSSTI,[19] and the emergence of VRE around

the world limits treatment options.[20] In the present study, contezolid was shown to exhibit antibacterial activity against enterococci that was greater than or similar to that of linezolid or vancomycin, and hence would be a promising future treatment of VRE infection. Linezolid may cause reversible myelosuppression in patients receiving treatment for >2 weeks,[21] and contezolid may be a more well-tolerated option for treating cSSTI and other infections caused by MRSA.

The PK/PD optimization of antibiotic dosage regimens is a crucial topic in the development of new drugs to optimize efficacy and reduce the emergence of drug resistance.[15] Hence, we evaluated the PTA and CFR values with different dosages of contezolid against MRSA using Monte Carlo simulations. The MIC_{90} of contezolid against MRSA was 1 mg/L, and the results demonstrate that the dosage regimen of 800 mg q12h, which is the therapeutic dosage in the ongoing Phase III clinical trial, can achieve adequate efficacy in the treatment of MRSA infection, with a MIC of $\leq$4 mg/L. The findings from a Phase II randomized, double-blind clinical trial in patients with cSSTI in China showed that the efficacy of 7 to 14 days of oral contezolid 800 mg BID was similar to that of 7 to 14 days of oral linezolid 600 mg BID. Collectively, these results support oral contezolid 800 mg q12h as an effective therapy in the treatment of cSSTIs, including those caused by MRSA. In addition, as the PK/PD index of contezolid was attained from a murine model of *S aureus* infection of the thigh, other gram-positive infection models are warranted to predict the efficacy of contezolid against other microorganisms. Thus far, PK and susceptibility studies have all been from China; studies in other ethnicities are needed. The results of the present study suggest that 800 mg q12h of contezolid should be an effective treatment for MRSA infection.

CONCLUSIONS

Due to its tolerability and potent antibacterial activity, the new-generation oxazolidinone contezolid could be valuable in treating infection with MRSA. The results indicate in-depth that a 800 mg q12h dosage regimen could attain sufficient efficacy against MRSA infection. Finally, the 2-compartment PK model will provide a reference option for the construction of a population PK model for contezolid.

CONFLICTS OF INTEREST

This research were sponsored in part by MicuRx Pharmaceuticals, which provided the study design reference standards and the investigational drug.

H. Yuan is an employee of MicuRx Pharmaceuticals and a PhD student at Fudan University. The authors have indicated that they have no other conflicts of interest with regard to the content of this article.

ACKNOWLEDGMENTS

The authors thank the sponsor of the study for study design reference standards and providing the investigational drug. This study was financially supported by Major Research and Development Project of Innovative Drugs, Ministry of Science and Techology of China (2017ZX09304005) and Jing Zhang was responsible for the project.

J. Zhang managed the clinical trial process. J. Wu contributed subject recruitment, sample and data collection, statistical analysis, and writing of the manuscript. H. Wu, Y. Wang, and B. Guo contributed sample measurements. Y. Chen contributed PK analysis, PK modeling, and PK/PD analysis. G. Cao, X. Wu, and J. Yu contributed to the study design and trial conduction. J. Wu contributed clinical observation as a physician. D. Zhu, F. Hu, and Y. Guo contributed to the *in vitro* susceptibility studies. H. Yuan contributed to the study design and analysis review. All of the authors approved the manuscript for submission and publication.

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APPENDIX

Table S1. Parameter of two-Compartment model.

Parameter	Mean (SD)		
	800 mg	1200 mg	1600 mg
tvV (L)	7.23 (3.63)	6.03 (1.89)	8.25 (4.84)
tvK_{tr} (h^{-1})	1.51 (0.70)	2.19 (0.38)	2.54 (0.89)
tvK_a (h^{-1})	1.51 (0.70)	2.19 (0.38)	2.54 (0.89)
tvK_e (h^{-1})	1.19 (0.39)	1.13 (0.44)	0.96 (0.33)
tvK_{12} (h^{-1})	0.20 (0.23)	0.94 (0.70)	1.11 (0.79)
tvK_{21} (h^{-1})	0.09 (0.24)	0.95 (0.73)	1.65 (0.48)
ε	2.53 (0.89)	2.29 (1.22)	3.51 (1.83)

tvV: typical value of volume of central compartment; tvKtr: typical value of transit absorption rate constant; tvKa: typical value of absorption rate constant; tvKe: typical value of elimination rate constant; tvK12: typical value of intercompartment transfer rate constant from central to peripheral compartments; tvK21: typical value of intercompartment transfer rate constant from peripheral to central compartments; ε: residual error; SD: standard deviation.

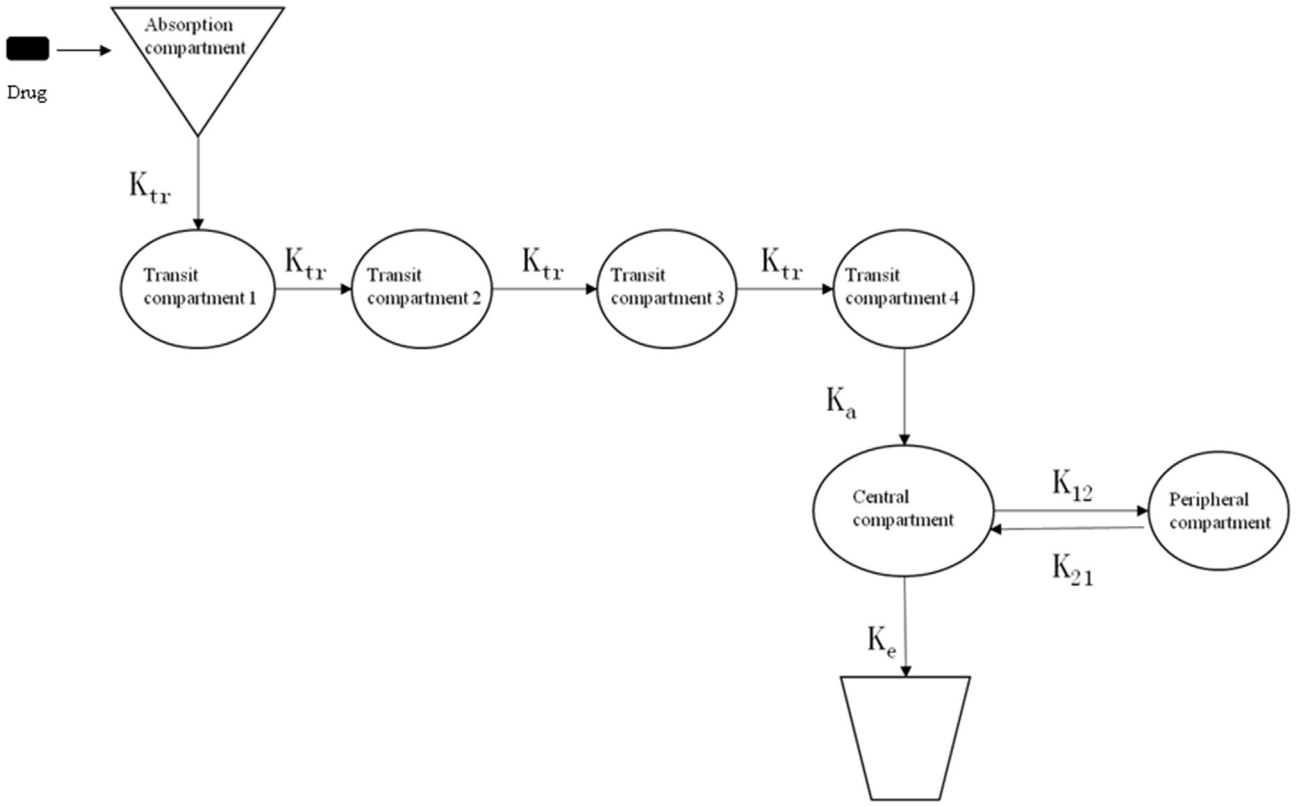

Figure S1. The schematic of the A two-compartment model with four absorption transit compartments and first-order elimination.

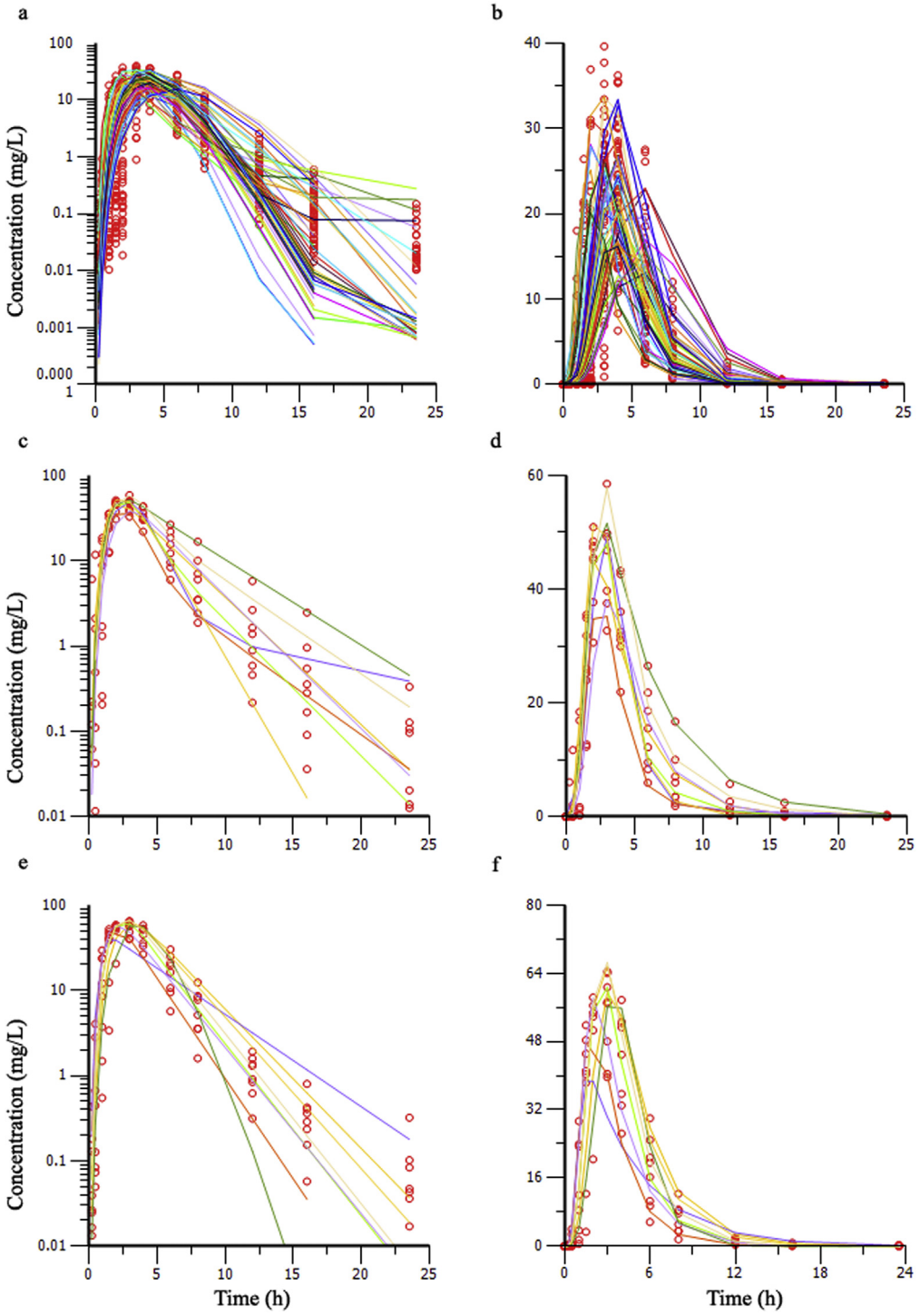

Figure S2. Observed contezolid plasma concentrations versus predicted contezolid concentrations. a,b: 800 mg cohort; c,d: 1200 mg cohort; e,f: 1600 mg cohort. a, c, e: in a semi-log scale for the y axis; b, d, f : in a linear scale for the y axis. The red open circles represent the observed contezolid plasma concentrations; the solid lines mean the predicted contezolid plasma contentrations. The different colors of lines in a-f represent different subjects.

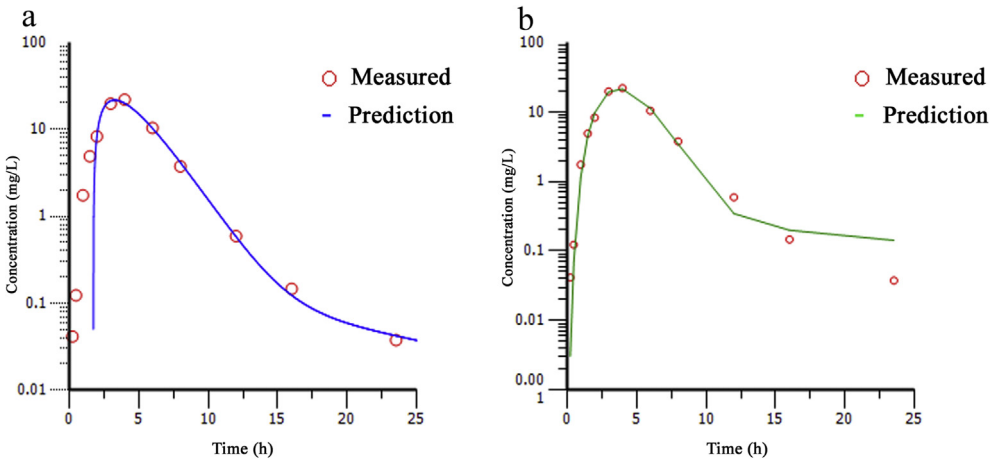

Figure S3. Simulated contezolid plasma concentrations (blue and green solid lines) from two-compartmental model fitted to mean measured concentrations of contezolid (red open circles). a: Two-compartmental model with with first-order elimination and T-lag time; b: A two-compartment model with first-order elimination and transit compartment absorption consisting of four transit compartments.