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Population pharmacokinetics and dose optimization of intravenous levofloxacin in hospitalized adult patients

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Although levofloxacin has been used for the last 25 years, there are limited pharmacokinetic data to guide levofloxacin dosing in adult patients. This study aimed to develop a population pharmacokinetic model of levofloxacin for adult hospitalized patients and define dosing regimens that attain pharmacokinetic/pharmacodynamic target associated with maximum effectiveness. Blood samples were drawn from 26 patients during one dosing interval. Population pharmacokinetic modelling and dosing simulations were performed using Pmetrics®. Pathogen minimum inhibition concentration (MIC) distribution data from the European Committee on Antimicrobial Susceptibility Testing database was used to analyse fractional target attainment (FTA). A two-compartment model adequately described the data. The final model included estimated glomerular filtration rate (eGFR) to describe clearance. The population estimate for clearance was 1.12 L/h, while the volume of distribution in the central compartment and peripheral compartments were 27.6 L and 28.2 L, respectively. Our simulation demonstrated that an area under free concentration–time curve to MIC ≥ 80 was hardly achieved for pathogens with MIC ≥ 1 mg/L. Low FTA against *Pseudomonas aeruginosa* and *Streptococcus pneumoniae* were observed for patients with higher eGFR (≥ 80 mL/min/1.73m²). A daily levofloxacin dose of 1000 mg is suggested to maximise the likelihood of efficacy for adult patients.

Lower respiratory tract infections (LRTIs), including pneumonia, are a leading cause of death in many developing countries, including Indonesia¹. Levofloxacin is a fluoroquinolone antibiotic and is considered a first-line empirical treatment for many patients with pneumonia^{2–4}, given it has good coverage against Gram-negative, Gram-positive and atypical bacteria⁵. In addition, high concentrations of levofloxacin are achieved in lung tissue⁶. Nevertheless, resistance to levofloxacin has been documented and this may jeopardise its use against pathogens with reduced susceptibility⁷. Optimising levofloxacin dosing is an important strategy to overcome this problem.

Similar to other fluoroquinolones, the pharmacokinetic/pharmacodynamic (PK/PD) indices that best describe levofloxacin antibacterial activity are the area under the free concentration–time curve to MIC ratio ($fAUC_{0-24}/MIC$) and the peak concentration to MIC ratio (C_{max}/MIC)^{8,9}. An $fAUC_{0-24}/MIC \geq 80$ has been recommended as a PK/PD target that is most likely to result in successful levofloxacin treatment⁹. Dosing regimens for levofloxacin should aim to achieve this PK/PD target, particularly in severely unwell patients.

An in-depth understanding of levofloxacin pharmacokinetics (PK), including PK variability and the covariate(s) influencing such variability, remains a key step in the design of dosing regimens that maximise the achievement of desired PK/PD targets. Previous data demonstrates differences in levofloxacin PK in patients^{10–15}

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Characteristic	Total patients (%)	ICU patients (%)	Non-ICU patients (%)
Total number	26	6	20
Male	16 (61.5)	3 (50.0)	13 (65.0)
With mechanical ventilation	4 (15.4)	4 (66.7)	0 (0)
Age (year) ^a	58.8 ± 16.4	62.5 ± 23.1	57.7 ± 14.5
Weight (kg) ^{a,b}	61.6 ± 12.1	63.0 ± 16.6	60.5 ± 7.82
Se _{Cr} (mg/dL) ^a	1.99 ± 1.48	2.03 ± 1.88	2.08 ± 1.56
Albumin (g/l) ^{a,c}	2.80 ± 0.50	3.13 ± 0.58	2.62 ± 0.26
eGFR _{CKD-EPI} (mL/min/1.73m ²) ^a	52.7 ± 33.7	55.2 ± 40.7	52 ± 32.5
Dose (once daily)			
500 mg	8 (30.8)	2 (33.3)	6 (30.0)
750 mg	14 (53.9)	2 (33.3)	12 (60.0)
500–750*	1 (3.85)	0 (0)	1 (5.00)
750–500**	3 (11.5)	2 (33.3)	1 (5.00)

Table 1. Demographic data of study participants. ^aPresented as mean ± SD. ^bNumber of patients with weight information: 5 (ICU patients) and 6 (non-ICU patients). ^cNumber of patients with albumin information: 3 (ICU patients) and 5 (non-ICU patients). *Patient received two doses of 500 mg and then one dose of 750 mg. **Two patients received one dose of 750 mg then a dose of 500 mg and another one patient received one dose of 750 mg then another two doses of 500 mg.

compared with healthy subjects^{16,17}. Additionally, levofloxacin population PK varies between ethnicities, with different PK parameters reported among Chinese^{10,11}, Korean¹², and Caucasian^{13–15} patients. Consequently, a standardised dose of levofloxacin may not always achieve comparable exposures across different ethnic groups of patients^{10–13}.

Levofloxacin PK data in Indonesian hospitalized patients is not available and this may be problematic in optimising dosing in pneumonia, particularly in pathogens with higher MICs. Therefore, the aim of the present study was to describe the population PK of levofloxacin in adult Indonesian hospitalized patients and then to apply Monte Carlo simulation to define appropriate levofloxacin dosing regimens that can attain a priori PK/PD targets.

Results

Patient and sampling characteristics. Five data points from one ICU patient were considered biologically implausible and they were excluded for PK analysis resulting a total of 124 blood samples from 26 patients being potentially included in the final PK analysis. Almost all patients (88.5%) were diagnosed with pneumonia. No patient was given more than 750 mg of levofloxacin in a 24 h period. Table 1 describes the demographic characteristics of the patients. No concomitant therapy classified as known to significantly interact with levofloxacin PK was given in any of the study patients. No adverse drug reactions to levofloxacin were reported in any of the study patients.

Population PK model. Three concentration data points were further excluded from the population PK modeling due to possible contamination either with the previous (n = 2) or next dose (n = 1). An average of 4.77 blood samples per-patient (2–6 samples per patient) were obtained. Two blood samples per-patient were collected in two patients (7.69%). Levofloxacin was best described as a two-compartment model. The value of -2LL and AIC in the best structural model are described in Table 2.

The only covariates that improved the goodness-of-fit of the scatter plots and decreased the value of -2LL and AIC significantly was eGFR_{CKD-EPI} on CL (Table 2). The CL of levofloxacin was best described as the following equation: $CL = (0.044 * eGFR_{CKD-EPI}) + 0.358$, where CL is the levofloxacin CL and eGFR_{CKD-EPI} is the estimated glomerular filtration rate calculated with CKD-EPI equation.

The goodness of fit of the observed versus predicted plots, both population, and individual prediction, for the final model with covariate were acceptable (Fig. 1). The distribution of the observed data within the percentiles of the simulated data is shown in Fig. 2. The estimated population PK parameters from the final model with covariate are presented in Table 2.

Dosing simulation. PTA. Figures 3 and 4 present the PTA of several dosing regimens of levofloxacin for several typical patient scenarios with eGFR_{CKD-EPI} level of 20–50 mL/min/1.73m² and 80–120 mL/min/1.73m², respectively. In all eGFR_{CKD-EPI} levels, there were various alternatives of dosing regimens observed that attained the PTA ≥ 90% against pathogens with MIC 0.5 mg/L even though not all resulted in a PTA ≥ 90% both on the first day of treatment and at a steady state condition. The highest MIC at which ≥ 90% PTA could be achieved among all dosing regimens is in the supplementary file (suppl. Table S1).

For patients with eGFR_{CKD-EPI} of 80 and 120 mL/min/1.73m², a daily dose of 500 mg every 24 h has a PTA ≥ 90% against an MIC of 0.25 mg/L which is the epidemiological cut-off value (ECOFF) for *Escherichia coli* and *Klebsiella pneumoniae*¹⁸. Increasing the dose to 750 mg every 24 h, ensures an acceptable PTA against an MIC value of 0.5 mg/L (the ECOFF for *Staphylococcus aureus*)¹⁸ for both the first day of treatment and at steady

Parameter ^a	Mean	SD	CV%	Median	Shrink (%)
Pharmacokinetic value					
V_c (L)	27.6	19.1	69.3	26.4	3.71
CL (L/h)	1.12	0.58	52	0.90	1.60
Q (L/h)	30.9	16.4	53.2	33.2	5.81
V_p (L)	28.2	16.2	57.7	27.9	5.20
Compartment model					
Model selection					
One	393	399			
Two (with $K_{PC}-K_{CP}$)	272	283			
Two (with Q)	256	267			
Two (with Q and the additional of $eGFR_{CKD-EPI}$ on CL) FINAL	250	261			

Table 2. Estimates of levofloxacin pharmacokinetic parameters from the final model with covariate and model selection. ^a V_c volume of central compartment, CL clearance, Q inter-compartment clearance, V_p volume of peripheral compartment, K_{PC} the rate constant from the central compartment to the peripheral compartment, K_{CP} the rate constant from the peripheral compartment to the central compartment, $-2LL - 2 * \text{Log-likelihood}$ at each cycle, AIC akaike information criterion at each cycle.

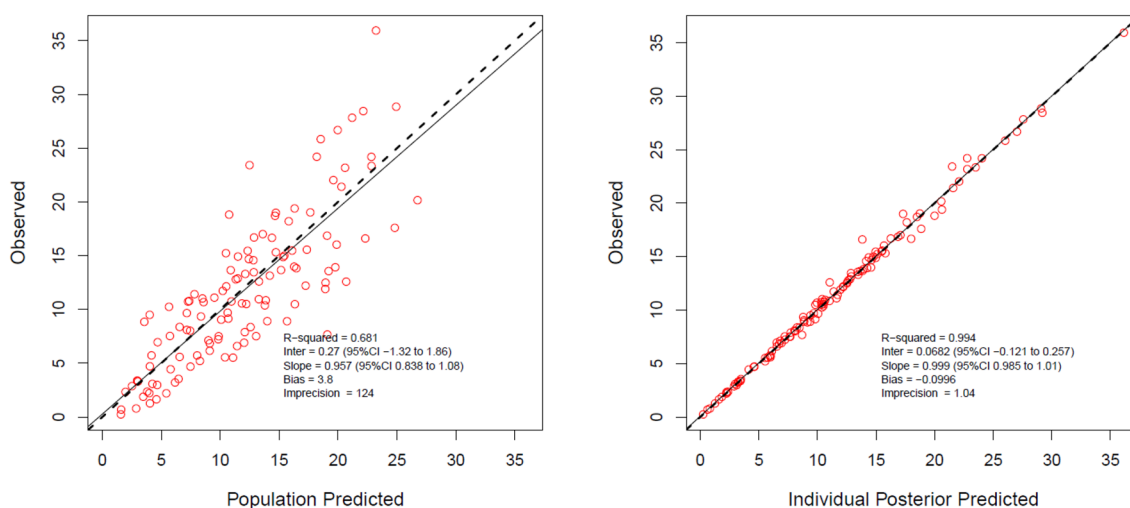


Figure 1. Diagnostic plot for the final covariate; (left) observed versus population predicted plasma concentrations and (right) individual predicted plasma concentrations.

state for patients with $eGFR_{CKD-EPI}$ 80 mL/min/1.73m² ($\geq 90\%$ and $\geq 90\%$) and $eGFR_{CKD-EPI}$ 120 mL/min/1.73m² (89.3% and $\geq 90\%$).

In all $eGFR_{CKD-EPI}$ levels and for all investigated dosing regimens, it was difficult to obtain a PTA $\geq 90\%$ for an MIC ≥ 1 mg/L. Even the highest daily dose of levofloxacin, either given as 1000 mg every 24 h or 500 mg every 12 h, could not attain a PTA $\geq 90\%$ against MIC ≥ 1 mg/L.

FTA. The FTA of all levofloxacin dosing regimens over four different $eGFR_{CKD-EPI}$ levels achieved $\geq 90\%$ against the empirical MIC distribution of *M. cattharalis* and *H. influenzae* (detailed percentages are not shown). The empirical FTAs were determined against *E. coli*, *K. pneumoniae*, *P. aeruginosa*, *S. aureus*, and *S. pneumoniae* for patients with $eGFR_{CKD-EPI}$ 20–120 mL/min/1.73m² (Table 3). While it can be found in each $eGFR_{CKD-EPI}$ level group that some dosing regimens attained an FTA $\geq 90\%$ against *E. coli*, *K. pneumoniae*, and *S. pneumoniae*, most levofloxacin dosing regimens only attained an FTA between 81 and 84% against *S. aureus*. Moreover, the FTAs of almost all of the simulated dosing regimens in all $eGFR_{CKD-EPI}$ levels are far lower than the target of $\geq 90\%$ against *P. aeruginosa*.

Considering the MIC distribution of the susceptible strains, an acceptable FTA ($\geq 90\%$) can be found in all different groups of $eGFR_{CKD-EPI}$ levels by giving any dosage regimen against *E. coli*, *K.pneumoniae*, *H. influenzae*, *M. cattharalis*, and *S. aureus* (detailed percentages for each pathogen are not shown). A dose of 1000 mg every 24 h is most likely to achieve desirable FTA for both *P. aeruginosa* and *S. pneumoniae* in patients with $eGFR_{CKD-EPI} \geq 50$ mL/min/1.73m² (Table 4).

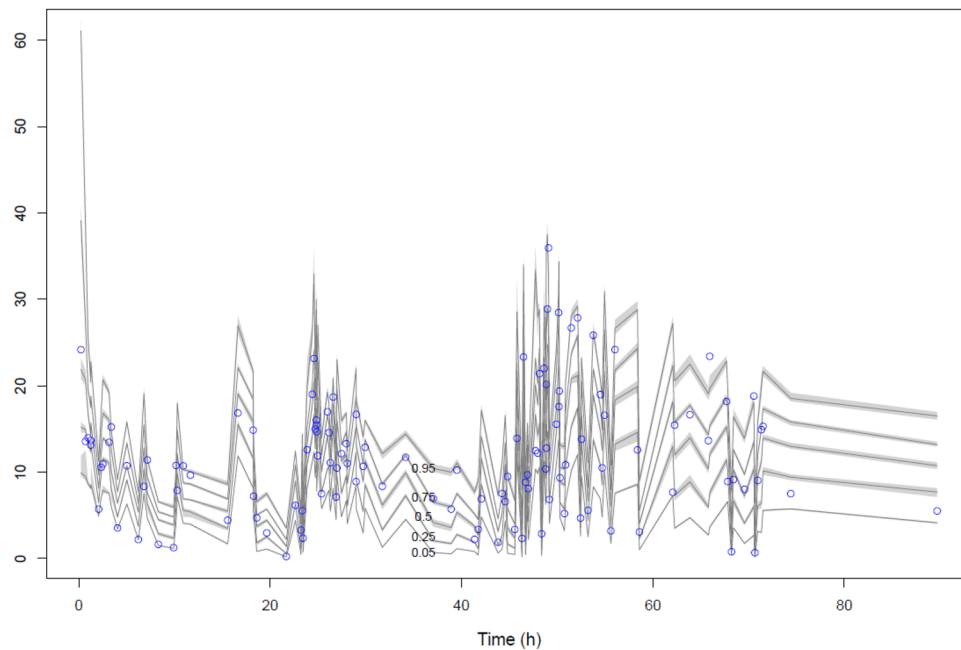


Figure 2. Visual predictive check plot of the final covariate two-compartment model; y axis indicated concentrations of levofloxacin (mg/L). Percentiles (with shaded 95% confidence interval) are the lines shown as 0.95, 0.75, 0.5, 0.25, and 0.05 values. Individual circles represent the observed concentration.

Discussion

To the best of our knowledge, ours is the first study to describe the population PK of levofloxacin in Indonesian hospitalized patients. In our study, we combined ICU and non-ICU patients into one group to increase the generalisability of the results. There was a non-statistically significant difference in levofloxacin PK observed in our study when compared to previous published data¹³. Our dosing simulation indicated that higher than 750 mg daily dose of levofloxacin may be required to achieve $fAUC/MIC \geq 80$ in patients with $eGFR_{CKD-EPI}$ values ≥ 80 mL/min/1.73m² and for infections with an $MIC \geq 1$ mg/L.

Our PK model is consistent with previously published articles describing levofloxacin PK using a two-compartment model^{12–15}. The value of estimated PK parameters, however, are relatively different compared with other studies. For example, we found that the CL of levofloxacin in Indonesian hospitalized patients is relatively lower (CL = 1.12 L/h) compared to what has been reported in other studies including Italian patients (CL = 8.66 L/h); Korean patients (CL = 6.19 L/h); and patients from the United States (9.27 L/h)^{12,13,15}. Given the significant influence of renal function on levofloxacin CL^{12–15}, the lower levofloxacin CL in our study could potentially be related to the lower renal function associated with this patient cohort. The mean (\pm SD) $eGFR$ of patients in our study was 53 (\pm 33) mL/min/1.73m² while the mean creatinine clearance (CL_{Cr}) reported in the study by Roberts et al., Kiem et al., and Preston et al. were 70 (\pm 67; in critically ill group) and 70 (\pm 32; in non-critically ill group), 80.6 (\pm 28.2) mL/min, 82.9 (\pm 31.6), respectively^{12,13,15}. It is worth noting, however, that renal function might not be sufficient to completely describe the variability of levofloxacin CL. In our model, renal function might contribute to explain around 52% of CL variability, while it was found to be 45% and 14% in models developed by Roberts et al. and Kiem et al., respectively^{12,13}. Given renal CL represents approximately 60% of total body CL of levofloxacin, it is understandable that renal function alone cannot explain CL variability¹⁹. Therefore, it is likely that the different reported CL value between our model and other reported models might also be influenced by non-renal CL.

Nevertheless, the value of $eGFR$ or CL_{Cr} is likely to be the foundation to optimise dosing of levofloxacin. Altered dosing approaches should be implemented in patients depending on their $eGFR_{CKD-EPI}$ values, especially if a causative organism with $MIC > 0.5$ mg/L is a concern. Our dosing simulation indicates that the majority of the simulated dosing regimens in all $eGFR_{CKD-EPI}$ groups could attain $PTA \geq 90\%$ against an MIC of ≤ 0.5 mg/L. This PTA is, more difficult to be attained against an MIC of ≥ 1 mg/L, especially in patients with higher $eGFR_{CKD-EPI}$ levels. For an MIC 1 mg/L, our study found that every alternate day dosing regimen with 750 or 500 mg every 48 h might still be appropriate for patients with $eGFR_{CKD-EPI}$ 20 mL/min/1.73m² should a PTA of around 85% be clinically acceptable at both the first day of treatment and steady state. A daily dosing regimen, however, should be implemented for patients with $eGFR_{CKD-EPI} \geq 50$ mL/min/1.73m² and this supports product information recommendations²⁰. It is worth mentioning that the highest dose of levofloxacin commonly prescribed at the research sites of our study, i.e., 750 mg every 24 h, could obtain acceptable PTA against MIC of 1 mg/L at the fifth day of treatment for patients with $eGFR_{CKD-EPI}$ 50 mL/min/1.73m² but not for patients with a $eGFR_{CKD-EPI}$ of 80 mL/min/1.73m² or 120 mL/min/1.73m². Maintaining a dose of 750 mg does not resolve the possibility of underexposure of levofloxacin at higher $eGFR$ s. However, it should be anticipated that $eGFR$ or CL_{Cr} may not

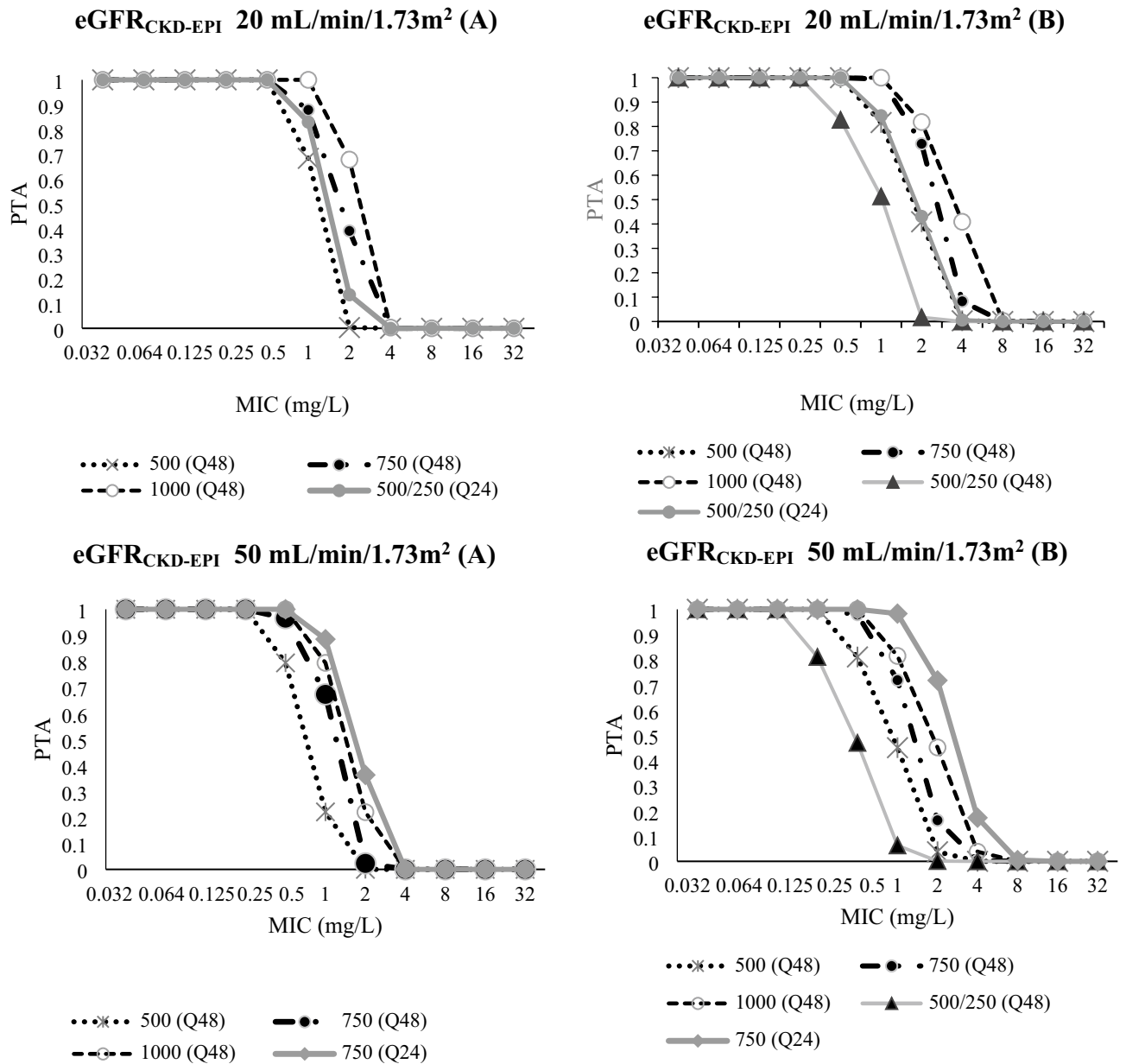


Figure 3. Probability of target attainment (PTA; $fAUC/MIC \geq 80$) of several dosage regimens of levofloxacin at (A) the first 24-h and (B) steady state; for patients with $eGFR_{CKD-EPI}$ 20 mL/min/1.73m² and $eGFR_{CKD-EPI}$ 50 mL/min/1.73m²; For the first dose (A) in both $eGFR_{CKD-EPI}$ groups: the lines for levofloxacin 500/250 (Q48) and 750/500 (Q48) were relatively similar to 500 (Q48) and 750 (Q48), respectively; For the steady state (B) in both $eGFR_{CKD-EPI}$: lines for 750/500 (Q48) was relatively similar to 500 (Q48). In $eGFR_{CKD-EPI}$ 50 mL/min/1.72m²: the lines for levofloxacin 500/250 (Q24) and 500 (Q24) at the first dose (A) was relatively similar to 750 (Q48). While at steady state (B), the lines for levofloxacin 500/250 (Q24) and 500 (Q24) were relatively similar to 500 (Q48) and 1000 (Q48), respectively. To provide better clarity, lines with similarity shapes were not presented.

always be available in the Indonesian hospital setting at the time when doses are initiated or dosing adjustments made²¹. Serum creatinine is more widely reported and so it is likely that dosing adjustments are made based on this reported value. Notably, Se_{Cr} may not accurately determine renal function²², and so this may limit the effort to optimise levofloxacin exposure.

Doses higher than 750 mg daily may be required to achieve $fAUC/MIC \geq 80$, particularly for patients with higher $eGFR_{CKD-EPI}$ values and for infections with an $MIC \geq 1$ mg/L. Our simulations emphasize that a daily dose of 1,000 mg levofloxacin could attain a PTA slightly below 90% for patients with $eGFR_{CKD-EPI}$ 80 mL/min/1.73m². Roberts et al., however, suggested that this higher daily dose could not attain a PTA $\geq 60\%$ in patients with CL_{Cr} 70 mL/min¹³. A higher model estimated population value for levofloxacin CL compared to what we found in our study is the likely explanation for this relatively lower reported PTA¹³. However, for patients with $eGFR_{CKD-EPI}$

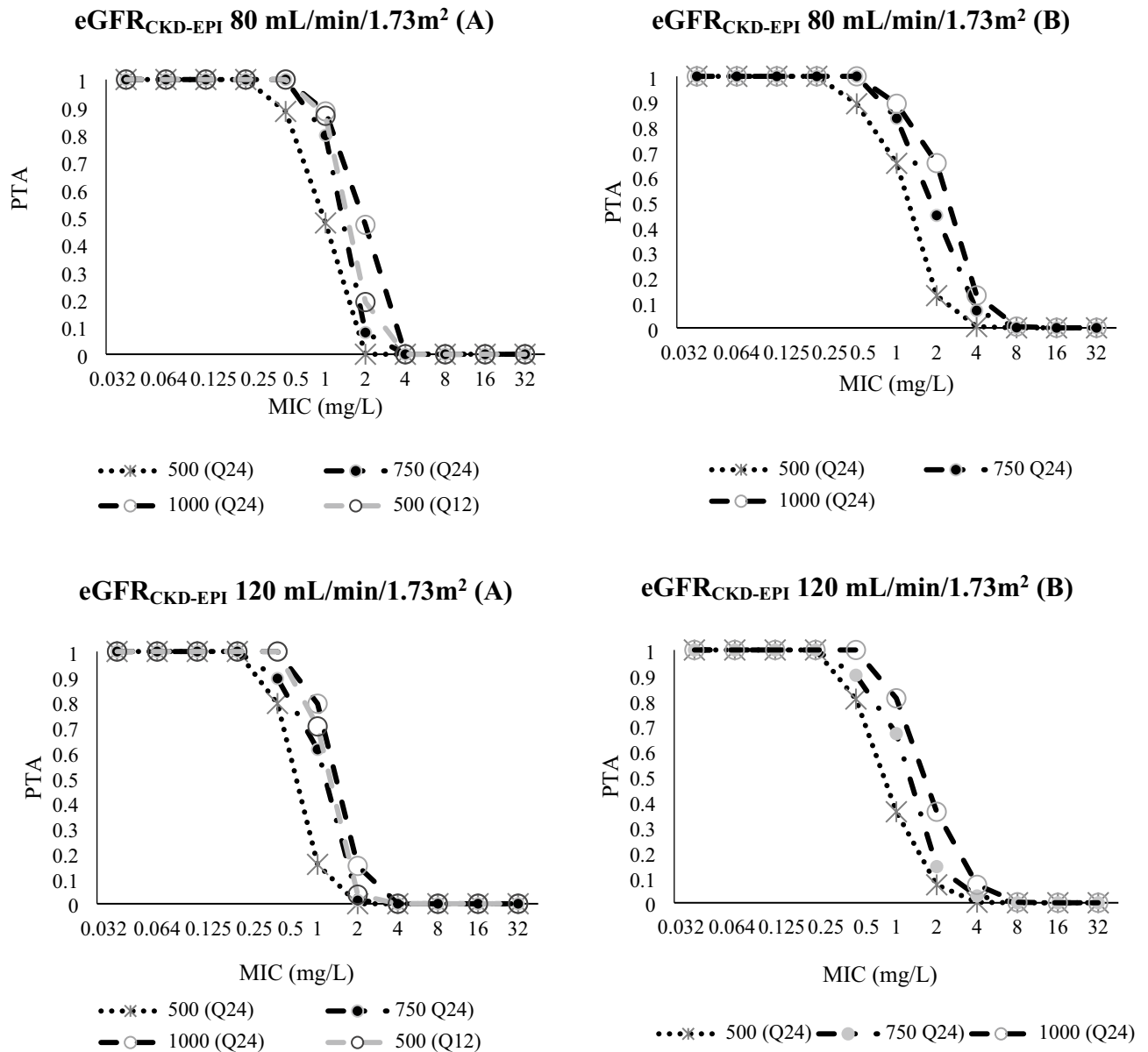


Figure 4. Probability of target attainment (PTA; $fAUC/MIC \geq 80$) of several dosage regimens of levofloxacin at (A) the first 24-h and (B) steady state; for patients with $eGFR_{CKD-EPI} 80 \text{ mL/min/1.73m}^2$ and $eGFR_{CKD-EPI} 120 \text{ mL/min/1.73m}^2$; Dotted horizontal line represent 90% of PTA which considered as a successful or acceptable; For the steady state (B) in both CKD-EPI groups: the lines for levofloxacin 500 (Q12) was relatively similar to 1000 (Q24). To provide better clarity, lines with similarity shapes were not presented.

higher than $80 \text{ mL/min/1.73m}^2$, our highest simulated dosing regimen could not provide adequate PTA against pathogens with an $MIC \geq 1 \text{ mg/L}$.

Given the MIC susceptibility profile and therefore more easily attainable PK/PD targets, we have demonstrated that a variety of levofloxacin dosing regimens can potentially achieve relatively optimal FTA against *H. influenzae*, *M. cattarhalis*, *E. coli*, *K. pneumoniae*. The borderline FTA (around 85%) could be potentially attainable against *S. aureus*. Of the pathogens studied, effective levofloxacin exposure against *P. aeruginosa* and *S. pneumoniae* are concerning. A high percentage of *P. aeruginosa* (45.7%) and *S. pneumoniae* (79.9%) in the EUCAST's MIC distribution data had $MIC \geq 1 \text{ mg/L}$ and this may contribute to the reported lower FTA attainment¹⁸. This may indicate that levofloxacin may not be a good choice to be used as an empirical therapy for both ICU and non-ICU patients where *P. aeruginosa* and *S. pneumoniae* are predominantly reported in Indonesian hospitals. Nevertheless, our study highlights the potential of using levofloxacin as targeted therapy in infections caused by susceptible strains of *P. aeruginosa* and *S. pneumoniae*. A dose of 750 mg every 24 h and 1,000 mg every 24 h may provide adequate exposure for Indonesian patients with $eGFR_{CKD-EPI} 50 \text{ mL/min/1.73m}^2$ and $\geq 80 \text{ mL/min/1.73m}^2$. Combination with other antibiotics or other broader antibiotics may be considered at the time when MIC is not known and then de-escalation to levofloxacin monotherapy with appropriately adjusted dosing after MIC is in hand. Furthermore, with the increasing number of LRTIs caused by *Stenotrophomonas maltophilia*

Levofloxacin dosage regimens	FTA (%) by bacteria and eGFR _{CKD-EPI} (mL/min/1.73m ²)																			
	<i>E. coli</i>				<i>K. pneumoniae</i>				<i>P. aeruginosa</i>				<i>S. aureus</i>				<i>S. pneumoniae</i>			
	20		50		20		50		20		50		20		50		20		50	
	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b
500 (Q48)	88	88	86	87	85	88	80	82	66	73	52	56	82	83	81	81	72	83	33	51
750 (Q48)	88	89	88	88	88	91	85	86	74	80	65	68	83	84	82	82	88	98	71	75
1000 (Q48)	89	89	88	88	90	92	87	88	79	84	70	73	83	87	83	83	98	98	81	83
500/250 (Q48)	88	87	86	85	85	83	80	76	66	58	52	35	82	81	81	73	72	56	33	14
750/500 (Q48)	88	88	88	87	88	88	85	83	74	74	65	57	83	83	82	81	88	84	71	52
500/250 (Q24)	88	88	88	87	87	88	85	83	70	73	65	57	82	83	82	81	83	85	71	52
500 (Q24)	-	-	88	88	-	-	85	88	-	-	65	74	-	-	82	83	-	-	71	85
750 (Q24)	-	-	88	89	-	-	88	91	-	-	73	80	-	-	83	85	-	-	88	97

Levofloxacin dosage regimens	FTA (%) by bacteria and eGFR _{CKD-EPI} (mL/min/1.73m ²)																			
	<i>E. coli</i>				<i>K. pneumoniae</i>				<i>P. aeruginosa</i>				<i>S. aureus</i>				<i>S. pneumoniae</i>			
	80		120		80		120		80		120		80		120		80		120	
	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b
500 (Q24)	87	87	86	87	83	85	80	82	59	63	50	55	81	82	80	81	54	68	28	44
750 (Q24)	88	88	87	87	87	88	84	85	68	74	61	64	82	84	81	82	81	85	64	69
1000 (Q24)	88	89	88	88	89	90	87	88	75	78	69	73	83	84	82	83	88	90	81	82
500 (Q12)	88	89	88	88	87	90	86	87	71	77	66	72	83	84	82	83	86	89	73	80

Table 3. FTA for various dosage regimens of levofloxacin against the empiric EUCAST MIC distributions of several Gram positive and Gram negative bacteria for patients with eGFR_{CKD-EPI} 20, 50, 80, 120 mL/min/1.73m². – not simulated, *a* simulation at the first dose, *b* simulation at the steady state, Q48 given every 48 h, Q24 given every 24 h, Q12 given every 12 h.

Levofloxacin dosage regimens	FTA (%) by bacteria and eGFR _{CKD-EPI} (mL/min/1.73m ²)															
	<i>P. aeruginosa</i>								<i>S. pneumoniae</i>							
	20		50		80		120		20		50		80		120	
	a	b	a	b	a	b	a	b	a	b	a	b	a	b	a	b
500 (Q48)	79	88	62	68	-	-	-	-	73	84	33	51	-	-	-	-
750 (Q48)	89	96	78	82	-	-	-	-	89	99	71	7	-	-	-	-
1000 (Q48)	95	97	85	88	-	-	-	-	99	99	82	84	-	-	-	-
500/250 (Q48)	79	69	62	42	-	-	-	-	73	56	33	14	-	-	-	-
750/500 (Q48)	89	89	78	69	-	-	-	-	89	85	72	52	-	-	-	-
500/250 (Q24)	84	89	79	69	-	-	-	-	84	86	72	52	-	-	-	-
500 (Q24)	-	-	79	89	71	76	65	72	-	-	72	85	54	69	40	58
750 (Q24)	-	-	89	96	83	89	80	84	-	-	89	98	82	85	73	79
1000 (Q24)	-	-	-	-	90	93	87	90	-	-	-	-	89	91	86	88
500 (Q12)	-	-	-	-	86	93	82	90	-	-	-	-	87	90	80	86

Table 4. FTA for various dosage regimens of levofloxacin against the directed EUCAST MIC distributions of several gram positive and negative bacteria for patients with eGFR_{CKD-EPI} 20, 50, 80, and 120 mL/min/1.73m². – not simulated, *a* simulation at the first dose, *b* simulation at the steady state, Q48 given every 48 h, Q24 given every 24 h, Q12 given every 12 h.

particularly in Asia–Pacific region, dosing simulations against the MIC distribution of this biofilm-forming bacterium could be considered as an important area for future research²³.

It should be noted that there are some limitations in our study. First, we could not collect all blood samples within the precise time specifications as originally planned due to some technical challenges. These unstructured blood samplings are prone to cause imprecision in estimating the PK of levofloxacin. However, by having a mean of 4 to 5 blood samples per-patient and employing a population PK approach with Bayesian priors, we believe that our analysis provides a reasonable population PK estimation. Second, our PK model may not truly describe the distribution of levofloxacin PKs in hospitalized Indonesian patients given that only 26 patients were included in the study. We performed Monte Carlo simulation to virtually enlarge the sample sizes and predicted the probability of several levofloxacin dosing regimens to attain adequate PD exposure. In the settings where the estimated PKs from a substantial number of sample size are unavailable, Monte Carlo simulation could be considered as a rational approach to identify the achievement of PK/PD exposure from varied dosing regimens.

Third, even though levofloxacin in our study was given predominantly to patients with pneumonia, we could not measure the concentration of levofloxacin at the site of infection. Given that PK/PD indices are most commonly based on blood exposures of drug and that levofloxacin extensively penetrates into the epithelial lining fluid⁶, we believe that our findings are translatable. Fourth, we used MIC data from EUCAST database in our dosing simulations and these may not represent the MIC distributions in Indonesian settings. Therefore, caution should be taken when extrapolating these results to Indonesian hospitals, with the recommendation being that each facility consider local antibiogram data when making therapeutic and dosing decisions involving levofloxacin. It is worth mentioning though that susceptibility surveillance is not adequately conducted in every hospital setting in developing countries such as Indonesia^{24,25}. Finally, although we included patients with $eGFR_{CKD-EPI} \leq 20$ mL/min/1.73m², our findings might not be applicable for those with renal replacement therapy (RRT) as they were excluded from our study.

Conclusions

We have described the PK of levofloxacin in Indonesian hospitalized patients. Changes in CL of levofloxacin in our study was significantly influenced by changes in $eGFR_{CKD-EPI}$ and dose adjustment should be made accordingly. Our simulations found that an acceptable PTA ($\geq 90\%$) could be obtained against a MIC of ≤ 0.5 mg/L among all simulated $eGFR_{CKD-EPI}$ values. While for an MIC of ≥ 1 mg/L, a PTA of 90% is likely difficult to attain, in particular among those patients with $eGFR_{CKD-EPI}$ of 120 mL/min/1.73m². Higher doses of levofloxacin provide adequate coverage against majority LRTI pathogens, however, high doses are needed to achieve acceptable FTAs against *P. aeruginosa* and *S. pneumoniae*.

Methods

Study design and setting. A prospective observational PK study was conducted in two Indonesian hospitals from November 2018 to November 2019. Patients aged ≥ 18 years old admitted to the intensive care unit (ICU) and non-ICU wards and receiving intravenous levofloxacin were included in this study. Patients with a plan for RRT or extracorporeal membrane oxygenation (ECMO) at the time of sampling and/or known to be pregnant were excluded. The study protocol was approved by the Ethics Committee of Dr. Ramelan Navy Hospital (approval number 76/EC/KERS/2019) and The University of Queensland Human Research Ethics Committee (approval number 2018001592). Written informed consent from the patient or legal substitute decision-maker was obtained prior to sampling.

Drug administration, sampling procedure, and data collection. Levofloxacin was administered as a 30-min intermittent infusion and the dosing regimens were at the discretion of the treating team. The aim was to obtain six blood samples (each sample: 3 mL, using lithium heparin as an anticoagulant) per-patient after the administration of 500 or 750 mg of levofloxacin intravenously during one dosing interval. All blood specimens were immediately centrifuged for 15 min at 3000 rpm after sampling and the aliquots were immediately frozen at -20 °C. All frozen aliquots were further moved to -80 °C within one week after the centrifugation.

The dose given, time of administration, number of blood samples and number of prior levofloxacin doses were recorded. Demographic data, including gender, age, body weight, and laboratory data, including serum creatinine (Se_{Cr}) and albumin, were collected from patients' medical records. The estimated glomerular filtration rate (eGFR) was calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation²⁶. For ICU patients, the use of mechanical ventilation was recorded. All data were recorded on the day of recruitment. All medications administered concomitantly with the levofloxacin were recorded and further screened for potential interactions with levofloxacin²⁷. Interactions classified as "avoid combination" and "usually avoid combination" in the reference used in our study were considered as clinically relevant interactions²⁷.

Bioanalytical methods. Determination of levofloxacin concentrations in plasma was performed by a validated ultra-high performance liquid chromatography with tandem mass spectrometry (UHPLC-MS/MS) method on a Nexera liquid chromatograph connected to a 8030+ triple quadrupole mass spectrometer (Shimadzu, Kyoto, Japan). Test samples were assayed in batches alongside calibrators and quality controls, and results were subject to batch acceptance criteria. Sample (10 μ L) was spiked with internal standard (ciprofloxacin) and protein was precipitated using acetonitrile. An aliquot of 0.2 μ L of the supernatant was injected onto the UHPLC-MS/MS instrument. The stationary phase was a Kinetex C8 100 \times 2.1 mm (1.7 μ m) analytical column preceded by a SecurityGuard-Ultra C8 guard cartridge (Phenomenex, Torrance, USA). Mobile phase A was 0.2% formic acid in water (v/v), and mobile phase B was 0.2% formic acid in acetonitrile (v/v). Separations were effected with a gradient from 10 to 80% of mobile phase B at a flow of 0.3 mL/min, producing back pressure of approximately 3600 psi. Levofloxacin was monitored in positive mode at the mass-to-charge ratio (m/z) of 362.0 \rightarrow 318.1. The standard internal ciprofloxacin was monitored in positive mode at m/z of 332.2 \rightarrow 314.1. The assay method was linear from 0.1 to 50 mg/L (precision of 10.2% and accuracy of -4.2% at LLOQ of 0.1 mg/L). Precision of 5.9, 2.1, and 5.8% and accuracy of 6.4, 0.9, and -4.7% at levofloxacin concentrations of 0.3, 2 and 40 mg/L. The precision and accuracy of LLOQ and QCs and the matrix effect validation met the US FDA guidelines²⁸.

Population pharmacokinetic data analysis. Levofloxacin plasma concentration–time data were fitted to generate the population PK model using non-parametric adaptive grid (NPAG) algorithm in Pmetrics[®] software (version 1.9; Laboratory of Applied Pharmacokinetics and Bioinformatics, Los Angeles, CA, USA) for R 3.41²⁹. Classic one-compartment and two-compartment models, with intercompartmental distribution, represented either as $K_{PC}-K_{CP}$ or Q were initially evaluated as a potential PK model. First-order processes were used to

describe the elimination of levofloxacin from the central compartment and inter-compartmental distribution in two-compartment model. Both lambda (ranging from 0.1 to 0.9) and gamma (ranging from 1 to 9) error models were tested for each PK model.

The following were evaluated as potential covariates for volume of distribution (V_d) and clearance (CL) of levofloxacin using linear, exponential regression and power (using population-median-normalised and allometric) model: gender, Se_{Cr} , hospitalisation type (ICU and non-ICU), mechanical ventilation, and $eGFR_{CKD-EPI}$. The value of 0.75 was used as a coefficient in the allometric model for age, Se_{Cr} , and $eGFR_{CKD-EPI}$. The final estimated PK parameters are presented as mean, standard deviation (SD), percentage coefficient of variation (%CV), and median value. The %CV was used to describe inter-individual PK variability.

PK model diagnostics. The goodness of fit of the PK model was assessed using inspection of observed versus predicted plots, both population and individual predictions. The following indicators were used to identify the best structural and error model: (1) improvement of the scatterplot, (2) improvement of the intercept (close to 0) and slope (close to 1) of linear regression, (3) an increased value of the coefficient of determination (r^2 ; close to 1) of the linear regression. In addition to this, a statistical reduction of -2 log-likelihood ($-2LL$; a decrease value of 3.84 corresponds to $P < 0.05$) and the lowest value of Akaike Information Criteria (AIC) scores were also used to choose the best structural and error model. Once the structural model was chosen, each covariate was separately added to that particular model. Only covariates that could improve the scatterplot, r^2 , intercept, slope, and a statistically significant improvement of $-2LL$ and AIC was retained in the final model. The internal validation of the final model with covariates was assessed by a visual predictive check (VPC) with 1000 simulations. The distribution of the observed concentration in this simulation was plotted and visually examined.

Dosing simulations. Monte Carlo simulations ($n = 1000$) were undertaken using Pmetrics® to identify the probability of target attainment (PTA) of achieving the a priori PK/PD target both at first 24-h and at the fifth day of levofloxacin treatment (steady state) against a specific MIC value ranging from 0.004 to 512 mg/L. The a priori PK/PD target was $fAUC_{0-24}/MIC \geq 80$ and the protein binding of levofloxacin was set at 30%^{9,30}. The simulations were conducted in four different $eGFR_{CKD-EPI}$ levels, including 20, 50, 80, and 120 mL/min/1.73m². The standard dosing regimens of levofloxacin for each $eGFR_{CKD-EPI}$ level were simulated, including 500 mg i.v. followed by 250 mg every 48 h, 750 mg followed by 500 mg every 48 h, 500 mg followed by 250 mg every 24 h, 500 mg every 48 h, and 750 mg every 48 h (for $eGFR_{CKD-EPI}$ 20 and 50 mL/min/1.73m²); 500 mg every 24 h and 750 mg every 24 h ($eGFR_{CKD-EPI}$ 50, 80, and 120 mL/min/1.73m²)^{20,31}. In addition, intensified dosing regimens up to the highest safe daily dose found in the literature (1000 mg/day) were also simulated for each $eGFR_{CKD-EPI}$ level group, including 1000 mg every 48 h (for $eGFR_{CKD-EPI}$ 20 and 50 mL/min/1.73m²); 500 mg every 12 h and 1000 mg every 24 h ($eGFR_{CKD-EPI}$ 80, and 120 mL/min/1.73m²)³². All PTA simulations were assessed on the first day of treatment (first 24 h) and at steady state (defined as the fifth day of treatment). A PTA value $\geq 90\%$ for a particular MIC value was considered optimal in our study.

To calculate the fractional target attainment (FTA), the PTA of each dosing regimen was compared against the MIC distribution of pathogens, commonly causative of LRTIs, obtained from the European Committee on Antimicrobial Susceptibility Testing (EUCAST) database (available from www.eucast.org; accessed 01 August 2020)¹⁸. The empirical FTA was calculated considering the whole range of MIC distribution of *Escherichia coli* ($n = 9144$), *Klebsiella pneumoniae* ($n = 3674$), *Haemophilus influenzae* ($n = 22,910$), *Pseudomonas aeruginosa* ($n = 14,871$), *Moraxella catarrhalis* ($n = 5259$), *Staphylococcus aureus* ($n = 27,556$), *Streptococcus pneumoniae* ($n = 85,564$). While the directed FTA was calculated by considering a range of MICs of the susceptible strains of each pathogen. Any dosage regimen that achieved the acceptable FTA (i.e. $\geq 90\%$) was considered a successful dosage regimen either for directed or empirical levofloxacin therapy.

Statistical analysis. Descriptive analysis using frequencies (%) for categorical data and mean (\pm standard deviation; SD) for continuous data in the demographic of patients were conducted using Microsoft Excel v2016.

Ethics approval. Approval was obtained from Ethics Committee of Dr. Ramelan Navy Hospital (approval number 76/EC/KERS/2019) and The University of Queensland Human Research Ethics Committee (approval number 2018001592). The procedures used in this study adhere to the tenets of the Declaration of Helsinki.

Consent to participate and for publication. Written informed consent from the patient or legal substitute decision-maker was obtained prior to sampling.

Data availability

The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

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References

1. GBD 2016 Lower Respiratory Infections Collaborators: Estimates of the global, regional, and national morbidity, mortality, and aetiologies of lower respiratory infections in 195 countries, 1990–2016: a systematic analysis for the Global Burden of Disease Study 2016. *Lancet. Infect Dis.* **18**(11), 1191–1210 (2018).

2. Kalil, A. C. *et al.* Management of adults with hospital-acquired and ventilator-associated pneumonia: 2016 clinical practice guidelines by the Infectious Diseases Society of America and the American Thoracic Society. *Clin. Infect. Dis.* **63**(5), e61–e111 (2016).
3. Metlay, J. P. *et al.* Diagnosis and treatment of adults with community-acquired pneumonia: An official clinical practice guideline of the American Thoracic Society and Infectious Diseases Society of America. *Am J. Respir. Crit. Care Med.* **200**(7), e45–e67 (2019).
4. Indonesian Medical Association. *Panduan praktik klinis bagi dokter di fasilitas pelayanan kesehatan primer* (Indonesian Medical Association, 2017).
5. Blondeau, J. M. Expanded activity and utility of the new fluoroquinolones: a review. *Clin. Ther.* **21**(1), 3–40 (1999).
6. Boselli, E. *et al.* Pharmacokinetics and intrapulmonary diffusion of levofloxacin in critically ill patients with severe community-acquired pneumonia. *Crit. Care Med.* **33**(1), 104–109 (2005).
7. Paiva, J. A. & Pereira, J. M. Fluoroquinolones: another line in the long list of their collateral damage record. *Crit. Care Med.* **43**(3), 708–710 (2015).
8. Wispelwey, B. Clinical implications of pharmacokinetics and pharmacodynamics of fluoroquinolones. *Clin. Infect. Dis.* **41**(Suppl 2), S127–135 (2005).
9. Abdul-Aziz, M. H. *et al.* Antimicrobial therapeutic drug monitoring in critically ill adult patients: A position paper. *Intensive Care Med.* **46**(6), 1127–1153 (2020).
10. Zhang, Y. *et al.* Population pharmacokinetics of intravenous levofloxacin 500mg/day dosage in infected patients. *Pharmazie.* **69**, 553–557 (2014).
11. Zhang, J. *et al.* Population pharmacokinetics of oral levofloxacin 500 mg once-daily dosage in community-acquired lower respiratory tract infections: results of a prospective multicentre study in China. *J. Infect. Chemother.* **15**, 293–300 (2009).
12. Kiem, S. *et al.* Population pharmacokinetics of levofloxacin in Korean patients. *J. Chemother.* **28**(4), 308–313 (2016).
13. Roberts, J. A., Cotta, M.O., Cojutti, P., Lugano, M., Della Rocca, G. & Pea, F. Does critical illness change levofloxacin pharmacokinetics?. *Antimicrob. Agents. Chemother.* **60**(3), 1459–1463 (2015).
14. Cojutti, P. G. *et al.* Population Pharmacokinetics and pharmacodynamics of levofloxacin in acutely hospitalized older patients with various degrees of renal function. *Antimicrob. Agents. Chemother.* **61**(3), e02134–e2216 (2017).
15. Preston, S. L. *et al.* Levofloxacin population pharmacokinetics and creation of a demographic model for prediction of individual drug clearance in patients with serious community-acquired infection. *Antimicrob. Agents. Chemother.* **42**(5), 1098–1104 (1998).
16. Chien, S. C. *et al.* Pharmacokinetic profile of levofloxacin following once-daily 500-milligram oral or intravenous doses. *Antimicrob. Agents. Chemother.* **41**(10), 2256–2260 (1997).
17. Chien, S. C. *et al.* Double-blind evaluation of the safety and pharmacokinetics of multiple oral once-daily 750-milligram and 1-gram doses of levofloxacin in healthy volunteers. *Antimicrob. Agents. Chemother.* **42**(4), 885–888 (1998).
18. European Committee on Antimicrobial Susceptibility Testing. *Data from the EUCAST MIC distribution website* <http://www.eucast.org> (2020).
19. Fish, D. N. & Chow, A. T. The clinical pharmacokinetics of levofloxacin. *Clin. Pharmacokinet.* **32**(2), 101–119 (1997).
20. United State Food and Drug Administration. Highlights of prescribing information: Levaquin® https://www.accessdata.fda.gov/drugsatfda_docs/label/2008/021721s020_020635s57_020634s52_lbl.pdf (2008).
21. Dowling, T. C., Matzke, G. R., Murphy, J. E. & Burckart, G. J. Evaluation of renal drug dosing: Prescribing information and clinical pharmacist approaches. *Pharmacotherapy* **30**(8), 776–786 (2010).
22. Faull, R. & Lee, L. Prescribing in renal disease. *Aust. Prescr.* **30**(1), 17–20 (2007).
23. Chang, Y. T., Lin, C. Y., Chen, Y. H. & Hsueh, P. R. Update on infections caused by *Stenotrophomonas maltophilia* with particular attention to resistance mechanisms and therapeutic options. *Front. Microbiol.* **6**, 893 (2015).
24. Ayuuekbong, J. A., Ntemgwa, M. & Atabe, A. N. The threat of antimicrobial resistance in developing countries: Causes and control strategies. *Antimicrob. Resist. Infect. Control.* **6**, 47 (2017).
25. Chokshi, A., Sifri, Z., Cennimo, D. & Horng, H. Global contributors to antibiotic resistance. *J. Glob. Infect. Dis.* **11**(1), 36–42 (2019).
26. Levey, A. S. *et al.* A new equation to estimate glomerular filtration rate. *Ann. Intern. Med.* **150**(9), 604–612 (2009).
27. Hansten, P. D. & Horn, J. R. *Drug interactions analysis and management* (Wolters Kluwer Health Inc., 2014).
28. U.S Department of Health and Human Services, Food and Drug Administration, Center for Drug Evaluation and Research, Center for Veterinary Medicine. *Bioanalytical method validation: guidance for industry* <https://www.fda.gov/files/drugs/published/Bioanalytical-Method-Validation-Guidance-for-Industry.pdf> (2018).
29. Neely, M. N., van Guilder, M. G., Yamada, W. M., Schumitzky, A. & Jelliffe, R. W. Accurate detection of outliers and subpopulations with Pmetrics, a nonparametric and parametric pharmacometric modeling and simulation package for R. *Ther. Drug. Monit.* **34**(4), 467–476 (2012).
30. Anderson, V. R. & Perry, C. M. Levofloxacin: a review of its use as a high-dose, short-course treatment for bacterial infection. *Drugs* **68**(4), 535–565 (2008).
31. Stanford Antimicrobial Stewardship Safety and Sustainability Program. *Stanford Health Care antimicrobial dosing reference guide* http://med.stanford.edu/bugsanddrugs/dosingprotocols/_jcr_content/main/panel_builder/panel_0/download/file.res/SHC%20ABX%20Dosing%20Guide.pdf (2019).
32. Noel, G. J. *et al.* Measuring the effects of supratherapeutic doses of levofloxacin on healthy volunteers using four methods of QT correction and periodic and continuous ECG recordings. *J. Clin. Pharmacol.* **44**(5), 464–473 (2004).

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Competing interests

The authors declare no competing interests.

Additional information

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