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# Applicability of a Single Time Point Strategy for the Prediction of Area Under the Concentration Curve of Linezolid in Patients: Superiority of $C_{trough}$- over $C_{max}$-Derived Linear Regression Models

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**Abstract**

*Background and Objectives* Linezolid, a oxazolidinone, was the first in class to be approved for the treatment of bacterial infections arising from both susceptible and resistant strains of Gram-positive bacteria. Since overt exposure of linezolid may precipitate serious toxicity issues, therapeutic drug monitoring (TDM) may be required in certain situations, especially in patients who are prescribed other co-medications.

*Methods* Using appropriate oral pharmacokinetic data (single dose and steady state) for linezolid, both maximum plasma drug concentration ($C_{max}$) versus area under the plasma concentration–time curve (AUC) and minimum plasma drug concentration ($C_{min}$) versus AUC relationship was established by linear regression models. The predictions of the AUC values were performed using published mean/median $C_{max}$ or $C_{min}$ data and appropriate regression lines. The quotient of observed and predicted values rendered fold difference calculation. The mean absolute error (MAE), root mean square error (RMSE), correlation coefficient ($r$), and the goodness of the AUC fold prediction were used to evaluate the two models.

*Results* The $C_{max}$ versus AUC and trough plasma concentration ($C_{trough}$) versus AUC models displayed excellent correlation, with $r$ values of >0.9760. However, linezolid AUC values were predicted to be within the narrower boundary of 0.76 to 1.5-fold by a higher percentage by the $C_{trough}$ (78.3 %) versus $C_{max}$ model (48.2 %). The $C_{trough}$ model showed superior correlation of predicted versus observed values and RMSE ($r = 0.9031$; 28.54 %, respectively) compared with the $C_{max}$ model ($r = 0.5824$; 61.34 %, respectively).

*Conclusions* A single time point strategy of using $C_{trough}$ level is possible as a prospective tool to measure the AUC of linezolid in the patient population.

**Key Points**

The linear regression model of maximum plasma drug concentration ($C_{max}$) versus area under the plasma concentration–time curve (AUC) $C_{max}$ and trough plasma concentration ($C_{trough}$) versus AUC showed excellent correlation.

Linezolid AUC values were accurately predicted with the $C_{trough}$ model compared with the $C_{max}$ model, with better error predictions.

The single time point $C_{trough}$ model can be utilized in a prospective fashion to measure the AUC of linezolid in patients.

## 1 Introduction

Linezolid, belonging to the oxazolidinone class of antibacterials, was the first in the class to be granted global approval for treating a variety of infections related to Gram-positive pathogens [1, 2]. Both oral and intravenous drug formulations are available to provide convenient therapy for patients [2]. Linezolid's mechanism of action is

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unique and suggested to occur via significant inhibition of the bacterial protein synthesis complex initiation in the bacterial system via the direct action of linezolid on the binding site for initiator transfer RNA (t-RNA) [3, 4]. Linezolid significantly inhibits the growth of a variety of Gram-positive bacterial strains, including staphylococci, streptococci, and enterococci. Furthermore, it shows antimicrobial activity against both methicillin-resistant *Staphylococcus aureus* (MRSA) and vancomycin-resistant enterococci (VRE) [5–7]. The hallmark of linezolid's antibacterial activity is its persistent and long-acting post-antibiotic effect, which may render it useful in strains that are difficult to treat. In addition, this effect may also curb the development of bacterial resistance to linezolid. Linezolid has been found to be an important option in the treatment of multiple drug-resistant tuberculosis [MDR-TB] [8]. Linezolid has an excellent minimum inhibitory concentration (MIC) against *Mycobacterium tuberculosis* and several first-line drug-resistant isolates [9–11]. The same dosing regimen (every 12 h) used to treat patients with Gram-positive infections has been used to treat patients with MDR-TB [11–13].

The safety, tolerability, and pharmacokinetics of linezolid in humans has been investigated for both intravenous and oral use [14–16]. It has been shown to be well tolerated in doses up to 625 mg given intravenously twice daily for up to 7 days in the clinic and in doses of either 400 mg or 600 mg given orally twice daily for up to 28 days [14–16]. Pharmacokinetic investigation has confirmed complete bioavailability of oral linezolid; this suggests it can be used interchangeably permitting oral and intravenous drug switches during therapy, if necessary. After oral administration, linezolid reached peak levels within 1–1.5 h, suggesting relatively rapid absorption of the drug. After intravenous administration, the peak levels were reached at the end of the 30-min drug infusion [14, 16]. Both maximum plasma drug concentration ($C_{max}$) and area under the plasma concentration–time curve (AUC) values appeared to increase in a dose-proportional manner after oral or intravenous routes of administration. Almost two-thirds of linezolid total clearance was renal; the remaining one-third was via non-renal routes [14–16]. Regardless of the administration route, the half-life of linezolid ranged from 5 to 7 h, supporting twice daily dosing of the drug. Drug accumulation occurred at steady state, albeit numerically small. A mass balance study showed that approximately 50 % of administered linezolid was recovered in the urine, and comprised two inactive metabolites; another 35 % of the dose was represented by the intact parent compound [14–16].

We were interested in predicting the AUC of linezolid using a simple and straightforward approach for universal application. To be rigorous, we assembled published pharmacokinetic data of linezolid from various studies with different subject populations to make the dataset very heterogeneous in nature. However, for the model development we used data from a single pharmacokinetic study that provided a wide spread of the pharmacokinetic parameters, such as $C_{max}$, trough plasma concentration ($C_{trough}$), and AUC for modelling purposes.

## 2 Scope

- To develop relationship using linear regression correlations of $C_{trough}$ versus AUC and $C_{max}$ versus AUC of linezolid from a published oral pharmacokinetic study.
- To perform an internal validation to predict the AUC of linezolid following intravenous dosing from the same study using both the developed models.
- To perform an external validation for the prediction of the linezolid AUC following oral and intravenous administration from scores of other published studies using the relevant $C_{trough}$ and $C_{max}$ data.

## 3 Methods

We searched the National Center for Biotechnology Information PubMed® database for relevant abstracts and full-length texts pertaining to the pharmacokinetics of linezolid. The keywords used in the search included linezolid, pharmacokinetics, humans, and clinical. The aim of the present analysis was to seek a relationship between $C_{trough}$ versus AUC and $C_{max}$ versus AUC for linezolid using unweighted linear regression analysis. Once established, we then used the appropriate regression lines in the prediction of AUC values for linezolid.

### 3.1 Data Source for Model Development

We obtained the mean pharmacokinetic data that provided $C_{max}$ and AUC values for linezolid from published pharmacokinetic data in healthy subjects [15–49]. The oral pharmacokinetic data to create the reference model for linezolid were from a double-blind, placebo-controlled study with 3:1 randomization of subjects to active relative or placebo at all dose levels [16]. The goal of the clinical study was to obtain clinical safety, tolerability, and pharmacokinetics data for linezolid after single and multiple oral administration to healthy subjects. In total, three doses (375, 500, and 625 mg) of linezolid were administered orally on day 1 (single dose) and from day 2 onwards (multiple doses). The same oral doses were administered for another 14.5 days every 12 h. The second study examined the safety, tolerability, and pharmacokinetics of

linezolid in healthy subjects following intravenous drug administration. Two doses (500 and 625 mg) of linezolid were administered via a 30-min infusion on day 1 (single dose) and from day 2 (multiple doses) onwards for another 7.5 days; the same intravenous doses were administered via a 30-min infusion every 12 h [16].

The pharmacokinetic data were gathered after single and multiple doses following both oral and intravenous administration of linezolid. The frequency of the blood samples was adequate to assess linezolid pharmacokinetics with single and multiple doses regardless of the drug administration route. The AUC values used for linezolid in the $C_{max}$ regression model represented both $AUC_{inf}$ (single-dose study) and $AUC_{tau}$ (multiple-dose study) values. However, for the $C_{trough}$ regression model, $AUC_{tau}$ values (multiple-dose study) were used. The AUC data for linezolid obtained from the intravenous study were used for internal validation of the two regression models. In addition, for each pair of observed $C_{max}$ versus AUC and $C_{trough}$ versus AUC, four additional data points were generated via the addition or subtraction of either one or two standard deviations from the corresponding mean values of each parameter (i.e., $C_{max}$, $C_{trough}$, and AUC). This provided a basis for a larger spread of the $C_{max}$, $C_{trough}$, and AUC data to facilitate the model development. The incorporation of standard deviation assisted spread of the parameter values has been documented in the linear regression analysis of cyclosporine [50].

For the $C_{max}$ model, 30 pairs of $C_{max}$ and AUC values for linezolid were used as raw reference data in establishing the regression model (Table 1). For the $C_{trough}$ model, 14 pairs of $C_{trough}$ and AUC values for linezolid were used as raw reference data in establishing the regression model (Table 1). The data spread of $C_{max}$, $C_{trough}$, and AUC for linezolid were approximately 7.67-fold (4.07–31.23 μg/ml), approximately 54.57-fold (0.28–15.28 ng/ml), and 16.74-fold (15.7–262.8 μg × h/ml), respectively (Table 1).

### 3.2 Linear Regression Model

Separate linezolid models representing $C_{max}$ versus AUC and $C_{trough}$ versus AUC were established by performing an un-weighted linear regression of the respective paired datasets to obtain the regression lines:

$$Y = mX + C,$$

where $m$ is the slope of the line and $C$ is the intercept value. For each regression model of the paired datasets, a correlation coefficient was established. The developed $C_{max}$ versus AUC model was utilized in the prediction of the AUC for the linezolid. The in-built statistical package in Microsoft® Excel 2010 (Microsoft Company, Redmond, WA, USA) was used to perform linear regressions and establish correlation coefficients.

### 3.3 Prediction Using Published $C_{max}$ and $C_{trough}$ Data

#### *3.3.1 Internal Dataset Validation*

The intravenous data obtained from the same study that supplied the raw reference data for establishing the regression models using both $C_{max}$ and $C_{trough}$ were used for the internal validation [16].

#### *3.3.2 External Dataset Validation*

Scores of publications that described the pharmacokinetics of linezolid after oral and intravenous dosing in a variety of patient populations and heathy subjects were gathered [15–49], and the respective observed individual, mean/median $C_{max}$ or $C_{trough}$ values were used to predict AUC for linezolid using the regression lines as applicable. The predicted AUC values obtained from the two models were then subjected for additional statistical tests.

### 3.4 Statistical Tests and Fold-Difference Computation

The fold difference of the linezolid AUC prediction was separately calculated for the two regression models and was defined as the quotient of observed AUC and predicted AUC value. Various categories of fold difference ranging from <0.5-fold, 0.51- to 0.75-fold, 0.76- to 1.25-fold, 1.26 to 1.5-fold, 1.51 to 2-fold, and >2-fold were created to understand the spread and goodness of the prediction.

For the purpose of the current analysis, a prediction within 0.5 to 2-fold difference was considered satisfactory for the external dataset validation and a narrower prediction of within 1.5-fold difference was considered appropriate for the internal dataset validation. Fold difference-based statistical comparison has previously been employed and validated for several drugs [50–56].

We used a double-sided paired $t$-test to evaluate the observed (literature data) versus predicted AUC for the linezolid. The mean absolute error (MAE) was defined as the mean of the observed AUC values minus the predicted AUC values of linezolid; 95 % confidence interval limits were generated and an appropriate p-value was assigned for the statistical significance using the T-test calculator (Graphpad, San Diego, CA, USA).

$$\text{MAE} = \sum_{i=1}^{N} (xi - yi)$$

**Table 1** Pharmacokinetic data used for developing linear regression models for linezolid

| Model type | Route, dose, type | Data tabulation | Single dose | | Multiple dose | | Reference |
|---|---|---|---|---|---|---|---|
| | | | $C_{max}$ (μg/ml) | $AUC_{inf}$ (μg × h/ml) | $C_{max}$ (μg/ml) | $AUC_{tau}$ (μg × h/ml) | |
| $C_{max}$ | Oral, 375 mg, single dose | Mean | 8.21 | 65.5 | 13.1 | 82.8 | Stalker et al. [16] |
| | | Mean (−1 SD) | 6.14 | 40.6 | 10.2 | 60.2 | |
| | | Mean (+1 SD) | 10.28 | 90.4 | 16 | 105.4 | |
| | | Mean (−2 SD) | 4.07 | 15.7 | 7.3 | 37.6 | |
| | | Mean (+2 SD) | 12.35 | 115.3 | 18.9 | 128 | |
| | Oral, 500 mg, single dose | Mean | 10.4 | 74.3 | 15.3 | 99.2 | |
| | | Mean (−1 SD) | 7.87 | 46.4 | 11.58 | 62.5 | |
| | | Mean (+1 SD) | 12.93 | 102.2 | 19.02 | 135.9 | |
| | | Mean (−2 SD) | 5.34 | 19.3 | 7.86 | 25.8 | |
| | | Mean (+2 SD) | 15.46 | 130.1 | 22.74 | 172.6 | |
| | Oral, 625 mg, single dose | Mean | 12.7 | 102 | 18.75 | 147 | |
| | | Mean (−1 SD) | 9.34 | 72.3 | 12.51 | 89.1 | |
| | | Mean (+1 SD) | 16.06 | 131.7 | 24.99 | 204.9 | |
| | | Mean (−2 SD) | 5.98 | 42.6 | 6.27 | 31.2 | |
| | | Mean (+2 SD) | 19.42 | 161.4 | 31.23 | 262.8 | |
| $C_{trough}$ | Oral, 375 mg, single dose | Mean | NA | NA | 3.9[a] | 82.8 | |
| | | Mean (−1 SD) | | | 2.05 | 60.2 | |
| | | Mean (+1 SD) | | | 5.75 | 105.4 | |
| | | Mean (−2 SD) | | | 0.18[b] | 37.6[b] | |
| | | Mean (+2 SD) | | | 7.6 | 128 | |
| | Oral, 500 mg, single dose | Mean | NA | NA | 5.04 | 99.2 | |
| | | Mean (−1 SD) | | | 2.66 | 62.5 | |
| | | Mean (+1 SD) | | | 7.42 | 135.9 | |
| | | Mean (−2 SD) | | | 0.28 | 25.8 | |
| | | Mean (+2 SD) | | | 9.8 | 172.6 | |
| | Oral, 625 mg, single dose | Mean | NA | NA | 8.02 | 147 | |
| | | Mean (−1 SD) | | | 4.39 | 89.1 | |
| | | Mean (+1 SD) | | | 11.65 | 204.9 | |
| | | Mean (−2 SD) | | | 0.76 | 31.2 | |
| | | Mean (+2 SD) | | | 15.28 | 262.8 | |

*AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration, *NA* not available

[a] $C_{trough}$ reported

[b] Value excluded from the regression analyses

In addition, we calculated mean square error and root means square error (RMSE) for linezolid (shown below) using Microsoft® Excel 2010.

$$\mathrm{MSE} = \frac{1}{N}\sum_{i=1}^{N}(xi - yi)^2$$

$$RMSE = \sqrt{\frac{1}{N}\sum_{i=1}^{N}(xi - yi)^2}$$

### 3.5 Data Utility and Conversions

All data points from the reference data, with the exception of a single pair for the $C_{trough}$ model were used in the model development for linezolid. For consistency for the data assessment, $C_{max}$ values were reported in μg/ml units; AUC values were reported in μg × h/ml. Data unit conversions, if necessary, were made as applicable during compilation and tabulation of the pharmacokinetic data using the same uniform unit format.

## 4 Results

As illustrated in Fig. 1, the $C_{max}$ versus AUC and $C_{trough}$ versus AUC linear regression models were established for linezolid using the reference data presented in Table 1. An excellent correlation coefficient ($r$) value of 0.9762 ($p < 0.001$) and 0.9979 ($p < 0.001$) were obtained for the $C_{max}$ and $C_{trough}$ models, respectively.

The prediction of AUC values for linezolid using the two models was performed using the regression equations described below:

$$\text{AUC(linezolid)} = C_{max}\text{(linezolid)} \times 8.8282 - 20.284$$

$$\text{AUC(linezolid)} = C_{trough}\text{(linezolid)} \times 15.598 - 20.557$$

### 4.1 Internal Dataset Prediction

As shown in Table 2, the use of either $C_{max}$ or $C_{trough}$ regression models developed using oral linezolid data adequately predicted the AUC values obtained after intravenous administration at steady state. The fold difference in the predicted AUC for linezolid was 0.84 and 1.15, for $C_{max}$ and $C_{trough}$ models, respectively.

### 4.2 External Dataset Prediction

#### *4.2.1 $C_{max}$ Model*

Figure 2 displays the comparison of the observed AUC values versus predicted AUC values for linezolid. Less than 50 % of the predicted AUC values were within the 0.76- to1.5-fold limit of the original values (Table 3). Furthermore, AUC fold difference was distributed across the various segments, suggesting a greater variability in the prediction of AUC (Table 3). For instance, 16.6 % of the AUC predictions were <0.5-fold difference, and 1.4 % of the AUC predictions were >2.0-fold difference. The plot of observed AUC versus predicted AUC values for linezolid is shown in Fig. 3 and had a correlation of 0.5824, $n = 222$ ($p < 0.001$). The MAE and RMSE (expressed as %) were 21.34 and 61.34 %, respectively (Table 3).

#### *4.2.2 $C_{trough}$ Model*

Figure 2 displays the comparison of the observed AUC values versus predicted AUC values for linezolid. More than 75 % of the predicted AUC values (i.e., 78.3 %) were within the 0.76- to 1.5-fold limit of the original values (Table 3). Unlike the $C_{max}$ model, no AUC predictions of linezolid were either <0.5- or >2.0-fold difference, suggesting the containment of the AUC values within 0.5- to 2-fold difference (Table 3). The plot of observed AUC versus predicted AUC values for linezolid is shown in Fig. 3 and had a correlation of 0.9031, $n = 120$ ($p < 0.001$). The MAE and RMSE (expressed as percentages) were 16.40 and 28.54 %, respectively (Table 3).

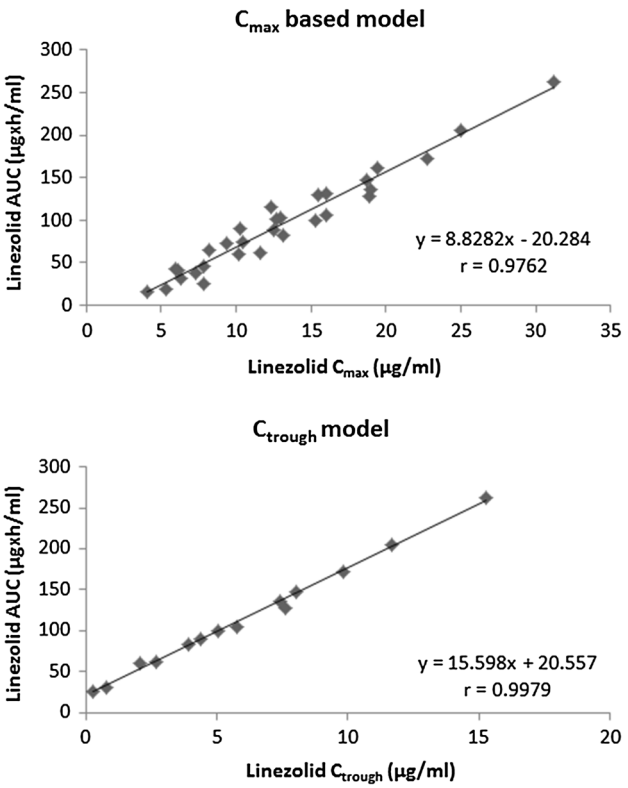


**Fig. 1** Linear regression models developed by linezolid $C_{max}$ vs. linezolid AUC and linezolid $C_{trough}$ vs. linezolid AUC. *AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration

## 5 Discussion

The increased risk posed by resistant Gram-positive pathogens causing frequent fatalities can be circumvented with the prudent use of linezolid to treat a variety of infections. Linezolid is one of the few antibiotics that possess excellent pharmacokinetic properties, such as almost 100 % [14–16] bioavailability and rapid $C_{max}$ after oral administration (almost matching the $C_{max}$ obtained after standard intravenous infusion of the drug), meaning it is easily possible to switch from intravenous to oral drug administration regimens. Therefore, transitioning patients from a hospital/institutional setting to a home setting is made easy with the possibility of changing an intravenous prescription of linezolid to an oral regimen with a dose alteration. This prompted us to establish simple regression models using oral pharmacokinetic data that would enable the prediction of AUC data for linezolid using a single time point strategy regardless of the administration route.

**Table 2** Internal dataset validation: prediction of intravenous area under the plasma concentration–time curve data for linezolid using regression models from oral data

| Model type | Route, dose, type | Observed $AUC_{tau}$ (μg × h/ml) | Predicted $AUC_{tau}$ (μg × h/ml) | Fold difference | Reference |
|---|---|---|---|---|---|
| $C_{max}$ | Intravenous, 500 mg, multiple dose | 81.2 | 106.84 | 0.76 | Stalker et al. [16] |
| | | 61.6 | 79.65 | 0.77 | |
| | | 100.8 | 134.03 | 0.75 | |
| | | 42 | 52.46 | 0.80 | |
| | | 120.4 | 161.22 | 0.75 | |
| | Intravenous, 625 mg, multiple dose | 93.4 | 118.32 | 0.79 | |
| | | 61.1 | 95.19 | 0.64 | |
| | | 125.7 | 141.45 | 0.89 | |
| | | 158 | 164.58 | 0.96 | |
| $C_{trough}$ | Intravenous, 500 mg, multiple dose | 81.2 | 75.31 | 1.08 | |
| | | 61.6 | 54.09 | 1.14 | |
| | | 100.8 | 96.52 | 1.04 | |
| | | 42 | 32.88 | 1.28 | |
| | | 120.4 | 117.73 | 1.02 | |
| | Intravenous, 625 mg, multiple dose | 93.4 | 80.45 | 1.16 | |
| | | 61.1 | 42.08 | 1.45 | |
| | | 125.7 | 118.82 | 1.06 | |
| | | 158 | 157.20 | 1.01 | |

*AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration

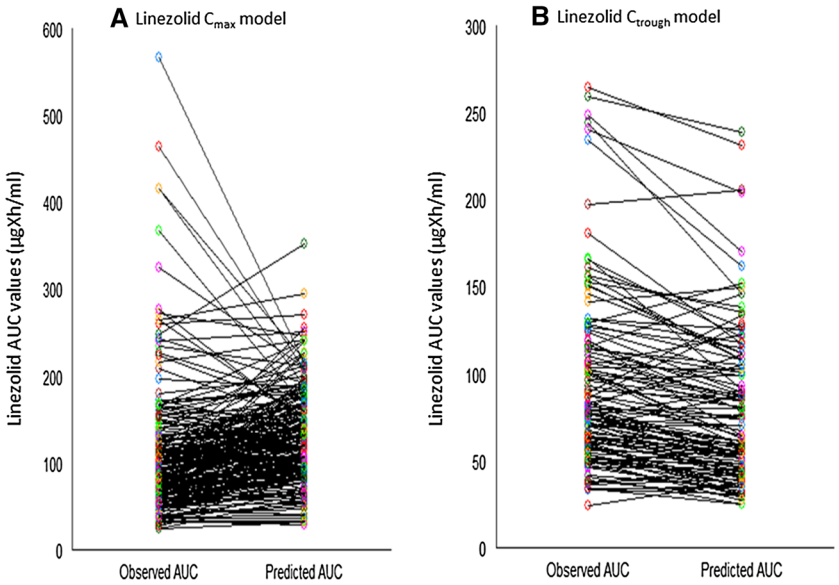


**Fig. 2** Spread of the observed AUC vs. predicted AUC for either linezolid $C_{max}$ model (**a**) or linezolid $C_{trough}$ model. *AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration

The AUC of linezolid is a vital parameter, and the ratio of AUC/MIC has been used as a surrogate for both bacteriological and clinical outcomes [14]. Note also that the linezolid AUC has also been linked to the occurrence of thrombocytopenia [14].

The reference data for linezolid AUC used for building either $C_{max}$ or $C_{trough}$ models represented either $AUC_{tau}$ (every 12 h dosing schedule) or $AUC_{inf}$ (single-dose) values. Because linezolid exhibits linear pharmacokinetics, steady state exposure was expected to be comparable to the single-dose $AUC_{inf}$ data. The calculated AUC values from either of the two models are representative of the exposure of linezolid in a dosing interval since the majority of the examples used in the

**Table 3** Statistical comparisons and fold difference summary between observed vs. predicted area under the plasma concentration–time curve values for linezolid

| Model type | *N* size | Prediction criteria[a] | | | | | Mean AUC (μg × h/ml) | | Mean absolute error (difference %) | Mean square error | Root mean square error (%) | Correlation coefficient (*r* value) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | <0.5-fold | >0.5 to 0.75-fold | 0.76 to 1.25-fold | 1.26 to 1.5-fold | 1.51 to <2-fold | >2-fold | Observed | Predicted | | | |
| $C_{max}$ | 222 | 37 (16.7) | 66 (29.7) | 96 (43.2) | 11 (5.0) | 9 (4.0) | 3 (1.4) | 107.26 | 130.22 | −22.90 (21.34)[b] | 4331.1 | 65.81 (61.34)[b] | 0.5824 |
| $C_{trough}$ | 120 | – | 4 (3.4) | 67 (55.8) | 27 (22.5) | 22 (18.3) | – | 93.05 | 77.79 | 15.26 (16.40)[c] | 705.37 | 26.56 (28.54)[c] | 0.9031 |

*AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration

[a] Data presented as *N* (%)

[b] Derived by the equation: % CV $= \frac{\text{error value}}{107.26} \times 100$

[c] Derived by the equation: % CV $= \frac{\text{error value}}{93.50} \times 100$

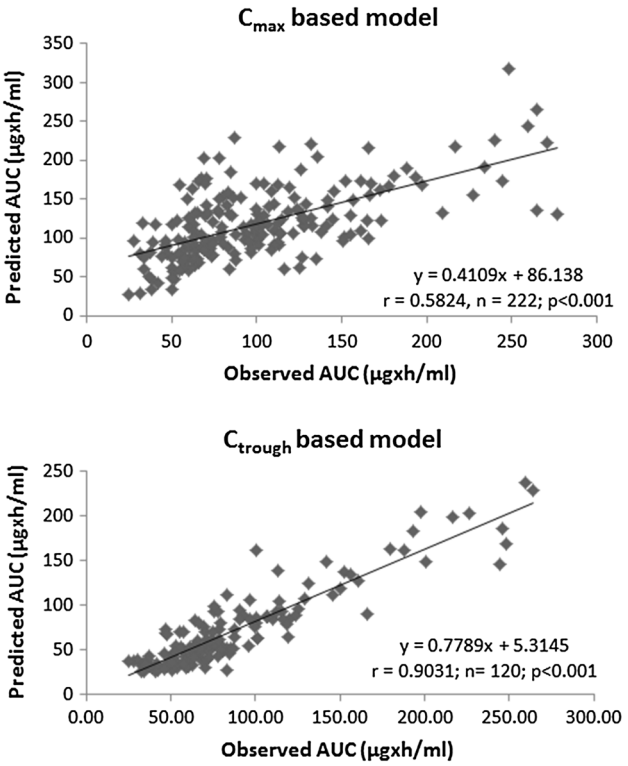


**Fig. 3** Correlation of the observed vs. predicted values for either the linezolid $C_{\max}$ model or the linezolid $C_{\text{trough}}$ model. *AUC* area under the plasma concentration–time curve, $C_{max}$ maximum plasma drug concentration, $C_{trough}$ trough plasma concentration

dataset were from multiple-dose pharmacokinetic studies of linezolid.

Although we were limited by not having individual datasets to build the $C_{\max}$ versus AUC and $C_{\text{trough}}$ versus AUC linear regression models, the mean ± standard deviation approach enabled us to generate additional data points. While this strategy enabled a wider spread of the $C_{\max}$, $C_{\text{trough}}$, and AUC values for linezolid, it did not compromise the scientific integrity of the analysis. For instance, the $C_{\max}$ versus AUC analysis would have yielded a slope value of 7.3458 using as is data, which was in close proximity to the value of 8.8282 with additional data points. Similarly, for the $C_{\text{trough}}$ versus AUC analysis, the slope value of 15.6750 (as is data) was almost overlapping with the slope value of 15.5980 (with additional data points). The internal validation unequivocally supported the ability of models developed with oral data to predict the intravenous exposure data of linezolid, irrespective of $C_{\max}$ or $C_{\text{trough}}$ models.

Based on statistical comparisons, the superiority of $C_{\text{trough}}$ over that of $C_{\max}$ in predicting the AUC of linezolid was established with >2-fold better error prediction rendered by the $C_{\text{trough}}$ model (RMSE: 28.54 %) as compared with the $C_{\max}$ model (RMSE: 61.34 %). The distribution of

AUC fold-differences in the prediction suggested that the $C_{trough}$ model predicted the AUC values to a large extent within the narrow band of 0.75- to 1.5-fold differences. This ability of the $C_{trough}$ model to consistently predict linezolid AUC values within a narrower boundary may be useful in determining the potential for any drug–drug interaction with other drugs co-administered with linezolid. For instance, in the drug–drug interaction study of clarithromycin with linezolid [35], the mean observed AUC for linezolid was 61 (34.6–63.9) ng × h/ml and the $C_{trough}$ model predicted AUC values were 53.1 (34.6–54.9) ng × h/ml, which confirmed its utility.

A clinical pharmacokinetic study was performed previously to explore a limited sampling strategy for the therapeutic drug monitoring (TDM) of linezolid in patients with MDR-TB [34]. Interestingly, the strategy comprised $C_{trough}$ (alone) and $C_{trough}$ combined with two to three additional time points within the 0- to 12-h dosing interval of linezolid. The use of $C_{trough}$ alone was identified as useful for the TDM of linezolid. This was a well planned and executed study with a homogenous patient population, and it yielded an $r$ value of 0.91 and an RMSE of 15 % [34]. To put things into perspective, the present analysis of linezolid was heterogeneous in terms of the nature of studies carried out in different geographies with applicable clinical protocols and collated data for over a decade, covering different patient populations being treated with linezolid for various resistant Gram-positive pathogens, and it also included oral and intravenous administration routes. Despite the enormous heterogeneity, we were able to establish an $r$ value of 0.90 and an RMSE of 29 % using the $C_{trough}C_{trough}$-based model. Furthermore, we also examined two individual patient studies of linezolid that had a sample size of at least $n = 10$ and performed the regression analysis of $C_{trough}$ versus AUC values to further validate our developed model, which was based on mean data in healthy subjects.

The first study involved critically ill patients with ventilator-associated pneumonia, where plasma and intrapulmonary linezolid concentrations were determined [25]—the $C_{trough}$ versus AUC regression analysis yielded:

$$\text{AUC (linezolid)} = C_{trough}\,\text{(linezolid)} \times 14.884 + 34.894 \quad (r = 0.8464).$$

The second study involved critically ill neurological patients where both cerebrospinal fluid and serum concentrations were measured [44]—the $C_{trough}$ versus AUC regression analysis yielded:

$$\text{AUC (linezolid)} = C_{trough}\,\text{(linezolid)} \times 16.145 + 38.795 \quad (r = 0.9771).$$

Using the examples of the individual patient studies, our present analysis when put into context with previously reported limited sampling strategy work on linezolid [34] strongly suggests that a $C_{trough}$ model could be used prospectively in patients. A single sample collection at $C_{trough}$ has the distinct advantage of minimizing the risk of other opportunistic infections in a community setting. Also, the $C_{trough}$ model would be beneficial when other concomitant drugs are administered, since the sample time is distant from absorption and metabolism processes that may affect the pharmacokinetics of the drug. Perhaps the same sample collected for linezolid may also be useful for measuring other concomitant drugs.

Although we understood that the $C_{max}$ versus AUC model may not be ideal, we attempted to build the model and validate it further. We believe that since $C_{max}$ is largely influenced by the sampling times to define the pharmacokinetic profile of the drug, it may exhibit more intra- and inter-subject variability. From a practicality viewpoint, it may be difficult to sample for a precise $C_{max}$ estimation because it would involve intensive pharmacokinetic sampling. In the present analysis, $C_{max}$ may also have been influenced by differences in the duration of intravenous infusion of linezolid (30 min vs. 1 h infusion). Therefore, institution of a $C_{max}$-based model as a strategy should be considered after carefully weighing the number of limitations it imposes.

As published pharmacokinetic data were lacking, we were unable to examine the predictability of linezolid AUC in obese subjects using either the $C_{max}$ or the $C_{trough}$ models. However, we used the recently published data by Bhalodi et al. [57] to examine the predictability of the $AUC_{tau}$ of linezolid using the $C_{max}$ model. Using the mean $C_{max}$ (20.9 μg/ml) of linezolid in moderately obese patients [57], the predicted $AUC_{tau}$ value was 182.4 μg × h/ml as compared with the observed $AUC_{tau}$ of 130.3 μg × h/ml. Similarly, using the mean $C_{max}$ (18.8 μg/ml) in morbidly obese patients [57], the predicted $AUC_{tau}$ was 161.9 μg × h/ml as compared with the observed $AUC_{tau}$ of 109.2 μg × h/ml. Although $C_{trough}$ data were not available in this study [57], using the $C_{max}$ model suggested that the developed models were applicable for the prediction of linezolid $AUC_{tau}$ in obese patients.

Our work has additional limitations: first, the linear regression models, either $C_{max}$ or $C_{trough}$, developed for linezolid were based on mean data but not on individual subject datasets; second, the AUC predictions for either of the models were based on mean data, while the prediction errors may not truly reflect the errors of the population at large. Third, although the $C_{trough}$ model appeared to provide the best accuracy and bias for predicting AUC values, the clinical pharmacokinetic data in patients should be interpreted with utmost caution, keeping in mind polypharmacy and/or attenuated pathophysiological considerations because of the disease state. Fourth, the $C_{trough}$ model can only be used to render the AUC prediction of

linezolid in a dosing interval ($\tau = 12$ h), but it may be less than ideal for the prediction of $AUC_{inf}$ following single-dose administration of linezolid.

## 6 Conclusions

The $C_{max}$ versus AUC and $C_{trough}$ versus AUC models were unambiguously established for linezolid using published data. The predictions of AUC values using the $C_{trough}$ model were found to be superior to those of the $C_{max}$ model as judged by fold-difference calculations and error predictions such as MAE and RMSE values and correlation coefficients. Since excellent predictions of the AUC values of linezolid were obtained by the $C_{trough}$ model, a single time point strategy of measuring $C_{trough}$ level is possible as a prospective tool in the patient population.

**Compliance with Ethical Standards**

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