Saturable elimination of piperacillin in critically ill patients: implications for continuous infusion


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Highlights

- Elimination of piperacillin (PIP) is saturable at therapeutic concentrations
- Same dose continuous PIP results in lower exposure compared with intermittent PIP
- Intermittent vs continuous PIP trials may be biased towards intermittent PIP
Saturable elimination of piperacillin in critically ill patients: implications for continuous infusion

1Dhaese S AM, 2,3 Colin P, 1Willems H, 4,5,6 Heffernan A, 1Gadeyne B, 7Van Vooren S, 1Depuydt P, 1Hoste E, 7,8Stove V, 7,8 Verstraete A G, 4,9,10 Lipman J, 4,6,9,11 Roberts J A, 11De Waele J J

1. Ghent University Hospital, Department of Critical Care Medicine, Ghent, Belgium
2. University of Groningen, University Medical Center Groningen, Department of Anesthesiology, Groningen, The Netherlands.
3. Ghent University, Laboratory of Medical Biochemistry and Clinical Analysis, Ghent, Belgium
4. University of Queensland Centre for Clinical Research, Faculty of Medicine, The University of Queensland, Brisbane, Australia
5. School of Medicine, Griffith University, Southport, Australia
6. Centre for Translational Anti-infective Pharmacodynamics, School of Pharmacy, The University of Queensland, Brisbane, Australia
7. Ghent University, Department of Diagnostic Sciences, Ghent, Belgium
8. Ghent University Hospital, Department of Laboratory Medicine, Ghent, Belgium
9. Royal Brisbane and Women’s Hospital, Department of Intensive Care Medicine, Brisbane, Australia
10. CHU Nîmes, Department of Anesthesiology and Critical Care, Nîmes, France
11. Royal Brisbane and Women’s Hospital, Department of Pharmacy, Brisbane, Australia
Address correspondence to:

Sofie Dhaese
C. Heymanslaan 10
9000 Ghent
Belgium

sofie.dhaese@ugent.be

+32 (0)9 332 28 70
Abstract

Purpose: To evaluate saturation of piperacillin elimination in adult critically ill patients.

Patients and methods: Seventeen adult critically ill patients received continuous and intermittent infusion piperacillin/tazobactam. Piperacillin plasma concentrations (n=217) were analyzed using population pharmacokinetic (PopPK) modeling. Post hoc simulations were performed to evaluate the type I error rate associated with our study. Unseen data was used to validate the final model. The mean error (ME) and root mean squared error (RMSE) were calculated as a measure of bias and imprecision respectively.

Results: A PopPK model with parallel linear and non-linear elimination best fitted our data. The median and 95% confidence intervals for model parameters drug clearance (CL), volume of the central compartment (V), volume of the peripheral compartment (Vp) and intercompartmental clearance (Q) were 9 (7.69 – 11) L/h, 6.18 (4.93 – 11.2) L, 11.17 (7.26 – 12) L and 15.61 (12.66 – 23.8) L/h. The Michaelis-Menten constant (Km) and the maximum elimination rate for Michaelis-Menten elimination (Vmax) were estimated without population variability in the model to avoid overfitting and inflation of the type I error rate. The population estimates for Km and Vmax were 37.09 mg/L and 353.57 mg/h respectively.

The ME was -20.8 (95% CI -26.2; -15.4) mg/L while imprecision (RMSE) was 49.2 (95% CI 41.2; 56) mg/L.

Conclusion: Piperacillin elimination is (partially) saturable. Moreover, the population estimate for Km lies within the therapeutic window and therefore saturation of elimination should be accounted for when defining optimum dosing regimens for piperacillin in critically ill patients.

Keywords: piperacillin, pharmacokinetics, critically ill, saturation
Introduction

The ureidopenicillin piperacillin combined with the beta-lactamase inhibitor tazobactam is frequently used to treat serious infections in critically ill patients [1,2]. In line with other beta-lactam antibiotics, piperacillin has time-dependent killing properties. The time (T) for which the free (f) concentration of piperacillin remains above the minimal inhibitory concentration (MIC) is the pharmacokinetic/pharmacodynamic (PK/PD) index of choice, i.e. %fT>MIC [3].

In the past few years, a wealth of evidence emerged demonstrating that the PK of antimicrobial drugs in critically ill patients is profoundly different from the PK of antimicrobial drugs in healthy volunteers or non-critically ill patients [4]. For beta-lactam antibiotics specifically, changes in volume of distribution and/or changes in renal function in critically ill patients may lead to considerable between- and within-patient PK variability [5]. Previously, a pharmacokinetic point-prevalence study of beta-lactam antibiotics in the ICU reported that 16% of the ICU patients did not achieve the PK/PD target of 50%fT>MIC [6]. As suboptimal antimicrobial use may lead to poor infection outcome, efforts are made to optimize the use of beta-lactam antibiotics [7–9]. Because beta-lactam antibiotics have time-dependent killing properties, prolonging the duration of beta-lactam infusion and thereby extending the time the concentration remains above the MIC, was recently introduced in clinical practice [10,11].

Currently, there is an ongoing debate on whether or not piperacillin elimination is saturable at therapeutic plasma concentrations [12–19]. This mechanism is particularly relevant in the context of the recent introduction of prolonged infusion of beta-lactam antibiotics. Indeed, saturation of piperacillin elimination at therapeutic plasma concentrations implies that, for the total antibiotic exposure in a patient to be the same, a higher daily dose could be necessary when piperacillin is infused continuously as opposed to intermittently. In
clinical practice however, the total daily dose of piperacillin is usually not adapted based on the mode of infusion used [11,20].

The aim of this study was to investigate saturation of piperacillin elimination in critically ill patients receiving both intermittent and continuous infusion piperacillin.

Patients and methods

1. Patients

This prospective interventional study was conducted in the Department of Critical Care Medicine of Ghent University Hospital (Ghent, Belgium). Ethical approval was obtained from the Ghent University Hospital Ethics Committee (registration number 2017/1354). Informed consent was signed by patients or their representatives. Patients were eligible for inclusion if they were admitted to the surgical or medical ICU and received piperacillin/tazobactam (TZP) in continuous infusion. Patients younger than 18 years of age and patients receiving extracorporeal membrane oxygenation (ECMO) or renal replacement therapy (RRT) during antibiotic therapy were excluded from the study. Creatinine clearance was determined by measuring urinary creatinine concentrations from an 8-hour urinary collection using an indwelling urinary catheter. Piperacillin antibiotic concentrations and additional data such as, biochemistry, demographic data, the modified Sequential Organ Failure Assessment score (SOFA) on the day of sampling, the Acute Physiology and Chronic Health Evaluation (APACHE II) score on admission and ICU survival were prospectively recorded via REDCap [21].

2. Administration of piperacillin antibiotic therapy and sampling

All patients received both continuous and intermittent infusion TZP. TZP dosing was as follows: loading dose of 4/0.5 g /30 min immediately followed by a continuous TZP
infusion: (measured creatinine clearance ($CL_{CR}$) < 15 mL/min: 8/1 g/24 h, $CL_{CR}$ 15-29 mL/min: 12/1.5 g/24h and for a $CL_{CR} \geq 30$ mL/min 16/2 g/24h). At the end of the antibiotic course as indicated by the treating physician, after a 3-hour washout period, a short infusion (0.5 h; 4500 g) of TZP was administered. In total, 13 samples were collected from every patient. The first two samples were taken 2 hours prior to and immediately before stopping the continuous infusion. Samples 3-13 were collected immediately before administration of the intermittent infusion and after 5, 30, 45, 60, 90, 120, 180, 240, 300 and 360 minutes as shown in Figure 1.

3. **Bioanalysis of piperacillin plasma concentrations**

Arterial blood collected in 4 mL blood tubes (lithium heparin blood collection tubes, BD Vacutainer®, BD Diagnostics, Erembodegem, Belgium) was sent to the core laboratory of the Dept. of Laboratory Medicine at the Ghent University Hospital where they were first stored in a refrigerator at 4°C until they were collected by the toxicology laboratory technicians. Storage at 4°C was never longer than 24 hours. After transferring to an Eppendorf tube, plasma samples were centrifuged at 16162xg for 8 minutes (Microfuge 16, Beckman Coulter, Brea, California). Immediately afterwards, the plasma samples were stored at -20°C until analysis. All samples were analyzed within 1 week. The plasma concentration of piperacillin was determined by ultra-performance liquid chromatography tandem mass spectrometry (UPLC – MS/MS). Tazobactam concentrations were not analyzed in this study. The lower limit of quantification (LLOQ) for piperacillin was 1.09 mg/L, the within-run assay imprecision at LLOQ level was 3.7 %CV and the between-run assay imprecision at the LLOQ level was 8.1 %CV [22].
4. **Population pharmacokinetic model building**

Piperacillin concentration-time data were analyzed using Pmetrics (version 1.5.2; Laboratory of Applied Pharmacokinetics, Los Angeles, CA, USA), an R-based software program for non-parametric and parametric pharmacokinetic-pharmacodynamic population and individual modelling and simulation. We used the non-parametric adaptive grid (NPAG) algorithm to build a PopPK model for piperacillin administered via continuous and intermittent infusion [23]. A digital Fortran compiler was used (Gfortran version 6.1; Free software foundation, Inc. Boston, MA, USA) and the runs were executed using R (version 3.5.1; The R Foundation for Statistical Computing. Vienna, Austria) and RStudio (version 1.1.383; RStudio, Inc. Boston, MA, USA). One- and two compartment models were fitted to the data using subroutines from the Pmetrics library. Modeling concentration-time data with both linear, parallel linear/Michaelis-Menten and Michaelis-Menten drug clearance was attempted. Subsequently, the statistical error model with the best fit was selected and a covariate model was developed. Covariates *a priori* considered for inclusion in the model were: measured creatinine clearance, estimated creatinine clearance (Cockroft-Gault formula), estimated glomerular filtration rate (Modification of Diet in Renal Disease (MDRD) formula), body weight, age, SOFA score and albumin, based on prior knowledge and biological plausibility [4,24–27]. Body weight was included as a primary covariate on all model parameters, except for $K_m$ and $V_{max}$, according to the allometric power model [28].

\[(1) \quad P \theta_i = TVP\theta_1\ast(\text{WEIGHT}/70)^{\text{power}} \quad \text{Eq. 1}\]

Where $P \theta_i$ is the individual parameter value, $TVP\theta_1$ is the parameter value for a typical adult with a body weight of 70kg, and power is an allometric exponent fixed to 0.75 for CL and Q and fixed to 1 for V and Vp. As an initial step, covariates measured creatinine, estimated creatinine clearance via Cockroft-Gault formula and estimated glomerular filtration rate using the MDRD formula were tested on the CL parameter as this is biologically plausible.
However, only one of these was retained as correlated variables may lead to collinearity and inflation of the parameter’s standard error [29]. In a next step, forward selection and backward elimination using the PMstep function in Pmetrics was used to assess the relationship between covariates and model parameters. The log likelihood ratio test (LRT) and the Akaike information criterion (AIC) were considered during model building. More specifically, a difference of 3.84 in the log likelihood was considered significant at the 5% level when performing the likelihood ratio test for comparing nested models. Estimated parameters are reported as mean, percent coefficient of variation (%CV) and median with interquartile range (IQR). The %CV is reported as a measure of between-subject variability in the model parameters. 95% Confidence intervals were estimated via a non-parametric bootstrap (n=1000) and quantify the uncertainty on the parameter estimates.

5. **Pharmacokinetic model diagnostics**

The PopPK model was assessed by visual evaluation of the goodness of fit of the observed versus *a posteriori* predicted plots and the coefficient of determination of the linear regression of the observed-predicted values ($r^2$ close to 1, intercept close to 0) from each run. The predictive performance was assessed on mean prediction error (bias) and the mean bias-adjusted square prediction error (imprecision) of the population predictions.

Internal model validation consisted of a visual predictive check (VPC) plot. The VPC (n=10.000) was performed by overlaying the 95% CI of the simulated profiles for 0.05, 0.5 and 0.95 quantiles with the corresponding quantiles of the observed data.

For external model validation, the final model population parameter distributions were used to predict concentrations for an independent validation dataset. We refer to Dhaese, et al [30] for a detailed description of this validation dataset. Prediction errors were evaluated based on the absolute bias (ME) and imprecision (MSE) as described in equation 2 and 3:
(2) Absolute bias[\(\hat{\theta} \)] (ME) = E[\(\hat{\theta} – \theta\)]  
Eq.2

(3) Absolute imprecision[\(\hat{\theta} \)] (MSE) = E[\((\hat{\theta} – \theta)^2\)]  
Eq.3

Where \(\hat{\theta}\) is the predicted piperacillin concentration and \(\theta\) is the observed concentration. The root mean square prediction error (RMSE) was calculated by taking the square root of MSE.

6. **Comparative AUC\(_u\) simulations for intermittent and continuous infusion dosing regimens**

Monte Carlo simulations (n=1000) were performed with the final PopPK model to compare the unbound (u) area under the curve (AUC\(_u\)) as a measure of total (unbound) drug exposure between intermittent and continuous infusion dosing regimens. Using AUC as a basis to compare intermittent and continuous infusion of beta-lactam antibiotics was previously reported by Firsov and Mattie [31]. Free piperacillin concentrations were calculated assuming a 30% level of protein binding in accordance with previous findings [32]. Four different scenarios were evaluated; i.e. a daily dose of 12/1.5g TZP for a patient with a measured CL\(_{CR}\) of 20mL/min, 16/2g TZP for a patient with a measured CL\(_{CR}\) of 70mL/min, 16/2g TZP for a patient with a measured CL\(_{CR}\) of 130mL/min and 16/2g TZP for a patient with a measured CL\(_{CR}\) of 200mL/min. The body weight for all patients was fixed at 70kg. For each of these four scenarios, both intermittent and continuous infusion dosing regimens were simulated and compared. The AUC\(_u\) was calculated using linear trapezoidal approximation. A 24-hour interval for AUC\(_u\) calculation was chosen after six doses for intermittent infusion and one bolus and five maintenance doses for continuous infusion.

7. **Post hoc estimation of type I error rate**

A type I error rate analysis was performed to evaluate the probability to reject the null-hypothesis (H\(_0\)) in favor of the alternative hypothesis (H\(_1\)) given that it is true, where H\(_0\) =
Piperacillin kinetics are best described by linear elimination and $H_1$: piperacillin kinetics are best described by non-linear elimination. [27]

In short, we simulated concentrations for 17 patients according to the design of this study (drug administration, blood sampling, etc.). For this, the PopPK model by Landersdorfer, *et al* [12] served as the $H_1$, i.e. piperacillin PKs are non-linear and elimination is characterized by a parallel first-order and Michaelis-Menten process. The $H_0$ was simulated by fixing the $V_{\text{max}}$ estimate in the model by Landersdorfer to zero, i.e. removing the non-linear component in piperacillin elimination. This process was repeated 5000 times, resulting in 10,000 simulated datasets. All simulated datasets were fitted with a two-compartmental model with linear elimination and a two-compartmental model with parallel linear and Michaelis-Menten elimination. Both models were compared using the LRT according to equation 4.

$$\text{(4) } \text{LRT} = 2*(L_{Lc} - L_{Lr})$$

where $L_{Lc}$ is the log likelihood (LL) for the more complex model and $L_{Lr}$ is the LL for the reduced model. The difference in the number of parameters between both models was 4 when between-subject variability was included in the estimation of $K_m$ and $V_{\text{max}}$ and was 2 otherwise. When considering the 5% level of significance, the critical values from the chi-square distribution were 9.49 and 5.99, respectively.

The type I error rate was calculated from the number of times the complex model was declared superior over the reduced model for the simulated datasets according to the $H_0$.

8. **Statistical analysis**

All statistical analyses were performed using R and RStudio. Continuous data are presented as median (interquartile range). Categorical data are presented as counts (%).

**Results**
1. **Patients and samples**

In total, 17 patients were included, and 221 samples were collected (Table 1). All patients were enrolled between 5/2/2018 and 18/10/2018. Samples 5-7 were lost for patient 13 and sample 8 was lost for patient 15, therefore only 217 samples were analyzed and used for PK model building. The focus of infection was respiratory in 11 patients, abdominal in 5 patients and bacteremia in 1 patient.

2. **Pharmacokinetic model building and model diagnostics**

Table 2 summarizes the log-likelihood values, the coefficients of determination ($r^2$ values), the AIC’s and the predictive performance of linear, parallel linear and Michaelis-Menten and Michaelis-Menten models (without covariates). Comparison of the coefficient of determination, the bias, imprecision and AIC indicated that the model with parallel linear and Michaelis-Menten kinetics was superior compared to both a model with linear elimination and a model with Michaelis-Menten elimination alone (Table 2).

Including measured creatinine clearance (mCRCL) normalized to 100 mL/min as opposed to estimated creatinine clearance using the Cockroft-Gault or the estimated glomerular filtration rate using the MDRD formula provided the model with the lowest AIC value (Table 3). Forward selection and backward elimination further revealed a relationship between albumin and clearance. However, when including albumin as a covariate on CL, no model improvement in terms of $\Delta$AIC or LRT was noted, hence albumin was not retained as a covariate in the final model.

The final model was described as:

\[
\begin{align*}
(5) \text{CL} &= \text{TVCL} \times \left(\frac{\text{mCL}_{CR}/100}{100}\right) \times (\text{WEIGHT}/70)^{**0.75} \\
(6) \text{V} &= \text{TVV} \times (\text{WEIGHT}/70) \\
(7) \text{Vp} &= \text{TVVp} \times (\text{WEIGHT}/70)
\end{align*}
\]

Eq. 5

Eq. 6

Eq. 7
(8) \( Q = TVQ^*(\text{WEIGHT}/70)^{0.75} \) \hspace{1cm} \text{Eq. 8}

where CL is piperacillin clearance, V is volume of distribution of the central compartment, Vp is volume of distribution of the peripheral compartment and Q is the intercompartmental clearance. TVCL refers to the population typical piperacillin clearance for a 70-kg patient with a mCL\textsubscript{CR} of 100 mL/min, TVV and TVVp refer to the population typical volume of distribution of the central, respectively the peripheral compartment for a 70-kg patient.

The mean, \%CV, median (IQR) and \%95 CI around the median for the population parameter estimates are listed in Table 4. The typical value for \( K_m \) and \( V_{\text{max}} \) was 37.09 mg/L and 353.57 mg/h respectively.

Between-subject variability was not estimated on \( K_m \) and \( V_{\text{max}} \) as this resulted in an over-parameterized model and an unacceptable inflation of the type I error rate (for further details see the section “Post hoc estimation of type I error rate”). Based on the diagnostic plots, the \( \gamma \) multiplicative error model was selected for modelling assay variance. In all model-building runs, each observation was weighted by \( 1/ (\gamma \times \text{SD}^2) \). We set \( \gamma \) equal to 1 initially and allowed Pmetrics to fit the value for the population. The final-cycle \( \gamma \) value was 1.26, indicating some additional process noise. The formula for the \( \gamma \) error model is \( \text{error} = \gamma \times \text{SD} \) where SD is the standard deviation of each observation. SD is modeled by equation 9 and was based on earlier validation work by Carlier, et al [33].

(9) \( \text{SD} = 2 + 0.1 \times C \) \hspace{1cm} \text{Eq. 9}

where C is the concentration of piperacillin.

The \textit{a posteriori} individual and population predicted versus observed plots and the VPC plots are shown in figure 2 and 3 respectively. The results of the Shapiro-Wilk test of normality for the NPDE indicated no violation of normality (\( p=0.195 \)).
The final PopPK models showed a bias (ME) in predicting serum concentrations from the validation dataset of -20.8 (95% CI -26.2 ; -15.4) mg/L while imprecision (RMSE) was 49.2 (95% CI 41.2 ; 56) mg/L. The Bland-Altman plot is shown in figure 4.

3. **Comparative AUC<sub>u</sub> simulations for intermittent and continuous infusion dosing regimens**

In all four scenarios, patients receiving continuous infusion had lower AUC<sub>u</sub> values when compared to simulated patients receiving the same dose via intermittent infusion (figure 5).

4. **Post hoc estimation of type I error rate**

If the between-subject variability was estimated for all model parameters, the type I error rate was 47.9%. If the between-subject variability was estimated for CL, Q, V and Vp and not estimated for K<sub>m</sub> and V<sub>max</sub>, the type I error rate was reduced to 6.6%.

**Discussion**

A PopPK model with parallel linear and Michaelis-Menten elimination of piperacillin best described this data, collected from 17 critically ill patients receiving both intermittent and continuous infusion piperacillin/tazobactam. These findings are in agreement with previous studies in healthy volunteers and non-critically ill patients [12,13,17] and in disagreement with other studies in healthy volunteers and critically ill patients [14,30,34].

Renal excretion of piperacillin is the major pathway of elimination. Approximately 74-89% of the administered dose of piperacillin is eliminated from the body by renal excretion [2,35]. More specifically, Tjandramaga, *et al.* [35] reported that 56-73% of the renally cleared piperacillin is eliminated through tubular secretion, which is a saturable process.
$V_{\text{max}}$ is the maximum elimination rate for Michaelis-Menten elimination and the drug concentration at which the elimination rate is half of the maximum elimination rate is called the Michaelis-Menten constant or $K_m$. Whether or not non-linear elimination of a drug is clinically relevant depends on the value of $V_{\text{max}}$ and $K_m$. Non-linear elimination is a clinically relevant process if saturation occurs at therapeutic concentrations (i.e. $K_m$ within the therapeutic window) and if $V_{\text{max}}$ is high relative to CL, indicating a substantial contribution of the non-linear elimination process to the total body clearance. It is postulated that the non-linear elimination pathway should contribute to at least 20% of the total body clearance for it to be clinically relevant [36]. If $K_m$ is very high, then saturation occurs but not at relevant plasma concentrations and it will therefore have no impact on the optimal dosing regimen [12]. Other researchers have reported $K_m$ estimates of 36.1 mg/L [12], 47.9 mg/L [13] and 90.13 mg/L [17], all well in the range of therapeutic piperacillin plasma concentrations and in line with our estimate of 37.09 mg/L.

The implications of these findings remain to be determined. Several institutions recently moved towards prolonged infusion of beta-lactam antibiotics yet conclusive evidence in favor of prolonged infusion is lacking and new clinical trials are in the pipeline [10,11,20,37]. Saturation of piperacillin elimination at therapeutic plasma concentrations is of particular relevance when randomized clinical trials compare intermittent versus continuous infusion piperacillin. Indeed, if saturation of piperacillin elimination occurs at therapeutic concentrations, clinical trials comparing the same daily dose of intermittent and continuous infusion piperacillin may unwillingly introduce a bias towards intermittent infusion as patients receiving the same daily dose of piperacillin via intermittent infusion may have a higher total antibiotic exposure when compared to patients receiving the same dose of piperacillin via continuous infusion as is demonstrated in the AUC$_{u \ 24}$ calculations using the final PopPK model (figure 5). While AUC$_{u}$/MIC may not be the PD index of choice for beta-
lactam antibiotics, the phenomenon of non-linear kinetics may impact antibiotic concentrations and indirectly also other PD indices such as $T_{>\text{MIC}}$. This study focused on piperacillin but tubular secretion of other beta-lactam antibiotics such as amoxicillin, oxacillin, fluocxacillin, cefazolin and cefuroxime has been reported as well [38,39].

When performing hypothesis testing and PK model selection, control of the type I error rate is pivotal to avoid false positive conclusions. Inflation of the type I error rate is expected when dealing with (very) small datasets [40,41]. In this study, including the between-subject variability on $K_m$ and $V_{max}$ resulted in an over-parameterized model and an unacceptable type I error rate (for further details see the section “Post hoc estimation of the type I error rate”). Therefore, the between-subject variability for $K_m$ and $V_{max}$ was not estimated. As few piperacillin population PK studies incorporate type I error calculations, it is difficult to determine how our findings with regard to the non-linear kinetics of piperacillin relate to the findings of other studies.

This study has several limitations. While our primary goal was to detect non-linear elimination of piperacillin with a low probability of falsely rejecting $H_0$, the between-subject variability was not estimated on $K_m$ and $V_{max}$ as this led to an unacceptable type I error. Determining urinary concentrations of renally eliminated drugs is helpful when non-linear kinetics are expected, however, in this study, piperacillin concentrations were not measured in the urine and no distinction could be made between the renal and non-renal clearance of piperacillin. The validation results indicate that the final model has a bias towards underpredicting antibiotic concentrations. While no bias is to be preferred, in case of underprediction, physicians may be inclined to increase the dose or dosing frequency. Given the low toxicity of beta-lactam antibiotics and the important risk of underdosing in ICU patients, models that underpredict concentrations of beta-lactam antibiotics are usually preferred over models that have bias towards overprediction [42]. Additionally, the sequence
of the infusion modes never changed and all patients received continuous infusion first, followed by intermittent infusion. Hence, a trend in piperacillin clearance over time could not be excluded.

In conclusion, piperacillin elimination was best described by a PopPK model incorporating parallel linear and Michaelis-Menten elimination. Nevertheless, in literature conflicting evidence is found on the importance of non-linear elimination for piperacillin PK. Non-informative study designs, and statistical inference based on over-parameterized models likely contribute to these conflicting findings. Future studies, appropriately powered and with a low type I error rate, should be conducted to provide conclusive evidence on the potential influence of non-linear elimination for piperacillin PK in critically ill patients.

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**Declarations**

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**Competing Interests:** Jeffrey Lipman has been a consultant for MSD, Australia and Pfizer. Jason Roberts has been a consultant for Accelerate Diagnostics, Astellas, Bayer, bioMerieux and MSD as well as having received investigator-initiated grants from MSD, The Medicines Company and Cardeas Pharma. Jan De Waele has been consultant for Accelerate Diagnostics, Bayer Healthcare, MSD and Pfizer.

**Ethical Approval:** Ghent University Ethics Committee (2017/1354)

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[38] Landersdorfer CB, Kirkpatrick CM, Kinzig M, Bulitta JBB, Holzgrabe U, Sörgel F.


Figures

Figure 1: Administration of piperacillin and timing of sampling

Figure 2: The population predicted versus observed concentrations (left) and the individual predicted versus observed concentrations (right) diagnostic plots for the final PK model. The dashed line is the line of unity and the solid line is the line of the best linear fit.
Figure 3: Visual predictive check plot of piperacillin plasma concentrations (log10 scale) vs. time for the final PopPK model. Black dots represent observed data, solid lines represent quantiles of the observed data and dashed lines represent quantiles of the simulated data.

Figure 4: Bland-Altman plot for comparison of predicted versus observed piperacillin concentrations from a validation dataset. The blue line represents the mean difference in concentrations. Red lines are mean-1.96*SD (lower line) and mean+1.96*SD (upper line).
Simulations of mean (sd) $AUC_u$ values and time-concentration curves for a total daily dose of 12/1g PIP (upper graph) or 16g PIP via intermittent (left) or continuous (right) infusion for a
patient with a body weight of 70kg and a measured CL\textsubscript{CR} of respectively 20, 70, 130 and 200mL/min. AUC\textsubscript{u} values were calculated for a 24-hour interval after the sixth dose.
Captions and legends of tables and figures

Tables

Table 1: Patient characteristics, laboratory data and infection characteristics.

<table>
<thead>
<tr>
<th>Patient characteristics</th>
<th>Median (IQR) or count (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male, n (%)</td>
<td>11 (64.7%)</td>
</tr>
<tr>
<td>Age in years, median (IQR)</td>
<td>64 (51-70)</td>
</tr>
<tr>
<td>Weight in kg, median (IQR)</td>
<td>75 (69-80)</td>
</tr>
<tr>
<td>APACHE II, median (IQR)</td>
<td>20 (14-24)</td>
</tr>
<tr>
<td>SOFA, median (IQR)</td>
<td>7 (5-8)</td>
</tr>
<tr>
<td>Duration of TZP therapy in days, median (IQR)</td>
<td>5.8 (4.3-6.8)</td>
</tr>
<tr>
<td>Mechanical ventilation during TZP therapy, n (%)</td>
<td>13 (76.5%)</td>
</tr>
<tr>
<td>Vasopressive therapy during TZP therapy, n (%)</td>
<td>6 (35.3%)</td>
</tr>
<tr>
<td>ICU length of stay in days, median (IQR)</td>
<td>17.9 (14.1-31.5)</td>
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<tr>
<td>ICU survival, n (%)</td>
<td>15 (88.2%)</td>
</tr>
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</table>

**Albumin in g/L**

<table>
<thead>
<tr>
<th>Time</th>
<th>Median (IQR)</th>
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</thead>
<tbody>
<tr>
<td>72h prior to sampling</td>
<td>26.5 (22-29.5)</td>
</tr>
<tr>
<td>48h prior to sampling</td>
<td>26 (21-27.5)</td>
</tr>
<tr>
<td>24h prior to sampling</td>
<td>26.5 (22.8-30.3)</td>
</tr>
<tr>
<td>Day of sampling</td>
<td>27 (21.5-30.5)</td>
</tr>
<tr>
<td>24h post sampling</td>
<td>27 (21.5-30.8)</td>
</tr>
</tbody>
</table>

**Timing**

<table>
<thead>
<tr>
<th>Timing</th>
<th>Estimated creatinine clearance (Cockroft-Gault) in mL/min</th>
<th>Estimated creatinine clearance (MDRD) in mL/min</th>
<th>Measured creatinine clearance (mCRCL) in mL/min</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Median (IQR)</td>
<td>Median (IQR)</td>
<td>Median (IQR)</td>
</tr>
<tr>
<td>72h prior to sampling</td>
<td>82.9 (52.3-147.3)</td>
<td>97.9 (49.8-145.6)</td>
<td>70 (30-138)</td>
</tr>
<tr>
<td>48h prior to sampling</td>
<td>85.2 (41.1-139.2)</td>
<td>92.9 (36.5-140.9)</td>
<td>49.5 (16.8-141.5)</td>
</tr>
<tr>
<td>Time Point</td>
<td>Predicted Value</td>
<td>Observed Value</td>
<td>Bias</td>
</tr>
<tr>
<td>--------------------------------</td>
<td>-----------------</td>
<td>----------------</td>
<td>-------</td>
</tr>
<tr>
<td>24h prior to sampling</td>
<td>84.7 (39.9-119.3)</td>
<td>70.3 (59.8-78.6)</td>
<td>-0.078</td>
</tr>
<tr>
<td>Day of sampling</td>
<td>86.1 (40.8-139.2)</td>
<td>101.1 (35.2-140.9)</td>
<td>-0.147</td>
</tr>
<tr>
<td>24h post sampling</td>
<td>100.1 (48.3-139.2)</td>
<td>72.9 (60.6-81.5)</td>
<td>-0.457</td>
</tr>
</tbody>
</table>

Table 2: Predictive performance of linear and non-linear piperacillin population PK models

<table>
<thead>
<tr>
<th>Model</th>
<th>-2LL</th>
<th>Intercept</th>
<th>Slope</th>
<th>r²</th>
<th>Bias</th>
<th>Imprecision</th>
<th>AIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>L</td>
<td>1842</td>
<td>3.73</td>
<td>0.98</td>
<td>0.977</td>
<td>0.995</td>
<td>1852</td>
<td></td>
</tr>
<tr>
<td>L/MM</td>
<td>1748</td>
<td>5.33</td>
<td>0.96</td>
<td>0.975</td>
<td>-0.147</td>
<td>1.31</td>
<td>1797</td>
</tr>
<tr>
<td>MM</td>
<td>2197</td>
<td>38.9</td>
<td>0.933</td>
<td>0.647</td>
<td>-0.457</td>
<td>0.779</td>
<td>2207</td>
</tr>
</tbody>
</table>

Predictive performance of linear, parallel linear and Michaelis-Menten and Michaelis-Menten model. R² is the coefficient of determination for the best-fit linear regression for the predicted-observed plot. LL is the log likelihood estimate. AIC is the Akaike information criterion. L = linear, MM= Michaelis-Menten.
Table 3: Predictive performance of piperacillin population PK models incorporating renal clearance as a covariate

<table>
<thead>
<tr>
<th>Model</th>
<th>-2LL</th>
<th>Intercept</th>
<th>Slope</th>
<th>$r^2$</th>
<th>Bias</th>
<th>Imprecision</th>
<th>AIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>mCL$_{CR}$</td>
<td>1796</td>
<td>4.87</td>
<td>0.97</td>
<td>0.986</td>
<td>-0.136</td>
<td>1.25</td>
<td>1806</td>
</tr>
<tr>
<td>GaG</td>
<td>1805</td>
<td>6.08</td>
<td>0.959</td>
<td>0.97</td>
<td>-0.172</td>
<td>1.29</td>
<td>1815</td>
</tr>
<tr>
<td>MDRD</td>
<td>1904</td>
<td>5.5</td>
<td>0.98</td>
<td>0.962</td>
<td>-0.12</td>
<td>0.96</td>
<td>1915</td>
</tr>
</tbody>
</table>

Predictive performance of linear, parallel linear and Michaelis-Menten and Michaelis-Menten model. $R^2$ is the coefficient of determination for the best-fit linear regression for the predicted-observed plot. LL is the log likelihood estimate. AIC is the Akaike information criterion. mCL$_{CR}$ = measured creatinine clearance, GaG = estimated creatinine clearance using the Cockroft-Gault formula, MDRD = estimated glomerular filtration rate using the MDRD formula.
Table 4: Mean, %CV, median (IQR), and 95% CI parameter estimates for the final PopPK model

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Mean</th>
<th>% CV</th>
<th>Median (IQR)</th>
<th>95% CI around the median</th>
</tr>
</thead>
<tbody>
<tr>
<td>V (L)</td>
<td>9.74</td>
<td>87.27%</td>
<td>6.18 (5.76 – 6.52)</td>
<td>4.93 – 11.2</td>
</tr>
<tr>
<td>CL (L/h)</td>
<td>9.29</td>
<td>26.19%</td>
<td>9 (8.68 – 9.43)</td>
<td>7.69 – 11</td>
</tr>
<tr>
<td>Q (L/h)</td>
<td>21.47</td>
<td>59.81%</td>
<td>15.61 (13.38 – 20.29)</td>
<td>12.66 – 23.8</td>
</tr>
<tr>
<td>Vp (L)</td>
<td>9.8</td>
<td>34.11%</td>
<td>11.17 (10.7 – 11.69)</td>
<td>7.26 – 12</td>
</tr>
</tbody>
</table>