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Population pharmacokinetics and pharmacodynamics of piperacillin in critically ill patients during the early phaseof sepsis

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POPULATION PHARMACOKINETICS AND PHARMACODYNAMICS OF PIPERACILLIN IN CRITICALLY ILL PATIENTS DURING THE EARLY PHASEOF SEPSIS

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เภสัชจลนศาสตร์ประชากรและเภสัชพลศาสตร์ของยาพิเพอราซิลินในผู้ป่วยวิกฤตในระหว่างช่วงแรกของภาวะพิษเหตุติดเชื้อ

น.ส.วรุณรัตน์ สุกาญจนาเศรษฐ์

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาเภสัชศาสตรดุษฎีบัณฑิต
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Thesis Title	POPULATION PHARMACOKINETICS AND PHARMACODYNAMICS OF PIPERACILLIN IN CRITICALLY ILL PATIENTS DURING THE EARLY PHASEOF SEPSIS
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Field of Study	Pharmaceutical Care
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วรุณรัตน์ สุกาญจนาเศรษฐ์ : เภสัชจลนศาสตร์ประชากรและเภสัชพลศาสตร์ของยาพิเพอราซิลินในผู้ป่วยวิกฤตในระหว่างช่วงแรกของภาวะพิษเหตุติดเชื้อ. (
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ที่มาและความสำคัญ: พิเพอราซิลิน/ทาโซแบคแทม เป็นยาปฏิชีวนะที่ใช้บ่อยในการรักษาเชิงประจักษ์ในผู้ป่วยภาวะพิษเหตุติดเชื้อ การเปลี่ยนแปลงทางพยาธิสรีรวิทยาในระหว่างช่วงแรกของภาวะพิษเหตุติดเชื้อส่งผลกระทบอย่างมีนัยสำคัญต่อลักษณะทางเภสัชจลนศาสตร์และเภสัชพลศาสตร์ของยา การศึกษานี้มีวัตถุประสงค์เพื่อศึกษาลักษณะทางเภสัชจลนศาสตร์ประชากรของยาพิเพอราซิลินและหาความน่าจะเป็นของการบรรลุเป้าหมายทางเภสัชจลนศาสตร์และเภสัชพลศาสตร์และการตอบสนองสะสม เมื่อใช้แผนบริหารยาต่างๆ ในผู้ป่วยวิกฤตในระหว่างช่วงแรกของภาวะพิษเหตุติดเชื้อ

วิธีวิจัย: ผู้ป่วยภาวะพิษเหตุติดเชื้อที่ได้รับยาพิเพอราซิลิน/ทาโซแบคแทมจำนวน 48 คน ได้รับการเก็บตัวอย่างเลือดคนละ 5 ตัวอย่างที่เวลาต่างๆดังนี้ ก่อนได้รับยาและ 0-0.5, 0.5-2, 2-4 และ 4-6 หรือ 8 ชั่วโมงหลังได้รับยา จากนั้นนำตัวอย่างเลือดมาหาระดับยาพิเพอราซิลินโดยใช้โครมาโตกราฟฟีเหลวที่มีประสิทธิภาพสูง การวิเคราะห์ทางเภสัชจลนศาสตร์ประชากรใช้โปรแกรม NONMEM จากนั้นการหาความน่าจะเป็นของการบรรลุเป้าหมายที่ระยะเวลาที่ระดับยาอยู่เหนือความเข้มข้นต่ำสุดที่จะยับยั้งเชื้อได้เป็นร้อยละ 90 ของระยะห่างระหว่างการให้ยา และการตอบสนองสะสมใช้การจำลอง Monte Carlo

ผลการวิจัย: เภสัชจลนศาสตร์ประชากรของยาพิเพอราซิลินสามารถอธิบายได้ด้วยแบบจำลองเภสัชจลนศาสตร์แบบ 2 ห้อง อัตราการขจัดยา ปริมาตรการกระจายห้องกลาง และ ปริมาตรการกระจายห้องรอบนอก คือ 5.37 ลิตรต่อชั่วโมง 9.35 ลิตร และ 7.77 ลิตร ตามลำดับ อัตราการขจัดครีอะตินินและค่าความดันโลหิตเฉลี่ยเป็นปัจจัยที่มีผลต่ออัตราการขจัดยาอย่างมีนัยสำคัญ น้ำหนักตัวที่ได้จากการปรับเป็นปัจจัยที่ส่งผลต่อปริมาตรการกระจายห้องกลาง แผนการให้ยามาตรฐาน (พิเพอราซิลิน 4 กรัม ทุก 6 ชั่วโมง โดยหยดยาเข้าทางหลอดเลือดดำเป็นเวลานาน 0.5 ชั่วโมง) สามารถบรรลุเป้าหมายในผู้ป่วยที่มีอัตราการขจัดครีอะตินิน 10-40 มิลลิลิตรต่อนาทีและได้รับเชื้อก่อโรคที่ไวต่อยา (MIC 16 mg/L) แต่ไม่สามารบรรลุเป้าหมาย เมื่อใช้ในผู้ป่วยที่มีอัตราการขจัดครีอะตินิน 40-120 มิลลิลิตรต่อนาที ผู้ป่วยกลุ่มนี้ควรต้องเพิ่มระยะเวลาการหยดยา แผนบริหารยาส่วนใหญ่เมื่อใช้ในการรักษาการติดเชื้อ *Escherichia coli* ให้การตอบสนองสะสมที่มากกว่าร้อยละ 90 ในขณะที่ไม่มีแผนบริหารยาใดเลยที่ให้การตอบสนองสะสมในการรักษาการติดเชื้อ *Pseudomonas aeruginosa* มากกว่าร้อยละ 90

สรุปผล:ในระหว่างช่วงแรกของภาวะพิษเหตุติดเชื้อในผู้ป่วยที่มีการทำงานของไตเป็นปกติอาจได้ระดับยาที่ต่ำไม่เพียงพอต่อการรักษา สำหรับผู้ป่วยที่มีอัตราการขจัดครีอะตินิน 40-120 มิลลิลิตรต่อนาที ขนาดยาที่แนะนำ คือ พิเพอราซิลิน 4 กรัม ทุก 6 ชั่วโมง โดยหยดยาเข้าทางหลอดเลือดดำเป็นเวลานาน 4 ชั่วโมง

สาขาวิชา การบริบาลทางเภสัชกรรม ลายมือชื่อนิสิต .....

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KEYWORD: population pharmacokinetics, pharmacodynamics, piperacillin, early phase, sepsis

Waroonrat Sukarnjanaset : POPULATION PHARMACOKINETICS AND PHARMACODYNAMICS OF PIPERACILLIN IN CRITICALLY ILL PATIENTS DURING THE EARLY PHASEOF SEPSIS. Advisor: Asst. Prof. Thitima Wattanavijitkul, Ph.D. Co-advisor: Prof. Sutep Jaruratanasirikul

Background: Piperacillin/Tazobactam is frequently used for empirical treatment in patients with sepsis. Pathophysiological changes during the early phase of sepsis have significant effects on pharmacokinetic/pharmacodynamic (PK/PD) behaviors. This study aimed to characterize the population PKs of piperacillin and investigate probability of target attainment (PTA) and cumulative fraction of response (CFR) of various dosage regimens in critically ill patients during the early phase of sepsis.

Methods: Forty-eight patients treated with piperacillin/tazobactam were recruited. Five blood samples were drawn before and during 0-0.5, 0.5-2, 2-4 and 4-6 or 8 hours after administration. Free piperacillin concentrations were determined using HPLC. Population PKs was analyzed using NONMEM®. The PTA of 90%$fT_{>MIC}$ target and CFR were determined by Monte Carlo simulation.

Results: The two compartment model best described the data. Piperacillin clearance (CL), central volume of distribution (V_1) and peripheral volume of distribution were 5.37 L/h, 9.35 L, and 7.77 L, respectively. Creatinine clearance (CL_{Cr}) and mean arterial pressure had a significant effect on CL while adjusted body weight had a significant impact on V_1. The standard regimen, 4-g of piperacillin infused over 0.5 hours every 6 hours, achieved the target for susceptible organisms with MIC ≤16 mg/L in patients with CL_{Cr} 10 to 40 ml/min, but not with CL_{Cr} 40-120 ml/min. In such patients, prolonged infusion is required. Most regimens provided CFR 90% for the *E. coli* infection while there was no dosage regimen achieved a CFR of 90% for the *P. aeruginosa* infection.

Conclusions: Due to high CL and V_1, subtherapeutic concentrations can occur during the early phase of sepsis in critically ill patients with normal renal function. Our proposed regimen for the patients with CL_{Cr} 40-120 ml/min was an extended 4-hour infusion of 4-g of piperacillin every 6 hours.

Field of Study:	Pharmaceutical Care	Student's Signature .....
Academic Year:	2018	Advisor's Signature .....
		Co-advisor's Signature .....

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Waroonrat Sukarnjanaset

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CHAPTER I INTRODUCTION

1.1 Background and rationale

Piperacillin/tazobactam is an extended-spectrum hydrophilic antibiotic used for empirical treatment in critically ill patients with sepsis.(1) In healthy volunteers, piperacillin is distributed into body fluids and tissues.(2) Volume of distribution (V_d) ranged between 11.9 and 18.6 L.(3-6) Protein binding is in the range of 20-30%.(3, 4) Piperacillin is primarily eliminated through renal clearance.(3, 4) The percentage of dose recovered in urine in its unchanged form ranged between 57 and 80%.(2-4, 7) The remainder is metabolized and then secreted into the bile.(3, 4) Total clearance (CL) ranged between 11.9 and 15.8 L/h.(3-6) The elimination half-life ranged from 0.83 to 1.22 hour.(3-6) Piperacillin is a time-dependent antibiotic and the percentage of time which free drug concentrations remain above minimum inhibitory concentration (MIC) during a dosing interval (%$fT_{>MIC}$) has been considered to be the best efficacy predictor.(8) Typically, 50% $fT_{>MIC}$ has been required for optimal activity of penicillins.(9) Recently, 75% $fT_{>MIC}$ has been proposed for bactericidal activity of piperacillin(10), however, higher targets (90 to 100% $fT_{>MIC}$) have been recommended for microbiological success and prevention of bacterial regrowth in patients with serious bacterial infections.(11-13)

Sepsis is defined as life-threatening organ dysfunction caused by a dysregulated host response to infection.(14) Sepsis remains a major cause of mortality and critical illness.(14-16) The mortality rate in this group of patients ranged between 14 and 45%.(15) Early antibiotic administration in patients with sepsis could reduce the mortality rate (17) but dosing strategies which enhance 100% $fT_{>MIC}$ attainment of piperacillin concentrations in these patients especially for less susceptible pathogens remains a perplexing issue. Pathophysiological changes, particularly during the early phase, have several significant effects on pharmacokinetic (PK) behaviors. Sepsis can cause endothelial damage and capillary leakage.(18-20) From previous works in patients with sepsis, V_d of piperacillin has been found to be larger than those in healthy volunteers and non-critically ill patients.(21-26) In addition, patients with sepsis might develop renal dysfunction which led to a decrease in piperacillin CL.(21, 23, 25, 27, 28)

On the other hand, hemodynamic changes including a high cardiac output(29) could lead to an increase in hepatic and renal blood flow.(30) Piperacillin CL could be higher in septic patients with normal renal function compared with in healthy volunteers.(24, 26) Because of these PK changes, subtherapeutic piperacillin concentrations have been found in patients with the first 24 hours of sepsis.(23, 27) Thus proper piperacillin dosing in patients with the early phase of sepsis persists the important and challenging issue.

The aims of this study were to estimate population PK parameters and variabilities, investigate the probability of target attainment (PTA) of various piperacillin dosage regimens and explore the cumulative fraction of response (CFR) against pathogens commonly found in critically ill patients with the early phase of sepsis.

1.2. Objectives

1.2.1. Primary objectives

- To estimate population PK parameters and variabilities of piperacillin in critically ill patients during the early phase of sepsis.
- To investigate the PTA of various piperacillin dosage regimens and the CFR against pathogens commonly found in critically ill patients during the early phase of sepsis.

1.2.2. Secondary objectives

- To assess clinical responses of critically ill patients with sepsis when receiving piperacillin/tazobactam.
- To explore 28-day mortality rate and the association with individual $\%fT_{>MIC}$ in critically ill patients with sepsis who were treated with piperacillin/tazobactam.

CHAPTER II LITERATURE REVIEW

2.1. Piperacillin

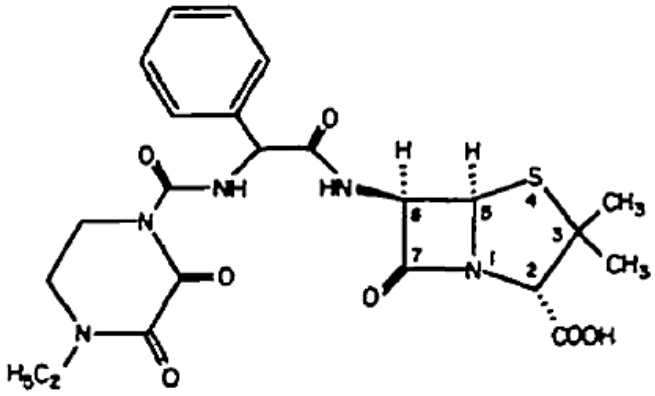

Figure 1 Chemical structure of piperacillin

2.1.1. Physicochemical properties

The chemical structure of piperacillin is shown in figure 1. Piperacillin is highly water soluble (714 g/L) with pK_a value of 4.14 (weak acid). Piperacillin will be present as an anionic substance at physiological pH values. (4)

2.1.2. Pharmacokinetic (PK) and pharmacodynamics (PD) characteristics

In healthy volunteers, piperacillin is distributed into body fluids and tissues including skin, muscle, lung, gall bladder, intestinal mucosa, female reproductive tissues, bile and interstitial fluid.(2) Volume of distribution (V_d) ranged between 11.9 and 18.6 L as shown in table 1.(3-6) Protein binding is in the range of 20-30%.(3, 4) Piperacillin is primarily eliminated through renal clearance.(3, 4) The percentage of dose recovered in urine in its unchanged form ranged between 57 and 80%.(2-4, 7) The remainder is metabolized and then secreted into the bile.(3, 4) Total clearance (CL) ranged between 11.9 and 15.8 L/h and the elimination half-life ($T_{1/2}$) ranged from 0.83 to 1.22 hour as shown in table 1.(3-6)

Piperacillin is always administered with tazobactam. Tazobactam inhibits beta-lactamases which destroy beta-lactam structures of piperacillin to broaden antimicrobial activity but it has no bactericidal activity by itself. However, when they are given together in a ratio of 8 : 1 (Piperacillin 4 g : Tazobactam 0.5 g), the PKs of piperacillin remain unaffected by tazobactam. Cheung et al investigated in

6 healthy volunteers receiving 3 treatments on 3 different occasions (1 week washout period); (i) 4-g of piperacillin, (ii) 0.5-g of tazobactam, and (iii) the combination of 4-g of piperacillin and 0.5-g of tazobactam, they found that mean PK parameters of piperacillin when administered with tazobactam were not significantly different from those when administered alone.(5)

Table 1 Pharmacokinetic characteristics of piperacillin in healthy volunteers

No	Authors, Year	Dose (Single dose)	V_d (L)	CL (L/h)	$T_{1/2}$ (h)
1	Tjandramaga et al, 1978(3)	Piperacillin 4g 3-min infusion	18.6 ± 1.1	15.3 ± 1.2	1.02 ± 0.05
2	Sorgel et al, 1993(4)	PIP/TAZ 4/0.5 g 5-min infusion	14.5 ± 2.5	12.3 ± 1.1	0.83 ± 0.23
		PIP/TAZ 4/0.5 g 30-min infusion	13.7 ± 0.9	15.8 ± 1.2	1.14 ± 0.12
3	Cheung et al, 1998(5)	Piperacillin 4 g 30-min infusion	11.9 ± 1.4	14.4 ± 2.4	0.87 ± 0.08
		PIP/TAZ 4/0.5 g 30-min infusion	12.3 ± 0.9	14.5 ± 1.2	0.83 ± 0.11
4	Bulitta et al, 2010(6)	Piperacillin 4g 5-min infusion	12.7 ± 2.3	11.9 ± 1.3	1.22 ± 0.46

PIP/TAZ, Piperacillin/Tazobactam

Piperacillin is a time-dependent antibiotic and the percentage of time which free drug concentrations remain above minimum inhibitory concentration (MIC) during a dosing interval (%$fT_{>MIC}$) has been considered to be the best efficacy predictor.(8) Typically, 50% $fT_{>MIC}$ has been required for optimal activity of penicillins.(9) Craig explored the relationship between duration of time which beta-lactams above the MIC and survival after day 4 of therapy in animal model infected with *S. pneumonia*. The results showed that when 50%$fT_{>MIC}$ was achieved, the survival would be 100%.(31) Recently, 75% $fT_{>MIC}$ has been proposed for bactericidal activity of piperacillin.(10) Zelenitsky performed *in vitro* study by simulating *P. aeruginosa* bacteremia in an immunocompromised host, they found that 75% $fT_{>MIC}$ provided bactericidal activity.(10) However, higher PK/PD targets (90 to 100% $fT_{>MIC}$) have been recommended for microbiological success and prevention of bacterial regrowth in patients with serious bacterial infections.(13)

2.1.3. Clinical uses

Piperacillin/tazobactam (PIP/TAZ) is an extended-spectrum beta-lactam antibiotic used for empirical treatment in critically ill patients with sepsis.(1) Overall, piperacillin antibacterial activity encompasses gram-positive, gram-negative aerobic and anaerobic bacteria, including many pathogens producing beta-lactamases such as *Escherichia coli, Klebsiella pneumoniae, and Pseudomonas aeruginosa*.(32)

Recommended dosage regimens of PIP/TAZ are 4/0.5 g every 6 hours, 2/0.25 g every 6 hours, 2/0.25 g every 8 hours for septic patients with creatinine clearance (CL_{Cr}) more than 40 mL/min, 20-40 mL/min, and less than 20 mL/min, respectively.(33) PIP/TAZ is usually administered by short infusion (over 20 to 30 min). Some clinicians prefer extended infusion; give the first dose over 30 min as a loading dose, followed by 4 hours infusion.(33) Extended infusion including 3-hour and 4-hour infusion, and continuous infusion of PIP/TAZ are increasingly proposed to help enhance the probability of target attainment (PTA) in patients with severe infections.(24) Previous studies in critically ill patients have documented that prolonged infusion strategy of PIP/TAZ showed significantly higher clinical cure rate (34, 35) and lower mortality rate in comparison with

conventional intermittent strategy.(35-37) However, a randomized control trial reported that there was no significant difference in clinical cure and 90 days-mortality rate between continuous and intermittent infusion group.(38) Likewise, Cutro et al. documented that clinical failure and inpatient mortality rate were similar between extended and short infusion.(39) Fan et al. also stated that both extended and short infusion demonstrated similar 14-day mortality.(40)

2.1.4. Adverse drug reaction

Common adverse drug reactions related to PIP/TAZ treatment include urticaria, thrombocytopenia, bone marrow suppression, and neurotoxicity.(41, 42) Regarding thrombocytopenia, the mechanism is unclear, however, drug-induced thrombocytopenia have three possible mechanisms; (i) immune-mediated thrombocytopenia, (ii) direct platelet number decreases, and (iii) bone marrow suppression.(43) Chen et al reported that thrombocytopenia occurred 12 hours after receiving 4/0.5-g PIP/TAZ every 8 hours. Platelets return to normal level within 3-5 days after discontinuation.(44) In addition, bone marrow suppression (thrombocytopenia, leukocytopenia, and anemia) could be found in patients receiving long-term PIP/TAZ (longer than 2 weeks). There are case reports to document PIP/TAZ induced bone marrow suppression.(45, 46) Ruiz-Irastorza et al stated that pancytopenia was observed in a patient receiving 4/0.5-g PIP/TAZ every 8 hours on day 17, however such condition was rapidly reversible after antibiotic cessation.(45) Likewise, Zhong-Fang et al also found that patients receiving 4/0.5-g PIP/TAZ every 8 hours had pancytopenia on day 17, and then it could be resolved after antibiotic cessation.(46)

2.2. Influence of pathophysiological changes in critically ill patients with sepsis on PK and PD properties of piperacillin

Sepsis is defined as life-threatening organ dysfunction caused by a dysregulated host response to infection.(14) Pathophysiological changes in patients with sepsis have several significant effects on the PK behaviors.

2.2.1. Change in piperacillin V_d

There have been several plausible explanations to describe the changes in V_d. Initially, the toxin release from pathogens causes the persistent production of various endogenous mediators that lead to endothelial damage and subsequently increased capillary permeability.(18-20) In addition, fluid retention can be found in patients with cardiac or renal failure.(47, 48) The administration of resuscitation fluids can also result in an expansion of fluid volume in the interstitial space.(13, 47, 48) Moreover, mechanical ventilation used to treat respiratory failure can cause an increase in airway and intrathoracic pressure leading to a decrease in cardiac filling pressure and cardiac output, subsequently.(49, 50) The lower cardiac output can stimulate the renin-angiotensin system which can cause an increase in antidiuretic hormone leading to water retention ultimately.(51) With these reasons, the V_d of hydrophilic drugs is likely to enlarge in patients with sepsis.

From most previous works, V_d of piperacillin in patients with sepsis has been found to be larger than those in healthy volunteers as shown in table 2. Joukhadar et al. compared pharmacokinetics of 6 septic shock patients with 6 healthy volunteers, both groups received a single dose of 4/0.5-g of PIP/TAZ (10-min infusion). They found that V_d of PIP in septic shock patients (40.7 L, 0.57 L/kg) was approximately 6 times larger than healthy volunteers (9.6 L, 0.13 L/kg).(21) Taccone et al. studied pharmacokinetics of 4 beta-lactams (PIP/TAZ, cefepime, ceftazidime, and meropenem) in 27 patients with severe sepsis and septic shock who received 4/0.5-g of PIP/TAZ (30-min infusion) every 6 or 8 hours, they reported piperacillin V_d of 0.38 L/kg smaller than meropenem (0.43 L/kg) and ceftazidime (0.48 L/kg).(23) Roberts et al. studied in 16 patients during the early phase of sepsis and normal renal function (8 patients receiving intermittent bolus and 8 patients receiving continuous infusion) on day 1 and day

2 of therapy. They found that the calculated piperacillin V_d (25.0 L, 0.33 L/kg).(24) In addition, Udy et al. investigated pharmacokinetics of piperacillin in 48 patients with sepsis and normal renal function who received 4/0.5-g of PIP/TAZ (20-min infusion) every 6 hours, they reported the calculated piperacillin V_d of 38.7 L (0.44 L/kg).(26) Recently, Andersen et al. conducted in 22 patients with sepsis receiving 4/0.5-g of PIP/TAZ (3-min infusion) every 8 hours, they reported the calculated piperacillin V_d of 15.9 L (0.21 L/kg).(52) On the other hand, Obrink-Hansen et al. which studied in 15 patients during the early phase of septic shock who received 4/0.5-g of PIP/TAZ (3-min infusion) every 8 hours found that the calculated piperacillin V_d was relatively small (11.2 L, 0.14 L/kg).(27)

2.2.2. Change in piperacillin CL

Regarding drug elimination, there have been various probable reasons to explain the changes in CL of patients with sepsis. Initially, the hemodynamic changes including high cardiac output have been observed. Increased cardiac output is associated with increased hepatic and/or renal blood flow, leading to elevated CL, subsequently. On the contrary, some patients with sepsis could develop hepatic and/or renal dysfunction which causes a decrease in CL.(13, 47, 48, 53, 54) In addition, the use of vasoactive medications in patients with sepsis has been required to improve their cardiovascular function.(55) Norepinephrine, a commonly employed vasoactive medication, can increase cardiac output, hepatic and renal blood flow.(56) Several studies in patients with septic shock have revealed that norepinephrine increased CL_{Cr}.(57-59) Likewise, a study in critically ill patients has shown that dopamine administration significantly increased CL_{Cr}.(60) Mechanical ventilation can result in lower cardiac output as previously described, therefore it can cause a decrease in renal(51) and hepatic blood flow(61), leading to a fall in CL.

With these reasons, piperacillin CL could be higher in septic patients with normal renal function compared with in healthy volunteers as shown in table 2.(24, 26) Robert et al. studied in patients with median CL_{Cr} 88 and 97 mL/min for intermittent bolus and continuous infusion groups, respectively. They found that the piperacillin CL of total population was 17.1 L/h.(24) Likewise, Udy et al.

investigated in patients with mean CL_{Cr} 122 mL/min, they found that the piperacillin CL was 16.3 L/h.(26) On the other hand, some PK studies found that piperacillin CL in septic patients could be lower in septic patients compared with in healthy volunteers as shown in table 2.(21, 23, 27, 28, 52) When considered by the renal function, Andersen et al. which investigated in patients with sepsis (excluding severe sepsis and septic shock) and median CL_{Cr} 83.9 mL/min found that piperacillin CL was 8.6 L/h.(52) Taccone et al which studied in patients with severe sepsis and septic shock reported piperacillin CL of 8.4 mL/min.(23) Likewise, Tsai et al which studied in Australian indigenous patients with severe sepsis stated that piperacillin CL was 5.6 L/h.(28) Joukhadar et al. and Obrink-Hansen et al. which conducted in only septic shock patients with plasma creatinine 1.8 and 1.9 mg/dL, respectively. They found that piperacillin CL were 8.2 and 3.6 L/h, respectively.(21, 27)

Table 2 Volume of distribution (V_d) and total clearance (CL) of piperacillin in critically ill patients with sepsis

No	Authors, Year	Patients (N)	Dosage regimens	V_d (L)	CL (L/h)
1	Joukhadar et al, 2001(21)	Septic shock (6)	A single dose of PIP/TAZ 4/0.5 g (10-min infusion)	40.7 ± 8.7 (0.57 L/kg)	8.2 ± 2.0
2	Taccone et al, 2010(23)	Severe sepsis and septic shock (27)	PIP/TAZ 4/0.5 g (30-min infusion) every 6 or 8 hours	0.38 L/kg	8.4 ± NA
3	Robert et al, 2010(24)	Sepsis with normal renal function (16) (During day 1 and day 2 of sepsis)	(i) PIP/TAZ 4/0.5 g (20-min infusion) every 6 or 8 hours (N=8) (ii) PIP/TAZ 4/0.5 g (20-min infusion) followed by PIP/TAZ 8/1 g continuous infusion (N=8)	25.0 ± NA (0.33 L/kg)	17.1 ± NA
4	Udy et al, 2015(26)	Sepsis with normal renal function (48)	PIP/TAZ 4/0.5 g (20-min infusion) every 6 hours	38.7 ± NA (0.44 L/kg)	16.3 ± NA
5	Obrink-Hansen et al, 2015 (27)	Septic shock (15) (During the third administration)	PIP/TAZ 4/0.5 g (3-min infusion) every 8 hours	11.2 ± NA (0.14 L/kg)	3.6 ± NA
6	Tsai et al, 2016(28)	Severe sepsis (9)	PIP/TAZ 4/0.5 g (30-min infusion) every 8 hours	14.5 ± 6.6 (0.19 L/kg) (V_1)	5.6 ± 3.2
7	Andersen et al, 2018 (52)	Sepsis (22)	PIP/TAZ 4/0.5 g (3-min infusion) every 8 hours	15.9 ± NA (0.21 L/kg)	8.6 ± NA

PIP/TAZ, Piperacillin/Tazobactam; NA, Not available; V_1, Central volume of distribution

2.2.3. Change in pharmacodynamics

Pathophysiological alteration could also have significant effects on the achievement of a PK/PD target.(62) Regarding the 50%$fT_{>MIC}$ target at MIC 16 mg/L, the probability of target attainment (PTA) of PIP/TAZ 4/0.5 g every 6 hour and 8 hour (usual dosage regimens) in patients with sepsis and normal renal function was 33% and 25%, respectively, which are below the accepted PTA of 90%(24), while the PTA was 100% in morbidly obese patients with sepsis whose renal function decreased.(25) Recently, Zelenitsky et al. found a pivotal PD relationship between bacterial kill and % $fT_{>MIC}$ with significant threshold of 75% $fT_{>MIC}$ for bactericidal activity of piperacillin.(10) A higher PK/PD target (90 to 100% $fT_{>MIC}$) was proposed as a proper PK/PD target to provide microbiological success and prevent bacterial regrowth.(13) Taccone et al. found that the PTA (50%$fT_{>4MIC}$: MIC 16 mg/L) of PIP/TAZ 4/0.5 g every 6 or 8 hour was only 44% in patients with the early phase of severe sepsis. Moreover, they also found that patients with CL_{Cr} less than 50 mL/min had a significantly higher PTA than patients with CL_{Cr} more than 50 mL/min.(23) In addition, Udy et al. found that the PTA (100%$fT_{>MIC}$: MIC 16 mg/L) of PIP/TAZ 4/0.5 g every 6 hour in patients with sepsis and normal renal function was 34%.(26) Obrink-Hansen et al and Andersen et al similarly reported that PIP/TAZ 4/0.5 g every 6 and 8 hour provided the PTA less than 90% for both targets (50%$fT_{>4MIC,}$ 100%$fT_{>MIC}$: MIC 16 mg/L)(27, 52), as shown in table 3.

Table 3 The PTA of piperacillin in patients with sepsis at MIC 16 mg/L

No	Authors, Year	Patients (N)	Dosage regimens	PK/PD targets	Cl_{Cr} (mL/min)	PTA (%)
1	Taccone et al, 2010(23)	Severe sepsis and Septic shock (27)	PIP/TAZ 4/0.5 g every 6 or 8 hours	50% $fT_{>4MIC}$	56	44
2	Robert et al, 2010(24)	Sepsis with normal renal function (16)	PIP/TAZ 4/0.5 g every 6 hours	50% $fT_{>MIC}$	92	33
			PIP/TAZ 4/0.5 g every 8 hours			25
3	Sturm et al, 2014(25)	Sepsis with Morbidly obese (9) [BMI ≥ 40 kg/m^2]	PIP/TAZ 4/0.5 g every 6 hours	50% $fT_{>MIC}$	75	100
			PIP/TAZ 4/0.5 g every 8 hours			100
4	Udy et al, 2015(26)	Sepsis with normal renal function (48)	PIP/TAZ 4/0.5 g every 6 hours	100% $fT_{>MIC}$	122	34
5	Obrink-Hansen et al, 2015 (27)	Septic shock (15)	PIP/TAZ 4/0.5 g every 6 hours	50% $fT_{>4MIC}$	S_{Cr} 170 µmol/L	< 90%
				100% $fT_{>MIC}$		< 90%
			PIP/TAZ 4/0.5 g every 8 hours	50% $fT_{>4MIC}$		< 90%
				100% $fT_{>MIC}$		< 90%
6	Andersen et al, 2018 (52)	Sepsis (22)	PIP/TAZ 4/0.5 g every 6 hours	50% $fT_{>4MIC}$	84	< 90%
				100% $fT_{>MIC}$		< 90%
			PIP/TAZ 4/0.5 g every 8 hours	50% $fT_{>4MIC}$		< 90%
				100% $fT_{>MIC}$		< 90%

PIP/TAZ, Piperacillin/Tazobactam; Cl_{Cr}, Creatinine clearance; S_{Cr}, Serum creatinine

2.3. Population pharmacokinetics using nonlinear mixed-effect model approach

2.3.1. Background

Population pharmacokinetics is the study of the sources and correlations of variability in drug concentrations among the target population receiving clinically relevant doses of a drug of interest. Population pharmacokinetics try to identify the measurable pathophysiologic features that cause changes in the dose-concentration relationship and the magnitude of these changes therefore if such changes are associated with clinically significant shifts in the therapeutic target, dosage can be properly adjusted.(63)

Nonlinear mixed-effect model approach is the current standard method used for population pharmacokinetics. This approach characterizes pharmacokinetics taking into account different types of variability such as between-subject and within-subject variability. One purpose of a nonlinear mixed-effect model is to model the relationship between an independent (time) and a dependent (concentration) variable. Another goal is to obtain estimates of the model parameters and their associated variance components. The nonlinear mixed-effect modeling has many advantages including (i) the data do not need to be intensive; they could be sparse with as little as one observation per patient or rich with many observations per patient, or a combination of both, (ii) the data do not have to follow any specific sampling time schedule and may have irregular sampling times, (iii) important covariates which explain between-patient variability can be identified. However, the limitation should be noted, sciences used in this approach are complicated and difficult to understand and implement therefore the modeling would be the time consuming process.(64)

2.3.2. Model development

Nonlinear mixed effect models consist of 2 components; the structural model and the variance model. The structural model is the model which best describes the data in the absence of covariates, such as one- and two- compartment models. Regarding the variance model, there are usually two main sources of variability; interindividual variability (IIV) and residual variability (RV). IIV refers to the variance of a parameter across different individuals in the population;

it could be described using the additive, proportional, or exponential model, as follows:

Additive IIV model

$$P_i = TVP + \eta_i$$

Proportional IIV model

$$P_i = TVP\ (1 + \eta_i)$$

Exponential IIV model

$$P_i = TVP\ e^{\eta i}$$

where P_i is the value of the PK parameter for the i^{th} subject.

TVP is the typical value of the population PK parameter.

η_i is the value of the deviation from the typical value for the i^{th} subject, it is assumed to have normal distribution with zero mean and ω^2 variance.

RV refers to the unexplained variability in the observed data after controlling for other sources of variability; it could be explained using the additive, proportional, combination between additive and proportional model or exponential model, as follows:

Additive RV model

$$C_{O,\,ij} = C_{P,\,ij} + \varepsilon_{ij}$$

Proportional RV model

$$C_{O,\,ij} = C_{P,\,ij}(1 + \varepsilon_{ij})$$

Combined additive and proportional RV model

$$C_{O,\,ij} = C_{P,\,ij} + \varepsilon_{1,\,ij} + (1 + \varepsilon_{2,\,ij})$$

Exponential RV model

$$C_{O,\,ij} = C_{P,\,ij}\ e^{\varepsilon ij}$$

where $C_{O,\,ij}$ is the observed concentration of the i^{th} subject at time j^{th}.

$C_{P,\,ij}$ is the predicted concentration of the i^{th} subject at time j^{th}.

ε_{ij} is the value of the difference between the observed and predicted concentrations of the i^{th} subject at time j^{th}, it is assumed to have normal distribution; zero mean and σ^2 variance.

The base model which consists of the structural, IIV, and RV models was chosen by considering the objective function value (OFV), Akaike information criterion (AIC), and graphical examination. Next step, covariate model building is performed, covariates selected should have some physiological rationale for their inclusion in the base model. The relation between a chosen covariate and a parameter is graphically explored. Different patterns of relationship are investigated, as follows:

- Continuous covariates

  Linear covariate model

  $$TVP = \theta_1 + \theta_2 \times (COV - COV_{Median})$$

  Power covariate model

  $$TVP = \theta_1 (COV/COV_{Median})^{\theta 2}$$

  Exponential covariate model

  $$TVP = \theta_1 e^{(\theta 2 \times (COV/COV_{Median}))}$$

  where TVP is the typical value of the population PK parameter.

  COV is the value of the continuous covariate.

  COV_{Median} is the median of the continuous covariate.

  θ_1 is the PK parameter value when the individual covariate is median (for linear and power model), zero (for exponential model)

  θ_2 is the value of the change in PK parameter for unit change in covariate for linear model, the value of the change in Ln(PK parameter) per unit change in Ln(covariate) for power model, and the value of the change in Ln(PK parameter) per unit change in covariate for exponential model.

- Categorical covariates

Fractional change covariate model

$$TVP = \theta_1 \times (1+(\theta_2 \times Cov))$$

where TVP is the typical value of the population PK parameter. COV is the value of the continuous covariate.

θ_1 is the typical PK parameter value without covariate

θ_2 is the value of the change in PK parameter when the covariate presents

Then the stepwise forward addition and backward deletion are implemented to obtain the final model. Based on the χ^2 test, a decrease in the OFV of 3.84 units is considered to be significant ($P < 0.05$) for forward addition step and an increase in the OFV of 6.64 units is considered to be significant ($P < 0.01$) for backward deletion step to avoid any possible false positives.(65)

2.3.3. Model evaluation

2.3.3.1. Goodness of fit plots

Goodness of fit plots are graphically assessed for accuracy of a model. Typical goodness of fit plots are shown in table 4. If the model could well describe the data, predicted concentrations should correspond to observed concentrations (data points are scattered around the identity line) and weighted residual errors (the difference between observed and predicted concentration) should close to zero (data points are scattered around the horizontal zero line)(66)

Table 4 Goodness of fit plots and interpretation

Plots	**Expectation if the model is correct**	**Possible solutions if the model does not fulfill the requirement**
1. Observed vs population predicted concentrations	• Data points are scattered around the identity line	• A modification of structural model, IIV model, or RV model
2. Observed vs individual predicted concentrations	• Data points are scattered evenly around the identity line • Points cluster closer to the line than observed vs population predicted concentrations (especially when IIV is large)	• A modification of structural model or RV model
3. Individual weighted residuals vs individual predicted concentrations	• Data points are scattered evenly around the horizontal zero-line • Most of points lie within -1.96 to 1.96	• A modification of structural model or RV model • A cone-shaped graph suggests a change in the RV model
4. Individual weighted residuals vs time	• Data points are scattered evenly around the horizontal zero-line • Most of points lie within -1.96 to 1.96	• A modification of structural model or RV model

2.3.3.2. Bootstrap

Bootstrap is implemented to assess reliability of the model. The primary data set was sampled with replacement to obtain various secondary data set, and then the model is fitted with all secondary data set. After that PK parameter estimates are considered whether they are within the range of 95% confidence interval of bootstrap parameter estimates or not. If the model is reliable, PK parameter estimates should be in the range of 95% confidence interval of bootstrap parameter estimates.(67)

2.3.3.3. Visual predictive check

Visual predictive check is used to assess predictive performance. The parameter estimates of the model are fixed and used to simulate a number of virtual data set, and then observed data is compared to the simulated data. If the model has adequate predictive performance, the percentiles of observed data should be within the 95% CI of corresponding percentiles of predicted data.(66)

2.4. Plausible covariates causing change in PK and PD properties of piperacillin in critically ill patients with sepsis

2.4.1. Creatinine clearance

Piperacillin is primarily eliminated by renal clearance(3, 7) therefore CL_{Cr} became an obviously reasonable covariate which should be explored. Previous PK studies in critically ill patients with sepsis found that the significant covariate of piperacillin CL was CL_{Cr}.(23, 26, 28, 52) Taccone et al studied in 27 patients with median CL_{Cr} 56 mL/min (calculated by Cockcroft and Gault equation). They found that there was a significant correlation between CL_{Cr} and piperacillin CL. Seventy one percent of patients with CL_{Cr} < 50 mL/min (n=14) could attain the target (50%$T_{>4xMIC}$, MIC 8 mg/L) while only 15% of patients with $CL_{Cr} \geq 50$ mL/min (n=13) could achieve this target.(23) Udy et al studied in 48 patients with mean measured CL_{Cr} 122 mL/min (46% of patients had CL_{Cr} > 130 mL/min (augmented renal clearance (ARC)). They also found that a correlation was observed between CL_{Cr} and piperacillin CL. In addition, CL_{Cr} was found to be the only one covariate significantly improved the fit of the PK model (change in the objective function value (ΔOFV) of -18.6).(26) Tsai et al studied in 10 indigenous

patients with severe sepsis (mean measured CL_{Cr} 91 mL/min). They found that CL_{Cr} was a significant covariate of piperacillin clearance.(28) Andersen et al studied in 22 patients with sepsis (excluding severe sepsis and septic shock) and median CL_{Cr} (calculated by Cockcroft and Gault equation) 83.9 mL/min. They found that inclusion of CL_{Cr} on piperacillin CL improved the PK model fit (ΔOFV of -32.3).(52) In another viewpoint, Obrink-Hansen et al studied in 15 septic shock patients with median plasma creatinine of 1.9 mg/dL. They found that plasma creatinine was found to be the most significant covariate to piperacillin CL by dropping interindividual variability (IIV) of piperacillin CL from 114.3% to 70.6%.(27)

2.4.2. Body weight

Body weight has been considered to be an important covariate for PKs and PDs of antibiotics. Regarding obese patients, obesity causes several physiological alterations, leading to changes in the V_d. When considering appropriate dosage of hydrophilic drugs, the water content in adipose tissue is approximately 30%(68), thus use of total body weight (TBW) in dosing a hydrophilic drug may result in a significant overdosage. Therefore dose increment in obese patients should use adjusted body weight (ABW), dosing weight is in proportion to the excess in body weight with use of a dosing weight correction factor (DWCF) as follows(69);

$$ABW = IBW + [DWCF \times (TBW-IBW)]$$

where ABW is adjusted body weight (kg)

IBW is ideal body weight (kg)

DWCF is dosing weight correction factor

TBW is total body weight (kg)

DWCF of hydrophilic drugs ranged between 0.38 and 0.58.(70-74) As for beta-lactams, the suggested DWCF is 0.30 however there is no clinical studies confirm this value. From previous PK studies of piperacillin in obese patients, Sturm et al which studied in 9 critically ill, morbidly obese surgical patients found that piperacillin V_d did not relate to any body weight (TBW, ABW, idael body weight (IBW), lean body weight (LBW)), probably resulting from the small sample size.(25) Alobaid et al investigated PKs of piperacillin in

3 groups of critically ill patients; (i) normal weight (n=13), (ii) obese (n=12), and (iii) morbidly obese (n=12), they found that body mass index (BMI) was the significant covariate for piperacillin V_1.(75) In patients with sepsis, TBW was found to be the significant covariate of piperacillin CL(24) and V_1 (28). The study of Robert et al reported that TBW (mean TBW 76 kg, BMI 26 kg/m^2) was the important covariate for piperacillin CL by reducing reduced IIV of 6.2%.(24) Likewise, the study of Tsai et al found that TBW (mean TBW 76 kg, BMI 27 kg/m^2) also was the significant covariate for V_1.(28) However, it should be noted that ABW was not investigated in both PK studies.

2.4.3. Mean arterial pressure

MAP is the measure of the strength of the blood pushing against the blood vessels as the heart pumps blood throughout the body. It is generally used in intensive care unit (ICU). MAP helps determine the actual pressure which carries oxygenated blood from the heart throughout the body.(76) A MAP ≥ 65 mmHg is a goal of therapy to maintain perfusion pressure and adequate flow at the arteriolar level.(77)

MAP is an average blood pressure during a single cardiac cycle. It is derived to represent the proportion of time in systole and diastole. It is calculated using the following equation(77);

$$MAP = DBP + (1/3 \times (SBP - DBP))$$

where MAP is mean arterial pressure (mmHg)

DBP is diastolic blood pressure (mmHg)

SBP is systolic blood pressure (mmHg)

Because MAP is associated with blood perfusion to vital organs such as kidney and liver, this may cause changing in PK behaviors. However, none of PK study in critically ill patients with sepsis have been documented that MAP had an effect on PK parameters of piperacillin.

2.4.4. Total bilirubin

Hyperdynamic state is a condition noticed in patients with the early phase of sepsis. In this condition, total bilirubin could be increased.(78) A PK study reported that total bilirubin was highly associated with piperacillin CL. Shikuma et al studied in 9 severely burn patients who had normal renal and hepatic

function. They found that total bilirubin had the non-linear relationship with piperacillin CL. This finding may result from stress induced hyperdynamic state reflected by changes in total bilirubin. The hyperdynamic state could accelerate physiologic and metabolic functions and likely lead to rapid piperacillin clearance.(78)

2.4.5. Resuscitation fluids

In critically ill patients with sepsis, capillary leakage condition and administration of resuscitation fluids enhances an expansion of fluid volume in the interstitial space and enlarge V_d for hydrophilic antibiotics, subsequently. (48, 79-82) Udy et al investigated 24-hour fluid balance but they did not mention any effect on V_d. Likewise, there is no previous PK studies which have reported a significant relation between amount of resuscitation fluids and piperacillin V_d.

2.4.6. Vasoactive medications

The use of vasoactive medications has been required to improve cardiovascular function in critically ill patients with septic shock.(55) Norepinephrine, a commonly employed vasoactive medication, can increase cardiac output, hepatic and renal blood flow.(56) Several studies in patients with septic shock have revealed that norepinephrine increased CL_{Cr}.(57-59) Likewise, a study in critically ill patients has shown that dopamine administration significantly increased CL_{Cr} but dobutamine did not have any effect on CL_{Cr}.(60) On the other hand, a study in animal has reported that epinephrine was associated with a significant decrease in renal blood flow.(83)

2.4.7. Mechanical ventilation

Mechanical ventilation can cause an increase in airway and intrathoracic pressure leading to a decrease in cardiac filling pressure and cardiac output, subsequently.(49, 50) The lower cardiac output can stimulate the renin-angiotensin system which can cause an increase in antidiuretic hormone leading to water retention.(51) There is a population PK study conducted by Georges et al. This study revealed that mechanical ventilation was a significant covariate for V_1 of ceftazidime.(84) In addition, mechanical ventilation can result in lower cardiac output as previously described, therefore it can cause a decrease in renal(51) and hepatic blood flow(61) leading to a fall in CL, subsequently.

2.5. Clinical outcomes of piperacillin/tazobactam in patients with sepsis

In patients with sepsis, clinical response and mortality rate have been primarily used to assess clinical outcomes of antibiotic treatment.(34, 35, 38-40) There have been several different measures to assess clinical responses. Clinical responses (improvement or failure) are usually evaluated from signs and symptoms of infection. In addition, microbiological test and changes in antibiotics (broadening or de-escalating measures) are also used to assess clinical responses. Cutro et al. assessed the clinical failure after 48 hours of administration from 3 criteria; (i) persistent/worsening systemic inflammatory response syndrome (SIRS), (ii) broadening of antibiotics, (iii) persistently positive cultures with same pathogens or persistence/worsening of clinical symptoms. They found that the clinical failure rate was 19.9% in patients who were administered PIP/TAZ by short infusion.(39) Two randomized control trials performed the clinical cure evaluation by considering disappearance of all signs and symptoms related to the infection at day 14 post antibiotic cessation. (34, 38) Abdul-Aziz et al. and Dulhunty et al. reported that the clinical cure rates of beta-lactam antibiotics with intermittent bolus were 34%(34) and 50%(38), respectively. In addition, another work of Abdul-Aziz et al. evaluated the clinical cure by considering the completion of treatment course without change or addition of antibiotics, and with no additional antibiotics commenced with 48 h of cessation. They found that 70% of patients who treated with PIP/TAZ were assessed as clinical cure.(35)

Regarding mortality rate, there is high variability of mortality rate in patients with sepsis.(85) Using PIP/TAZ to treat presumed or microbiologically confirmed infections, Cutro et al. reported that inpatient mortality rate was 14% in patients with sepsis (39),while Fan et al. found that 14-day and inpatient mortality rate were 14% and 30%, respectively in critically ill patients.(40) Findings from 2 randomized controlled trials, Abdul-Aziz et al. reported that 30-day survival rate was 63% (34) while Dulhunty et al. found that 90-day survival rate was 75% (38) in patients with severe sepsis who were treated with beta-lactam antibiotics including PIP/TAZ. Additionally, another study of Abdul-Aziz et al. documented that 30-day survival rate was 72% in critically ill patients who received PIP/TAZ throughout the therapy.(35) Moreover, Acute Physiology and Chronic Health

Evaluation II (APACHE II) score has been found to be a significant predictor for mortality rate.(36, 40) Robert et al. reported that APACHE II score was significantly related to hospital mortality in patients with severe sepsis.(36) Likewise, Fan et al. found that APACHE II score of 29.5 or higher was the significant predictor for 14-day mortality rate in critically ill patients.(40)

CHAPTER III RESEARCH METHODOLOGY

3.1. Research design

This study is a prospective open-label study to investigate population pharmacokinetics (PKs) and pharmacodynamics (PDs) of piperacillin in critically ill patients during the early phase of sepsis using sparse sampling data.

3.2. Scope

This study was conducted in critically ill patients with sepsis receiving piperacillin/tazobactam in Songklanagarind hospital between March 2014 and March 2017.

3.3. Operational definitions

- Critically ill patients with sepsis: Patients who are considered according to the third international consensus definitions for sepsis and septic shock (sepsis-3) (14)
- The early phase of sepsis: The first 24 hours after sepsis diagnosis and piperacillin/tazobactam prescription.
- Resuscitation fluids: Fluids used to restore intravascular volume.(86) In this study, they refer to normal saline solution and albumin colloidal solution.
- Vasoactive medications: Medications used to improve tissue perfusion and oxygen delivery.(87) In this study, they refer to norepinephrine, epinephrine, dobutamine, and dopamine.
- Probability of target attainment (PTA): Probability which the pharmacokinetic/pharmacodynamics target is achieved at a certain minimum inhibitory concentration (MIC).(88) In this study, the PK/PD target is 90% of dosing interval which free drug concentrations are above MIC in the range of 0.008 to 512 mg/L.

- Cumulative fraction of response (CFR): The expected population probability of target attainment for a specific drug dose and a specific population of microorganisms. It is calculated by multiplying the PTA at each MIC by the fraction of organisms susceptible at that concentration of the respective MIC distribution.(88)
- The 28-day all-causes mortality rate: The percentage of dead patients with all causes within 28 days after piperacillin/tazobactam administration.

3.4. Research methods

3.4.1. Population and sample

Population

Critically ill patients with sepsis who were treated with piperaciliin/tazobactam

Sample

Critically ill patients with sepsis who were treated with piperacillin/tazobactam in Songklanagarind hospital

Inclusion criteria

1. Patients who aged 15 years or older.
2. Patients who were treated with piperacillin/tazobactam.
3. Patients with sepsis defined according to the third international consensus definitions for sepsis and septic shock (Sepsis-3).(14)
4. Patients or their next of kins who consented.

Exclusion criteria

1. Patients who were known or suspected hypersensitivity to piperacillin/tazobactam.
2. Pregnant patients.
3. Patients who used peritoneal dialysis, hemodialysis, or continuous renal replacement therapy.
4. Patients who were expected to death within 48 hours by the principle investigator.

3.4.2. Data collection

Demographic and clinical data of patients were recorded including gender, age, total body weight (TBW), systolic/diastolic blood pressure (SBP/DBP), ratio of partial pressure of oxygen and fraction of inspired oxygen (Pao_2/Fio_2), serum creatinine (S_{Cr}), total bilirubin, complete blood count (CBC), acute physiology and chronic health evaluation II (APACHE II) score (see appendix A), sequential organ failure assessment (SOFA) score (see appendix B), suspected infections, microbial cultures, antibiotic therapy, total amount of resuscitation fluids per day, the uses of vasoactive medications and mechanical ventilators, concomitant medication and diseases.

3.4.3. Doses and Drug administration

According to the standard practice of Songklanagarind hospital, dose of piperacillin/tazobactam for patients with CL_{Cr} > 60 mL/min is 4/0.5 g every 6 hours. Dose for patients with $CL_{Cr} \leq 60$ mL/min is 2/0.25 g every 6 hours; however, at the physician's discretion, the dose could be 4/0.5 g every 6 hours in case of sepsis. It was reconstituted and diluted with 100 mL of normal saline solution before given intravenously as a 30-minute infusion.

3.4.3. Blood sampling

This study carried out during the initial 24 hours of sepsis and piperacillin/tazobactam administration. Blood samples (5 mL) were obtained at the following times: before (time 0) and then during 0 to 0.5, 0.5 to 2, 2 to 4 and 4 to 6 or 8 hours (depending on the physician's prescription) after piperacillin/tazobactam administration by a trained research nurse. All blood samples were added to heparinized tubes and centrifuged at 1000 × g for 10 min within 30 min. All samples were stored at -80°C and analyzed within 30 days.

3.4.4. free piperacillin assay

The free concentrations of piperacillin were determined using a validated high-performance liquid chromatography (HPLC) (89) with minor modifications. Briefly, 300 μL of plasma was subjected to ultrafiltration using a Nanosep 10K device with omega membrane (Pall Corp., Ann Arbor, MI, USA); the device was centrifuged at 10,000 x g for 15 min at 4 °C. A 30 μL aliquot of the sample was

injected into a Nova-Pack C18 column (150 mm by 3.9 mm inside diameter, 4 μm particle size; Waters associates) using an automated injection system (Waters e2695 Plus autosampler; Waters associates, Milford, MA) at 4 °C. Piperacillin was eluted from the column in 7.2 min with a gradient of 20 mM KH_2PO_4 pH 2.4 (buffer A) and acetonitrile (buffer B) (0-7 min 95%A; 7-7.1 min 50%A; 7.1-10 min 95%A), at a flow rate of 1.0 mL/min. The column effluent was monitored at 220 nm with a photodiode array detector (Waters 2996; Waters associates, Milford, MA). Peaks were recorded and integrated with a Waters 746 data module (Waters associates). The lower limit of quantification for piperacillin was 0.25 μg/mL. The standard curve was linear over the concentration range of 0.25 to 500 μg/mL ($r \geq 0.999$). The intraassay and interassay reproducibility values, characterized by coefficients of variations (CVs), were ranged from 0.31 to 9.79% and 0.80 to 12.81%, respectively. The determination of piperacillin concentrations was performed by the clinical pharmacology laboratory, department of internal medicine, faculty of medicine, Prince of Songkla University.

3.4.5. Population PK analysis

Nonlinear mixed effects model building

The piperacillin concentration versus time data were analyzed by a nonlinear mixed-effects modeling approach using NONMEM® version 7.2 (ICON Development Solutions, Ellicott city, MD, USA) through PDx-Pop® version 5.1 (ICON Development Solutions, Ellicott city, MD, USA). Data were fitted with a structural model and 2 variance models including interindividual variability (IIV) and residual variability (RV) models.

- Structural model (the model that best describes data in the absence of covariates): one-compartment model (ADVAN1 TRANS2) and two-compartment model (ADVAN3 TRANS4) were examined with the data.
- Interindividual variability (IIV) model describes the variance of a parameter across different individuals in the population. Three IIV models were investigated;

Additive IIV model

$$P_i = TVP + \eta_i$$

Proportional IIV model

$$P_i = TVP\ (1 + \eta_i)$$

Exponential IIV model

$$P_i = TVP\ e^{\eta i}$$

where P_i is the value of the PK parameter for the i^{th} subject.

TVP is the typical value of the population PK parameter.

η_i is the value of the deviation from the typical value for the i^{th} subject, it is assumed to have normal distribution with zero mean and ω^2 variance.

▪ Residual variability (RV) model describes the unexplained variability in the observed data. Four RV models were investigated;

Additive RV model

$$C_{O,\,ij} = C_{P,\,ij} + \varepsilon_{ij}$$

Proportional RV model

$$C_{O,\,ij} = C_{P,\,ij}(1 + \varepsilon_{ij})$$

Combined additive and proportional RV model

$$C_{O,\,ij} = C_{P,\,ij} + \varepsilon_{1,\,ij} + (1 + \varepsilon_{2,\,ij})$$

Exponential RV model

$$C_{O,\,ij} = C_{P,\,ij}\ e^{\varepsilon ij}$$

where $C_{O,\,ij}$ is the observed concentration of the i^{th} subject at time j^{th}.

$C_{P,\,ij}$ is the predicted concentration of the i^{th} subject at time j^{th}.

ε_{ij} is the value of the difference between the observed and predicted concentrations of the i^{th} subject at time j^{th}, it is assumed to have normal distribution; zero mean and σ^2 variance.

The appropriate base model was chosen based on objective function value (OFV), Akaike information criterion (AIC), PK parameter estimates, and goodness of fit plots. The model which had the lowest OFV and AIC was considered that it could best describe the data; however PK parameter estimates

of such model had to be reasonable and reliable. Then the chosen base model would be added with predefined covariates.

Covariate exploration

There were 8 plausible covariates explored as follows:

1. Total body weight (TBW)

2. Adjusted body weight (ABW) was calculated by

ABW = IBW + [0.4 x (TBW-IBW)](69)

3. Creatinine clearance was calculated by the Cockcroft-Gault equation if renal function was stable or the Jelliffe equation(90) (see appendix C) if renal function was unstable (an increase in serum creatinine by at least 0.3 mg/dL within 24 to 48 hours)

4. Mean arterial pressure (MAP) was calculated by

MAP = DBP + (1/3(SBP - DBP))

5. Total bilirubin

6. Total amount of resuscitation fluids per day

7. The uses of vasoactive medications

8. The uses of mechanical ventilators

Collinearities between studied covariates were investigated. Then Individual PK parameter estimates from the chosen base model were plotted against individual covariate values to assess relationships. After that studied covariates were included in the base model with different patterns of relationship as follows:

- Continuous covariates

  Linear covariate model

  $$TVP = \theta_1 + \theta_2 \times (COV - COV_{Median})$$

  Power covariate model

  $$TVP = \theta_1 (COV/COV_{Median})^{\theta 2}$$

  Exponential covariate model

  $$TVP = \theta_1 e^{(\theta 2 \times (COV/COV_{Median}))}$$

  where TVP is the typical value of the population PK parameter.

  COV is the value of the continuous covariate.

  COV_{Median} is the median of the continuous covariate.

θ_1 is the PK parameter value when the individual covariate is median (for linear and power model), zero (for exponential model)

θ_2 is the value of the change in PK parameter for unit change in covariate for linear model, the value of the change in Ln(PK parameter) per unit change in Ln(covariate) for power model, and the value of the change in Ln(PK parameter) per unit change in covariate for exponential model.

- Categorical covariates

  Fractional change covariate model

  $$TVP = \theta_1 \times (1+(\theta_2 \times Cov))$$

  where TVP is the typical value of the population PK parameter. COV is the value of the continuous covariate.

  θ_1 is the typical PK parameter value without covariate

  θ_2 is the value of the change in PK parameter when the covariate presents

The stepwise approach was implemented to obtain the final model. Based on the χ^2 test, a decrease in the OFV of 3.84 units is considered to be significant ($P < 0.05$) for forward addition step and an increase in the OFV of 6.64 units is considered to be significant ($P < 0.01$) for backward deletion step to avoid any possible false positives.

Estimation methods

To obtain accurate and precise PK parameter estimates, the chosen estimation method is an important part of a population PK analysis. The first-order conditional estimation with interaction (FOCE-I) is a classical method commonly used while the stochastic approximation expectation maximization (SAEM) is a newer estimation algorithm suggested for better performance when applied with complex models and/or sparse data. However, Sukarnjanaset et al conducted the work to evaluate the performance (accuracy, precision, completed estimations, and runtimes) of FOCEI and SAEM estimation methods in population PK analysis using NONMEM® when implemented with the simple models (one- and two-compartment models) across rich, medium, and

sparse sampling data. (See the appendix D) The work found that FOCEI provided comparable performance similar to SAEM but with significantly shorter runtimes across sparse, medium, and rich data scenarios.(91) Therefore FOCEI was chosen to use in PK parameter estimation of this study.

Model Evaluation

Basic goodness-of-fit plots of models were used to evaluate by visual inspection; (i) observed and population predicted concentrations versus time, (ii) observed and individual predicted concentrations versus time, (iii) individual weighted residual errors versus time and (iv) individual weighted residual errors versus individual predicted concentrations. In addition, the nonparametric bootstrap of 1,000 datasets was performed to assess the model reliability.(67) Moreover, visual predictive check was performed by simulating 1,000 subjects to assess the predictive performance of the model.(66, 67) Graphical displays of basic goodness-of-fit plots and visual predictive check were performed using R program version 3.1.0 (Free Software by the R project for statistical computing) and Xpose program version 4.4.0 (Free Software by the Xpose development team).

3.4.6. Pharmacodynamic assessment using Monte Carlo simulation (MCS)

Population PK parameter estimates and their variabilities from the validated final model were used to simulate 10,000 patients using Crystal ball software (Decisioneering Inc., Denver, CO, USA). Virtual patients were simulated in 4 different renal functions (CL_{Cr} < 20, 20 to 40, 40 to 60, and 60 to 120 mL/min). Forty dosage regimens were created based on 4 dosage regimens; (i) piperacillin/tazobactam 2/0.25 g every 8 hours (6/0.75 g/day), (ii) piperacillin/ tazobactam 2/0.25 g every 6 hours (8/1 g/day), (iii) piperacillin/ tazobactam 4/0.5 g every 8 hours (12/1.5 g/day), and (iv) piperacillin/tazobactam 4/0.5 g every 6 hours (16/2 g/day). Each dosage regimen was varied with 6 different infusion times including 0.5, 1, 2, 3, 4 hours, and continuous infusion (CI). Concerning loading dose (LD), LD 2/0.5 g and 4/0.5 g were added to 3 CI dosage regimens; (i) piperacillin/tazobactam 6/0.75 g CI, (ii) piperacillin/tazobactam 8/1 g CI, and (iii) piperacillin/tazobactam 12/1.5 g CI (since maximum daily dose is 16/2 g/day,

16/2 g CI would not be added with any LD). In addition, 2 dosage regimens; (i) piperacillin/tazobactam 2/0.25 g every 6 hours and (ii) piperacillin/tazobactam 2/0.25 g every 8 hours were implemented with LD 4/0.5 g (because maximum dose of each dose should not exceed 4/0.5 g, piperacillin/ tazobactam 4/0.5 g every 6 and 8 hours would not be implemented with any LD). Then the 90%$fT_{>MIC}$ was observed to determine the PTA for a range of MICs (0.008 to 512 mg/L).

MIC distributions of *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, and *Escherichia coli* from the European Committee for Antimicrobial Susceptibility and Testing (EUCAST) database were used to determine the CFR (See appendix E)(92), it should be noted that there has been no published MIC distribution of these pathogens in Thailand. The calculation was done by multiplying the PTA at each MIC by the fraction of organisms susceptible at that MIC. Then the summation of those results was the CFR for the respective MIC distribution. The dosage regimen was considered successful if the CFR value was ≥90%.

3.4.7. Clinical outcomes

Clinical responses

Patients with sepsis who received piperacillin/tazobactam more than 48 hours or less than 48 hours because of broadening antibiotics were eligible for the clinical response assessment. The criteria of clinical improvement were (i) improved clinical signs and symptoms in relation to the suspected infections or (ii) negative repeat culture or (iii) de-escalation of antibiotic therapy (changed to antibiotics which the pathogens were susceptible to). The criteria of clinical failure were (i) persistent/worsening clinical signs and symptoms in relation to the suspected infections or (ii) persistent positive culture with the same pathogen or (iii) broadening of antibiotic therapy (changed to carbapenems or added vancomycin or aminoglycosides).

The 28-day all-cause mortality rate

The percentage of patients who died with all causes within 28 days after initiation of piperacillin/tazobactam was recorded and investigated the association with individual %$fT_{>MIC}$.

3.5. Studied outcomes

3.5.1. Primary outcomes

- Population PK parameter estimates and variabilities of piperacillin in critically ill patients during the early phase of sepsis.
- The PTA of various piperacillin dosage regimens and the CFR against pathogens commonly found in critically ill patients during the early phase of sepsis.

3.5.2. Secondary outcomes

- Clinical responses of critically ill patients with sepsis when receiving piperacillin/tazobactam.
- Twenty eight-day all-cause mortality rate and the association with individual $\%fT_{>MIC}$ in critically ill patients with sepsis who were treated with piperacillin/tazobactam.

3.6. Ethical considerations

Investigators concern about the rights of patients. Before recruitment, investigators gave all patients or caregivers both oral and written research information until patients or caregivers fully understood and could make a decision to participate or not in the study according to their willingness. Patients can leave the study anytime without effect on the regular therapy. All data would keep confidential and present by concealing patients' private profiles. The study was approved by human subjects institutional review board, faculty of medicine, Prince of Songkhla University. (See appendix E)

CHAPTER IV RESULTS

4.1. Demographic and clinical data

Forty-eight patients with sepsis participated in the study. The demographic and clinical data are shown in table 5. Seventy-seven percent of patients were male. Median age and total body weight (TBW) were 60 years and 56.6 kg, respectively. Most patients had low respiratory function; median ratio of partial pressure of oxygen and fraction of inspired oxygen (Pao_2/Fio_2) was 234 mmHg and 60% of patients used mechanical ventilators. In addition, most patients had slightly low cardiovascular function; median of mean arterial pressure (MAP) was 68 mmHg. Thirty-three percent of patients received vasoactive medications (29% of patients progressed septic shock and 4% of patients developed cardiovascular shock). Most patients had renal impairment; median creatinine clearance (CL_{Cr}) was 54.9 mL/min. Median acute physiology and chronic health evaluation II (APACHE II) and sequential organ failure assessment (SOFA) score were 22 and 6, respectively. Suspected infection mostly found was respiratory tract infection. Thirty-five pathogens were found in 28 patients, 77% of pathogens were susceptible to piperacillin/tazobactam. Most patients received piperacillin/tazobactam monotherapy. All patients received piperacillin/ tazobactam 4/0.5 g 30-min infusion every 6 hours, except for two patients who received 4/0.5 g 30-min infusion every 8 hours and two patients who received 4/0.5 g (first dose) then 2/0.25 g 30-min infusion every 6 hours.

4.2. Population pharmacokinetic (PK) analysis

A total of 237 blood samples were available for the analysis (except for 3 blood samples which could not be drawn because patients underwent surgery). Piperacillin plasma concentration versus time and time after dose profiles are depicted in figure 2 and 3, respectively.

Table 5 Demographic and clinical data

Data	**Values (n=48)**
Gender (male), n (%)	37 (77)
Age (years)	60 (49-78)
Total body weight (kg)	56.6 (49.6-69.5)
Ideal body weight (kg)	62.3 (53.8-66.4)
Adjusted body weight (kg)	56.0 (48.1-65.0)
Body mass index (kg/m^2)	21.0 (18.9-25.5)
Pa_{O2}/F_{IO2} (mmHg)	234 (142-333)
Mean arterial pressure (mmHg)	68 (61-75)
Serum creatinine (mg/dL)	1.1 (0.7-1.5)
Creatinine clearance (mL/min)[a]	54.9 (41.6-86.5)
Platelets ($x10^3/\mu L$)	185 (125-283)
Total bilirubin (mg/dL)	0.8 (0.4-2.9)
APACHE II score	22 (18-26)
SOFA score	6 (5-8)
Total amount of resuscitation fluids per day (mL)	1,265 (405-2,250)
Patients with septic shock, n (%)	14 (29)
The uses of vasoactive medications, n (%)	16 (33)
The uses of mechanical ventilators, n (%)	29 (60)
Suspected infections, n (%)[b]	
Respiratory tract infection	23 (48)
Urinary tract infection	10 (21)
Septicemia	7 (15)
Peritonitis	1 (2)
Severe dengue infection	1 (2)
No known source of infection	9 (19)

Data are presented as median (interquartile range) or number (percentage). [a]Creatinine clearance was estimated using the Cockcroft-Gault equation for patients with stable renal function and the Jelliffe equation for patients with unstable renal function. [b]Some patients had more than one suspected infection.

Table 5 Demographic and clinical data (continue)

Data	**Values (N=48)**
Pathogens, n (%)[c]	
Escherichia coli (ESBL)	7 (20)
Escherichia coli	6 (17)
Klebsiella pneumoniae (ESBL)	5 (14)
Pseudomonas aeruginosa	5 (14)
Klebsiella pneumoniae	3 (9)
Acinetrobacter baummanii	3 (9)
Enterococcus cloacea	3 (9)
Streptococcus pneumonia	1 (3)
Staphylococcus aureus	1 (3)
Enterobacter aerogenes	1 (3)
Susceptibility of pathogens, n (%) (N=35)	
Susceptible to piperacillin/tazobactam	27 (77)
Not susceptible to piperacillin/tazobactam	6 (17)
No known susceptibility	2 (6)
Comorbidities, n (%)[d]	
Cancer	19 (37)
Diabetic mellitus	6 (12)
Hypertension	6 (12)
Cirrhosis	5 (10)
Myocardial infarction	4 (8)
Dyslipidemia	2 (4)
Chronic obstructive pulmonary disease	2 (4)
Schizoaffective and mood disorder	2 (4)

Data are presented as median (interquartile range) or number (percentage). [c]Some patients were affected by more than one pathogen. [d]Some patients had more than one comorbidity.

Table 5 Demographic and clinical data (continue)

Data	**Values (N=48)**
Comorbidities, n (%)[d]	
Thyroid	1 (2)
Burn	1 (2)
Atrial fibrillation	1 (2)
Alzheimer disease	1 (2)
Lung alectasis	1 (2)
Aplastic anemia	1 (2)
Concomittent antibiotics, n (%)	
No concomittent antibiotics	43 (90)
Vancomycin	3 (6)
Metronidazole	2 (4)

Data are presented as median (interquartile range) or number (percentage). [d]Some patients had more than one comorbidity.

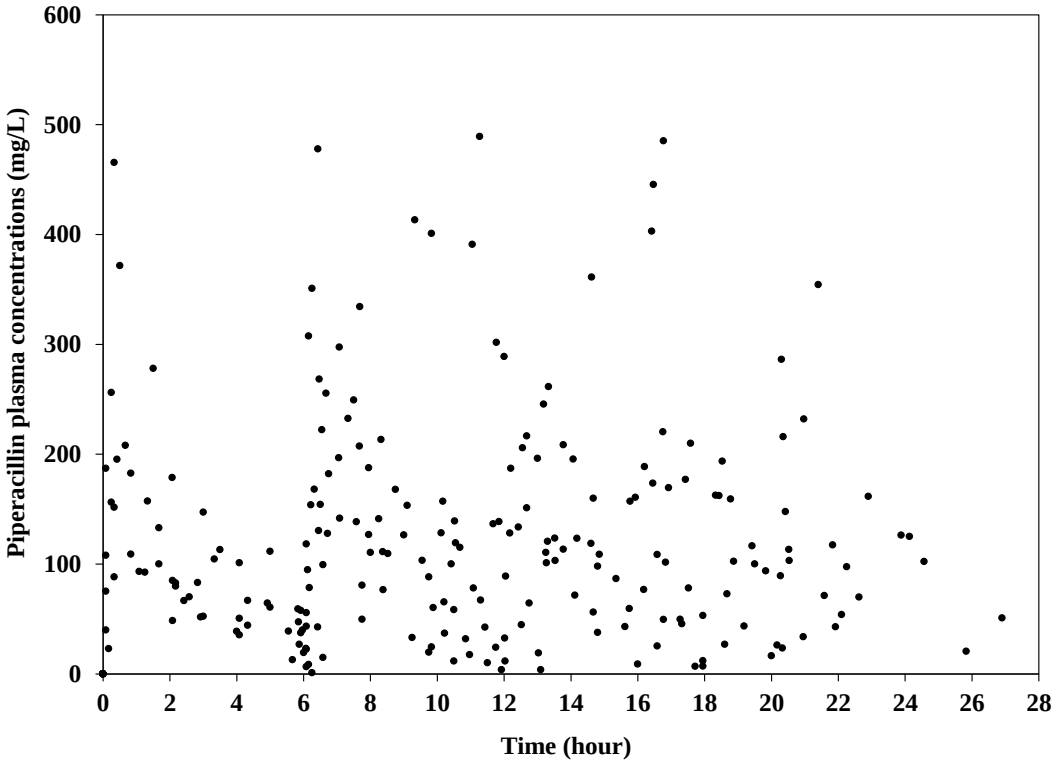

Figure 2 Piperacillin plasma concentrations (mg/L) versus time (hour) of critically ill patients with sepsis (n=48)

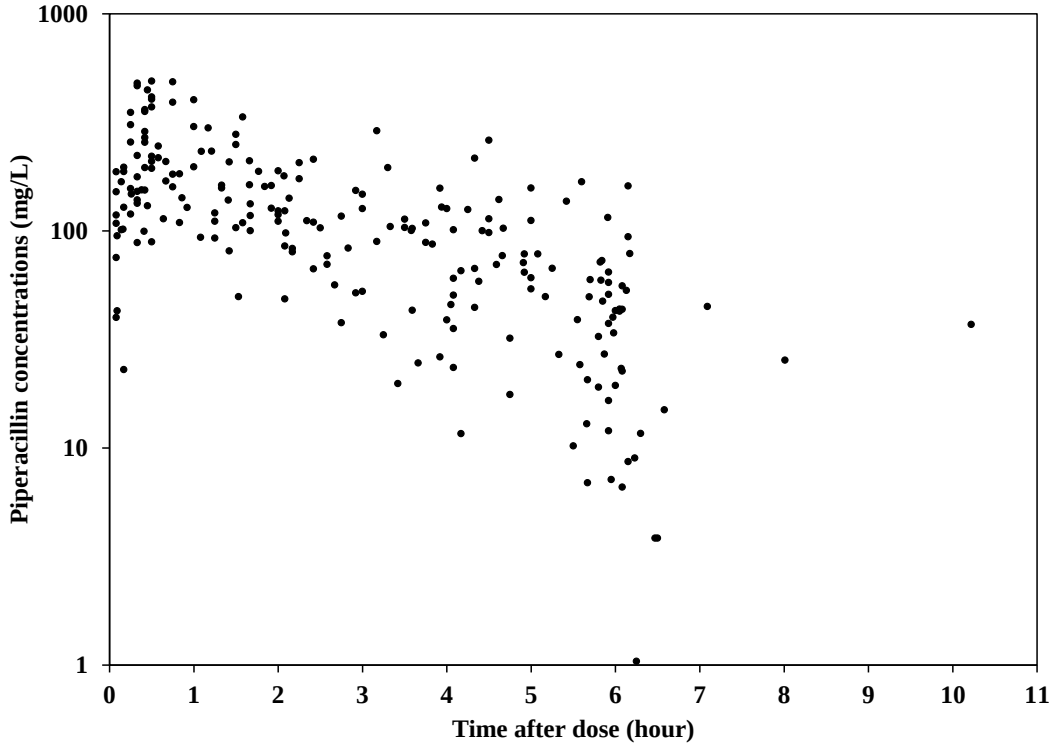

Figure 3 Piperacillin plasma concentrations (mg/L) versus time after dose (hour) of critically ill patients with sepsis (n=48)

4.2.1. Base models

Piperacillin plasma concentration-time profiles were fitted with the one and two-compartment models with different interindividual variability (IIV) and residual variability (RV). Results of objective function value (OFV), Akaike information criterion (AIC) and population PK parameter estimates from the one and two-compartment models with different IIV and RV models are shown in table 6 and 7, respectively. Overall, two-compartment models provided lower OFV and AIC than one-compartment models did. Among the two-compartment models, exponential IIV models could provide successful estimations (minimization successful). Regard as the two-compartment models with exponential IIV models, combined RV model provided the least OFV and AIC; however, an estimate of additive RV was not reliable (95% CI crossed zero). Therefore the two-compartment model with exponential IIV and proportional RV models was chosen as the appropriate base model. The goodness-of-fit plots of the base model were shown in figure 4. The base model could provide predicted concentrations which corresponded to observed concentrations although they were not consistent for the high observed piperacillin concentrations (data points were scattered around the identity line, excluding high observed piperacillin concentrations), as presented in figure 4a and b. In addition, weighted residuals (the differences between observed and predicted concentrations) were scattered around the horizontal zero line and were within ±1 along with all predicted concentrations and time, as shown in figure 4c and d.

Table 6 OFV, AIC and population PK parameter estimates from the one-compartment model with different IIV and RV models

IIV models	RV models	OFV	AIC	CL (L/h)[a]	V_d (L)[a]	IIV_{CL}[b]	IIV_{Vd}[b]	RV[b]
Additive	Additive[c]	1983.552	1993.550	6.57 (8.42)	15.0 (8.3)	3.06	6.47	28.7
	Proportional[c]	1904.624	1914.620	5.46 (0.10)	14.8 (0.1)	2.04	5.06	29.1
	Combined additive and proportional[c]	1911.584	1923.580	5.75 (6.63)	15.8 (7.2)	2.09	5.10	0.348 (Add) 30.0(Prop)
	Exponential[c]	1904.624	1914.620	5.46 (0.10)	14.8 (0.1)	2.04	5.06	29.1
Proportional	Additive[c]	1983.552	1993.550	6.57 (8.42)	15.0 (8.3)	46.6	43.2	28.7
	Proportional[d]	1904.197	1914.200	5.66	14.9	36.1	33.6	29.9
	Combined additive and proportional[d]	1911.584	1923.580	5.74	15.8	36.5	32.2	0.346 (Add) 30.0(Prop)
	Exponential[d]	1904.197	1914.200	5.66	14.9	36.1	33.6	29.9
Exponential	Additive[c]	1973.984	1983.980	5.87 (7.85)	13.7 (8.9)	52.0	55.9	28.2
	Proportional[c]	1908.928	1918.930	5.37 (6.72)	15.5 (7.3)	40.5	43.2	29.2
	Combined additive and proportional[c]	1898.787	1910.790	5.35 (6.67)	14.8 (7.3)	41.8	46.8	3.58 (Add) 26.6(Prop)
	Exponential[c]	1908.928	1918.930	5.37 (6.72)	15.5 (7.3)	40.5	43.2	29.2

[a] Data were presented as population parameter estimates (% relative standard error). b IIV and RV of additive models were presented as standard deviation, IIV and RV of proportional and exponential models were presented as % coefficient variation. [c] The model estimated parameters successfully (minimization successful). [d] The model could not throughout estimate parameters (minimization terminated). Add, Additive model; Prop, Proportional model.

Table 7 OFV, AIC and population PK parameter estimates from the two compartment model with different IIV and RV models

IIV models	RV models	OFV	AIC	CL (L/h)[a]	V_1 (L)[a]	V_2 (L)[a]	Q (L/h)[a]	IIV_{CL}[b]	IIV_{V1}[b]	RV[b]
Additive	Additive[c]	-	-	-	-	-	-	-	-	-
	Proportional[d]	1861.315	1879.320	6.69	4.3	14.4	53.9	3.41	2.00	32.9
	Combined additive and proportional[d]	1860.277	1880.280	6.69	4.3	14.4	54.3	3.41	2.03	0.417(Add) 32.7(Prop)
	Exponential[d]	1861.315	1879.320	6.69	4.3	14.4	53.9	3.41	2.00	32.9
Proportional	Additive[c]	-	-	-	-	-	-	-	-	-
	Proportional[d]	1835.206	1853.210	6.49	8.6	11.6	38.5	68.9	62.0	26.1
	Combined additive and proportional[d]	1808.725	1828.720	6.34	9.7	9.3	28.9	45.1	55.6	3.06 (Add) 19.4(Prop)
	Exponential[d]	1821.666	1839.670	6.00	9.3	8.6	30.0	42.9	42.3	24.1
Exponential	Additive[e]	1884.848	1902.850	5.34 (7.47)	11.5 (9.1)	7.1 (11.9)	6.7 (18.5)	51.8	59.8	19.7
	Proportional[e]	1811.109	1829.110	5.65 (7.47)	9.6 (13.2)	7.6 (15.8)	20.1 (27.9)	46.3	64.0	22.1
	Combined additive and proportional[e]	1805.582	1825.580	5.64 (7.41)	9.7 (14.7)	7.5 (16.2)	19.1 (32.9)	46.6	64.4	1.94[f] (Add) 20.9(Prop)
	Exponential[e]	1811.109	1829.110	5.65 (7.47)	9.6 (13.2)	7.6 (15.8)	20.1 (27.9)	46.3	64.0	22.1

[a] Data were presented as population parameter estimates (% relative standard error). [b] IIV and RV of additive models were presented as standard deviation, IIV and RV of proportional and exponential models were presented as % coefficient variation. [c] The model could not estimate parameters at the initial evaluation. [d] The model could not throughout estimate parameters (minimization terminated). [e] The model estimated parameters successfully (minimization successful). [f]95%CI of the value crossed zero (The value was not reliable). Add, Additive model; Prop, Proportional model.

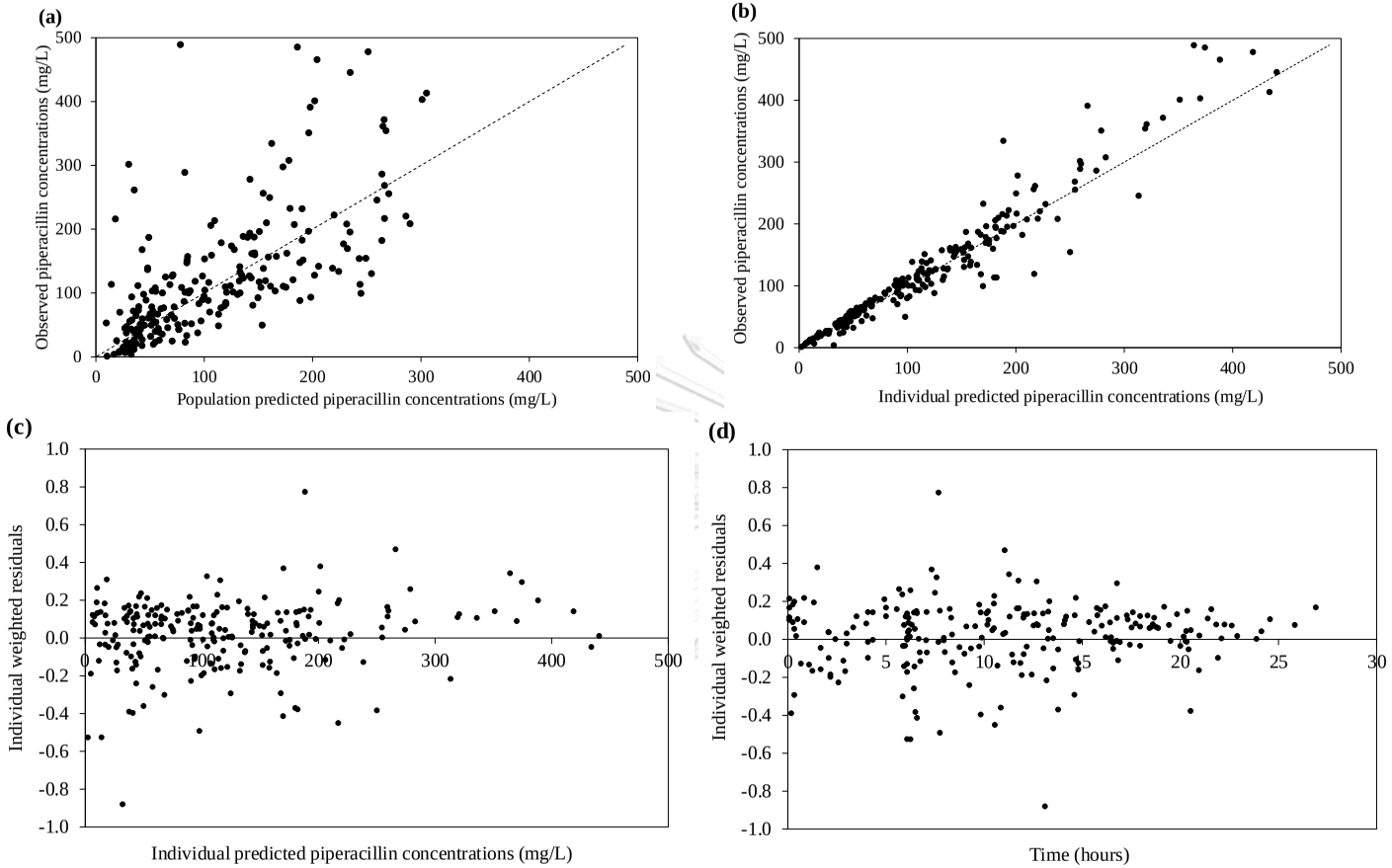

Figure 4 The goodness-of-fit plots of the base model.

(a) Observed versus population predicted piperacillin concentrations; (b) Observed versus individual predicted piperacillin concentrations; (c) Individual weighted residuals versus individual predicted piperacillin concentrations; (d) Individual weighted residuals versus time. Dotted lines, identity lines.

4.2.2. Covariate models and the final model

The collinearity between studied covariates were explored before covariate model building, as shown in figure 5 and 6. There was no collinearity between studied covariates except total body weight (TBW) and adjusted body weight (ABW). The relationships between plausible covariates and PK parameters including piperacillin clearance (CL) and central volume of distribution (V_1) are shown in figure 7 and 8, respectively. Regarding CL, among 8 studied covariates; TBW, ABW, creatinine clearance (CL_{Cr}), mean arterial pressure (MAP), total amount of resuscitation (RFLU), total bilirubin (TBIL), the uses of vasoactive medications (VASO) and mechanical ventilators (VENT); TBW, ABW, CL_{Cr} and MAP were likely to have a correlation with CL but their relationship patterns were unclear. Concerning V_1, 4 covariates; TBW, ABW, RFLU and VENT, were examined, the plots show that TBW and ABW tended to have a linear relationship with central volume of distribution (V_1).

The impacts of covariates on PK parameters were examined by using stepwise forward addition and backward deletion approach. The results of forward addition step 1 are shown in table 8. The relationships between CL_{Cr} and CL (linear), MAP and CL (linear), TBW and V_1 (linear) and ABW and V_1 (all patterns) significantly reduced the OFV of the base model. The relation between CL_{Cr} and CL (linear) provided the largest decline of the OFV; therefore it was firstly added in the base model.

Based on the base model added with the relation between CL_{Cr} and CL (linear), the rest covariates were individually included in this model. The results of forward addition step 2 are shown in table 9. The relation between MAP and CL, TBW and V_1, and ABW and V_1 significantly decreased the OFV of the model regardless patterns of relation. Because the relation between MAP and CL (linear) most decreased the OFV, it was included in the model, subsequently.

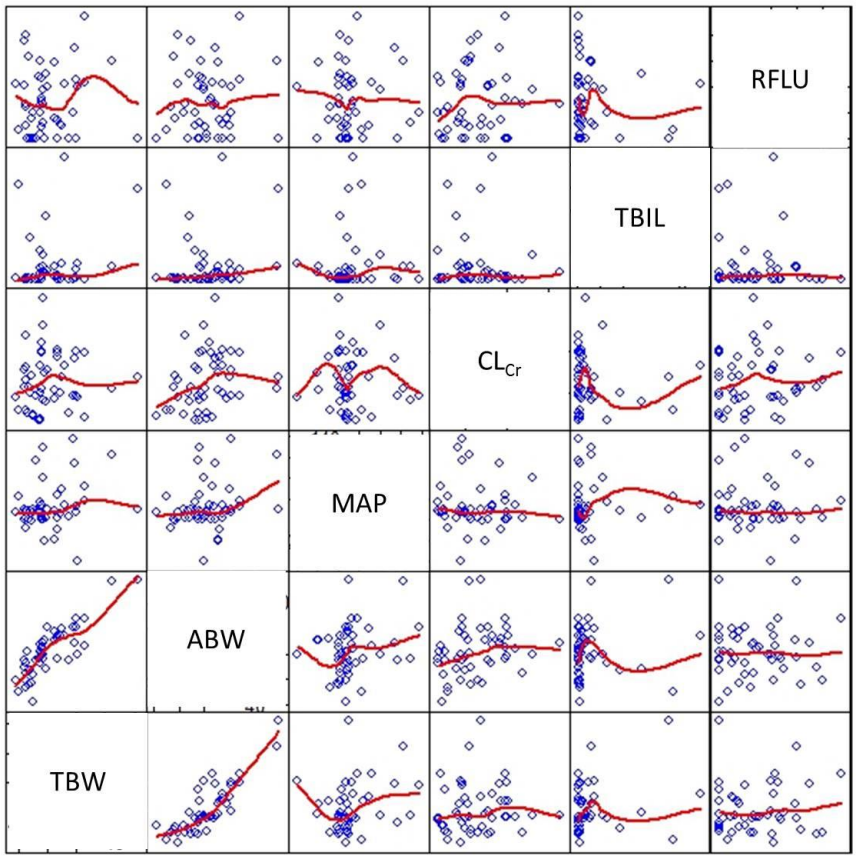

Figure 5 Scatterplot matrix of continuous covariates

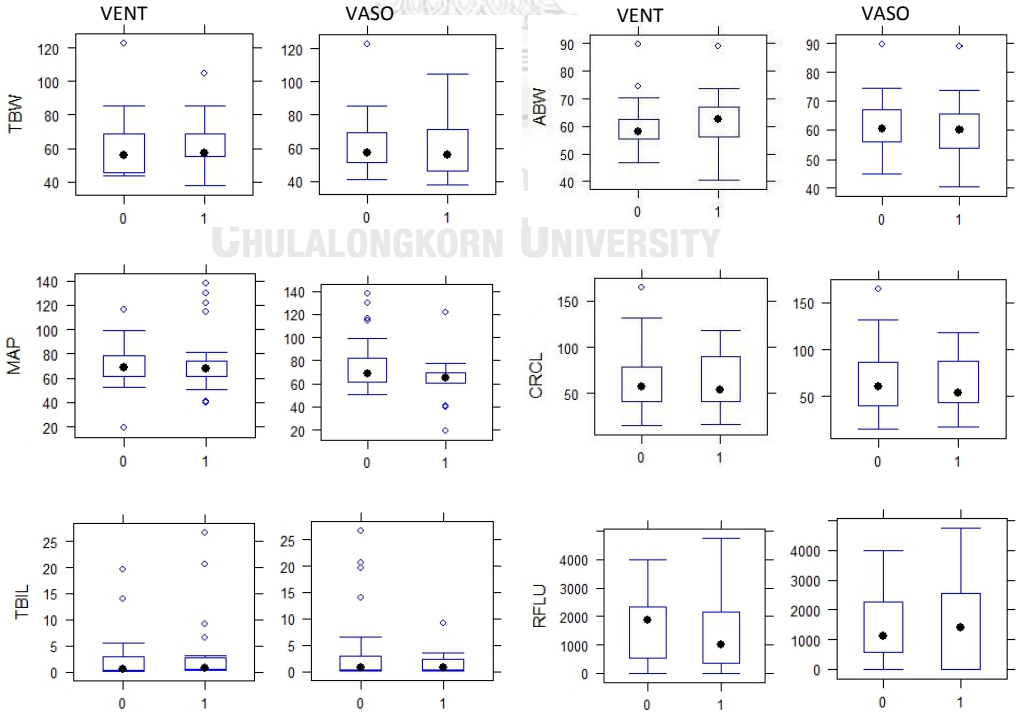

Figure 6 Boxplot matrix of studied covariates

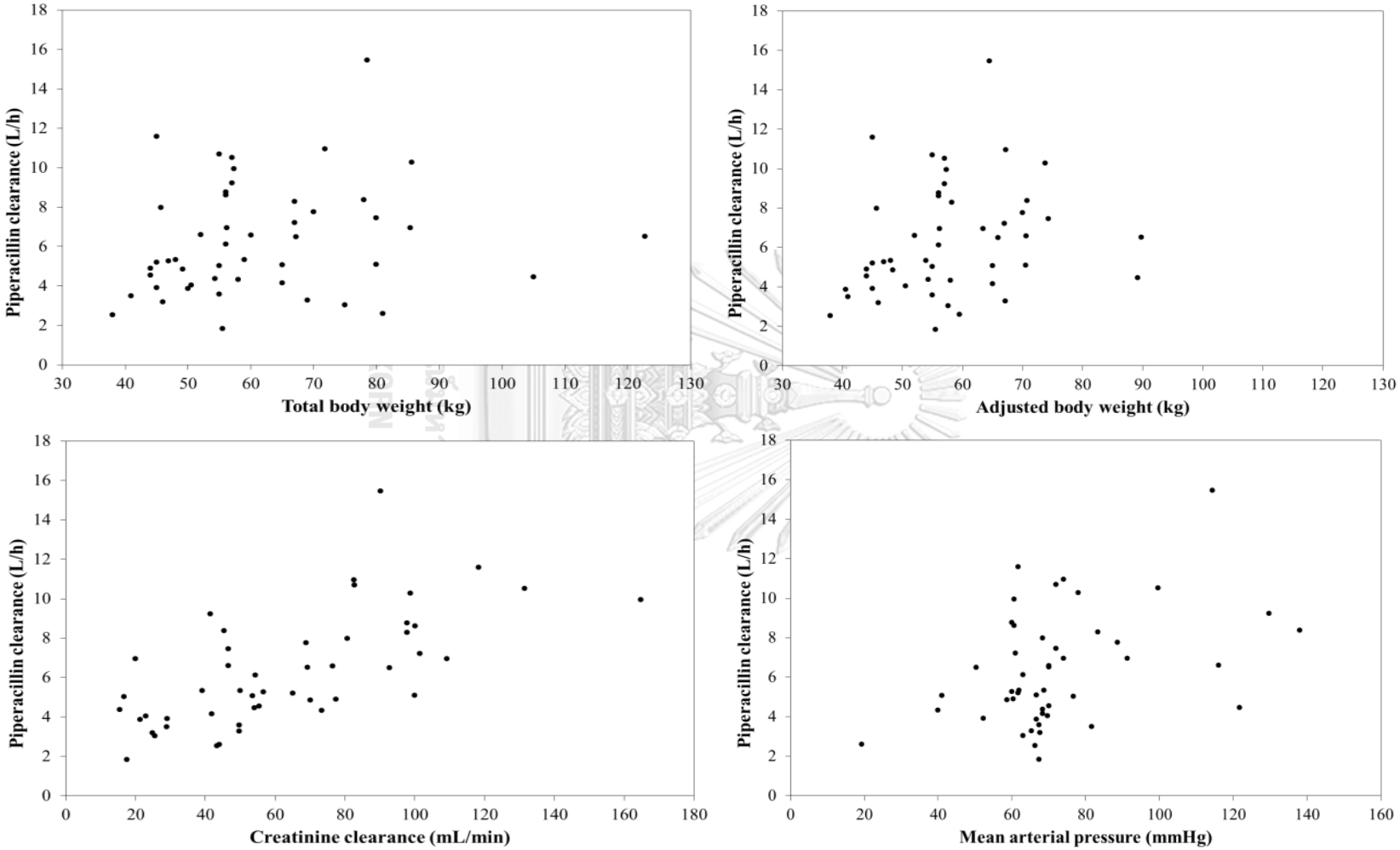

Figure 7 Plots of the relationships between individual covariate values and piperacillin clearance

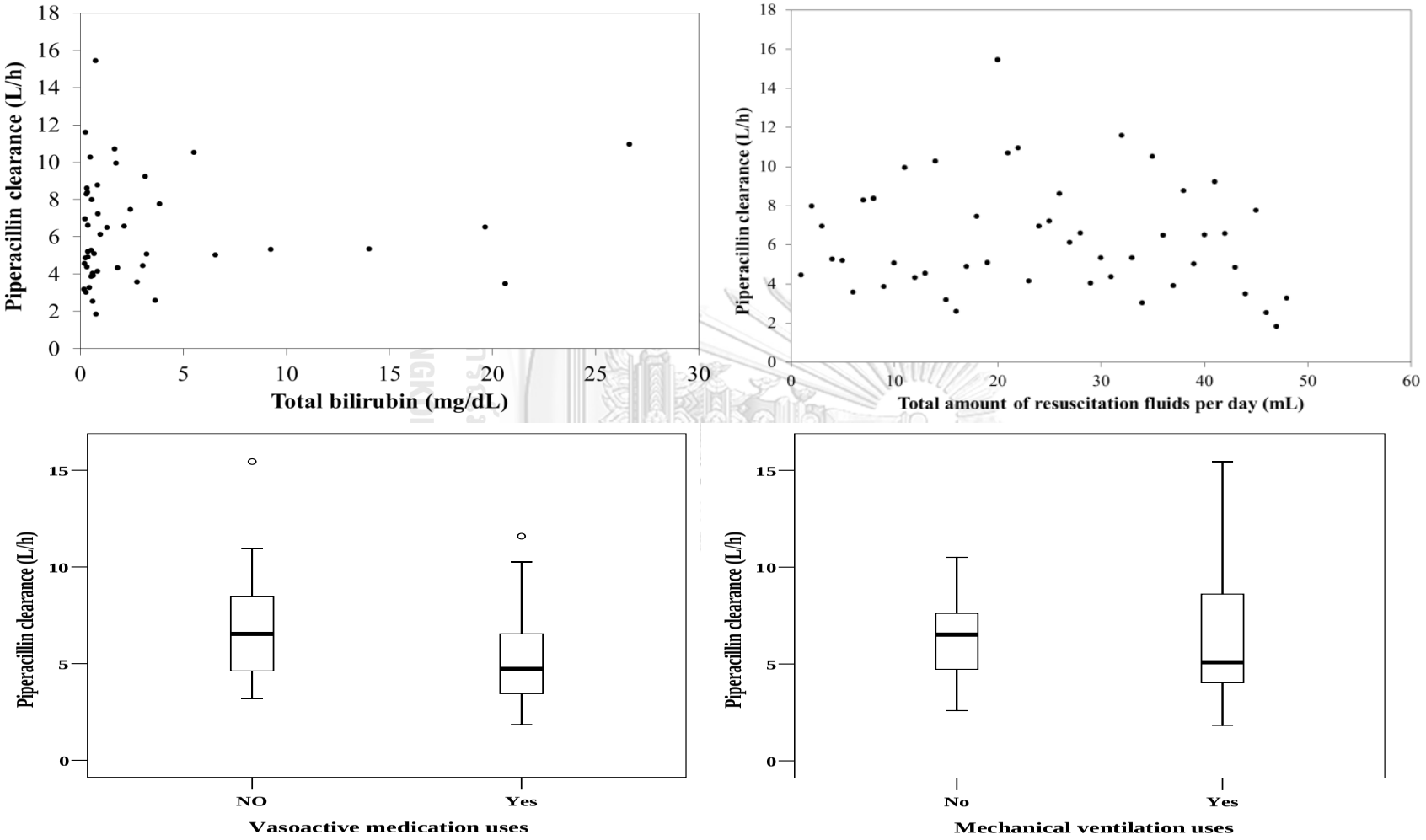

Figure 7 Plots of the relationships between individual covariate values and piperacillin clearance (continue)

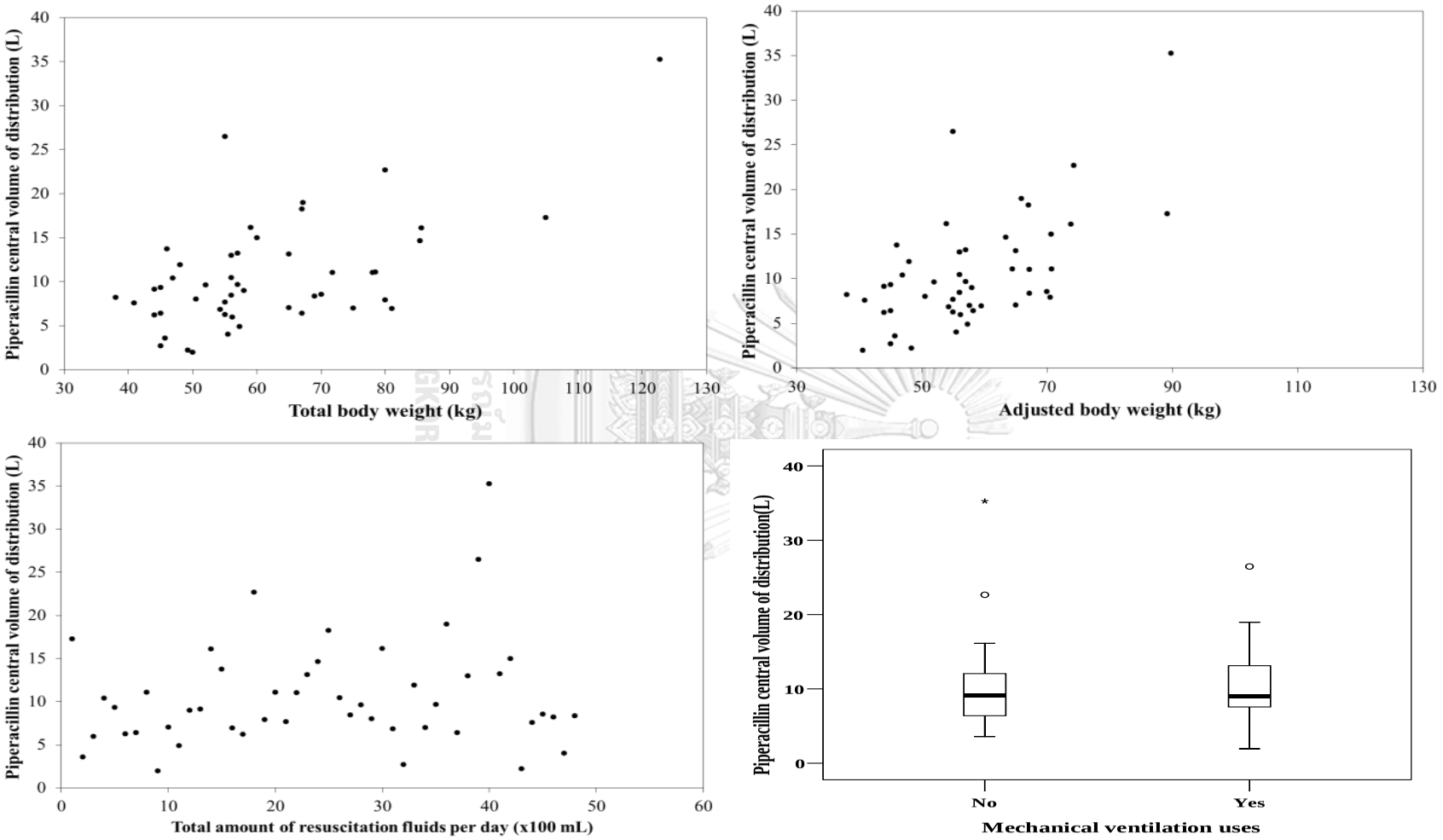

Figure 8 Plots of the relationships between individual covariate values and piperacillin central volume of distribution

Regarding the base model with the 2 linear relations; (i) CL_{Cr} and CL, and (ii) MAP and CL, the remainder covariates were separately included in this model. The results of forward addition step 3 are shown in table 10. The association of V_1 with TBW and ABW significantly lowered the OFV of the model regardless patterns of relationship. ABW was chosen to be in the model because ABW provided higher OFV reduction than TBW (ABW fitted with the data better than TBW). Among 3 patterns of relations between ABW and V_1, the linear relationship was applied in the model because it is a simpler way to describe the influence of ABW on V_1. Then there was no significant relation to improve model further as shown in table 11. The full model consisted of 3 significant linear relations including (i) CL_{Cr} and CL, (ii) MAP and CL, and (iii) ABW and V_1.

Afterward, the backward deletion was implemented by removing each covariate from the full model. There was no insignificant relation as shown in table 12. Therefore the final model was represented by equation (1) and (2);

$$TVCL = 5.37 + (0.06 \times (CL_{Cr} - 55)) + (0.05 \times (MAP - 68)) \quad (1)$$

$$TVV_1 = 9.35 + (0.26 \times (ABW - 56)) \quad (2)$$

where TVCL is the typical value of CL.

TVV_1 is the typical value of V_1.

Compared with the base model, the final model could reduce the IIV values of CL and V_1 of 17.8 and 8.6 %, respectively as shown in table 13. The final model showed that population CL and V_1 were 5.37 L/h and 9.35 L, respectively. CL_{Cr} and MAP had the significant effect on CL. In other words, if CL_{Cr} increases 1 mL/min, CL would heighten 0.06 L/h (when MAP is 68 mmHg). Likewise, if MAP increases 1 mmHg, CL would escalate 0.05 L/h (when CL_{Cr} is 55 mL/min). In addition, ABW was found to be the significant covariate for V_1, if ABW rises 1 kg, V_1 would increase 0.26 L.

Table 8 Change in OFV during forward addition step 1

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model			1811.109	
CL	TBW (Lin)	$CL = \theta_1 + (\theta_2 \times TBW-57)$	NA[a]	
	TBW (Pow)	$CL = \theta_1 \times (TBW/57)^{\theta 2}$	1839.817	+28.708
	TBW (Exp)	$CL = \theta_1 e^{(\theta 2 \times TBW/57)}$	1840.549	+29.440
	ABW (Lin)	$CL = \theta_1 + (\theta_2 \times ABW-56)$	NA[a]	
	ABW (Pow)	$CL = \theta_1 \times (ABW/56)^{\theta 2}$	1807.279	-3.830
	ABW (Exp)	$CL = \theta_1 e^{(\theta 2 \times ABW/56)}$	1808.128	-2.981
	CL_{Cr} (Lin)	**$CL = \theta_1 + (\theta_2 \times CL_{Cr}-55)$**	**1785.696**	**-25.413***
	CL_{Cr} (Pow)	$CL = \theta_1 \times (CL_{Cr}/55)^{\theta 2}$	1822.629	+11.52
	CL_{Cr} (Exp)	$CL = \theta_1 e^{(\theta 2 \times CL_{Cr}/55)}$	1823.691	+12.582
	MAP (Lin)	$CL = \theta_1 + (\theta_2 \times MAP-68)$	1803.714	-7.395*
	MAP (Pow)	$CL = \theta_1 \times (MAP/68)^{\theta 2}$	1833.824	+22.715
	MAP (Exp)	$CL = \theta_1 e^{(\theta 2 \times MAP/68)}$	1834.305	+23.196
	TBIL (Lin)	$CL = \theta_1 + (\theta_2 \times TBIL-0.8)$	1810.699	-0.410
	TBIL (Pow)	$CL = \theta_1 \times (TBIL/0.8)^{\theta 2}$	1810.802	-0.307
	TBIL (Exp)	$CL = \theta_1 e^{(\theta 2 \times TBIL/0.8)}$	1810.678	-0.431
	RFLU (Lin)	$CL = \theta_1 + (\theta_2 \times RFLU-1300)$	NA[b]	
	RFLU (Pow)	$CL = \theta_1 \times (RFLU/1300)^{\theta 2}$	NA[a]	
	RFLU(Exp)	$CL = \theta_1 e^{(\theta 2 \times RFLU/1300)}$	NA[a]	
	VASO (Frac)	$CL = \theta_1 \times (1+(\theta_2 \times VASO))$	1837.316	+26.207
	VENT (Frac)	$CL = \theta_1 \times (1+(\theta_2 \times VENT))$	1842.134	+31.025

Table 8 Change in OFV during forward addition step 1

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model			1811.109	
V_1	TBW (Lin)	$V_1 = \theta_1 + (\theta_2 \times TBW-57)$	1802.252	-8.857*
	TBW (Pow)	$V_1 = \theta_1 \times (TBW/57)^{\theta 2}$	1834.077	+22.968
	TBW (Exp)	$V_1 = \theta_1 e^{(\theta 2 \times TBW/57)}$	1833.301	+22.192
	ABW (Lin)	$V_1 = \theta_1 + (\theta_2 \times ABW-56)$	1800.391	-10.718*
	ABW (Pow)	$V_1 = \theta_1 \times (ABW/56)^{\theta 2}$	1799.782	-11.327*
	ABW (Exp)	$V_1 = \theta_1 e^{(\theta 2 \times ABW/56)}$	1799.017	-12.092*
	RFLU (Lin)	$V_1 = \theta_1 + (\theta_2 \times RFLU)$	NA[a]	
	RFLU (Pow)	$V_1 = \theta_1 \times (RFLU/1300)^{\theta 2}$	NA[a]	
	RFLU(Exp)	$V_1 = \theta_1 e^{(\theta 2 \times RFLU)}$	1841.041	+29.932
	VASO (Frac)	$V_1 = \theta_1 \times (1+(\theta_2 \times VASO))$	1838.388	+27.279
	VENT (Frac)	$V_1 = \theta_1 \times (1+(\theta_2 \times VENT))$	1841.553	+30.444

[a] The model could not estimate parameters at the initial evaluation. [b] The model could not throughout estimate parameters (minimization terminated). *A decrease in OFV ≥ 3.84 indicates that the covariate has a significant effect on the PK parameter (p-value ≤ 0.05). Lin, Linear model; Pow, Power model; Exp, Exponential model; Frac, Fractional change model.

Table 9 Change in OFV during forward addition step 2

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model added with CL_{Cr}		$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$	1785.696	
CL	TBW (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times TBW\text{-}57)$	NA[a]	
	TBW (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times (TBW/57)^{\theta 3}$	1783.981	-1.715
	TBW (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times e^{(\theta 3 \times TBW/57)}$	1784.519	-1.177
	ABW (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times ABW\text{-}56)$	NA[a]	
	ABW (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times (ABW/56)^{\theta 3}$	1783.969	-1.727
	ABW (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times e^{(\theta 3 \times ABW/56)}$	1784.440	-1.256
	MAP (Lin)	**$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$**	**1769.331**	**-16.365***
	MAP (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times (MAP/68)^{\theta 3}$	1770.163	-15.533*
	MAP (Exp)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) \times e^{(\theta 3 \times MAP/68)}$	1769.885	-15.811*
	TBIL (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times TBIL\text{-}0.8)$	1784.906	-0.790
	TBIL (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times (TBIL/0.8)^{\theta 3}$	1785.229	-0.467
	TBIL (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) \times e^{(\theta 3 \times TBIL/0.8)}$	1784.495	-1.201
	RFLU (Lin)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) + (\theta_3 \times RFLU\text{-}1300)$	NA[a]	
	RFLU (Pow)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) \times (RFLU/1300)^{\theta 3}$	NA[a]	
	RFLU(Exp)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) \times e^{(\theta 3 \times RFLU/1300)}$	1784.715	-0.981
	VASO (Frac)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) \times (1+(\theta_3 \times VASO))$	1785.701[b]	+0.005
	VENT (Frac)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55) \times (1+(\theta_3 \times VENT))$	1785.697[b]	+0.001

Table 9 Change in OFV during forward addition step 2

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model added with CL_{Cr}		$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$	1785.696	
V_1	TBW (Lin)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 + (\theta_4 \times TBW\text{-}57)$	1777.248	-8.448*
	TBW (Pow)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 \times (TBW/57)^{\theta 4}$	1777.111	-8.585*
	TBW (Exp)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 e^{(\theta 4 \times TBW/57)}$	1776.162	-9.534*
	ABW (Lin)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 + (\theta_4 \times ABW\text{-}56)$	1775.148	-10.548*
	ABW (Pow)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 \times (ABW/56)^{\theta 4}$	1774.597	-11.099*
	ABW (Exp)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 e^{(\theta 4 \times ABW/56)}$	1773.942	-11.754*
	RFLU (Lin)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 + (\theta_4 \times RFLU)$	NA[a]	
	RFLU (Pow)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 \times (RFLU/1300)^{\theta 4}$	NA[a]	
	RFLU(Exp)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 e^{(\theta 4 \times RFLU)}$	1784.481	-1.215
	VASO (Frac)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 \times (1+(\theta_4 \times VASO))$	1785.698[b]	+0.002
	VENT (Frac)	$CL = \theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)$ $V_1 = \theta_3 \times (1+(\theta_4 \times VENT))$	1785.697[b]	+0.001

[a] The model could not estimate parameters at the initial evaluation. [b] The model could not throughout estimate parameters. *A decrease in OFV ≥ 3.84 indicates that the covariate has a significant effect on the PK parameter (p-value ≤ 0.05). Lin, Linear model; Pow, Power model; Exp, Exponential model; Frac, Fractional change model.

Table 10 Change in OFV during forward addition step 3

Parameters	Added covariates (Patterns)	Models	OFV	ΔOFV
Base model added with 2 linear relations: CL_{Cr} and CL, MAP and CL		$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$	1769.331	
CL	TBW (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times TBW\text{-}57)$	1768.440	-0.891
	TBW (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (TBW/57)^{\theta 4}$	1768.476	-0.855
	TBW (Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times TBW/57)}$	1768.936	-0.395
	ABW (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times ABW\text{-}56)$	1768.842	-0.489
	ABW (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (ABW/56)^{\theta 4}$	1768.910	-0.421
	ABW (Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times ABW/56)}$	1769.207	-0.124
	TBIL (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times TBIL\text{-}0.8)$	1768.264	-1.067
	TBIL (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (TBIL/0.8)^{\theta 4}$	1768.375	-0.956
	TBIL (Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times TBIL/0.8)}$	1768.041	-1.290
	RFLU (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times RFLU\text{-}1300)$	NA[a]	
	RFLU (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (RFLU/1300)^{\theta 4}$	NA[a]	
	RFLU(Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times RFLU/1300)}$	1767.342	-1.989

Table 10 Change in OFV during forward addition step 3

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model added with 2 linear relations: CL_{Cr} and CL, MAP and CL		$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$	1769.331	
Cl	VASO (Frac)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (1+(\theta_4 \times VASO))$	1769.335 [b]	+0.004
	VENT (Frac)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (1+(\theta_4 \times VENT))$	1769.333[b]	+0.002
V_1	TBW (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 + (\theta_5 \times TBW\text{-}57)$	1761.073	-8.258*
	TBW (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 \times (TBW/57)^{\theta 5}$	1760.954	-8.377*
	TBW (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 e^{(\theta 5 \times TBW/57)}$	1760.062	-9.269*
	ABW (Lin)	**$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$** **$V_1 = \theta_4 + (\theta_5 \times ABW\text{-}56)$**	**1759.001**	**-10.330***
	ABW (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 \times (ABW/56)^{\theta 5}$	1758.496	-10.835*
	ABW (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 e^{(\theta 5 \times ABW/56)}$	1757.903	-11.428*
	RFLU (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 + (\theta_5 \times RFLU)$	NA[a]	
	RFLU (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 \times (RFLU/1300)^{\theta 5}$	NA[a]	
	RFLU(Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 e^{(\theta 5 \times RFLU)}$	1768.302	-1.029

Table 10 Change in OFV during forward addition step 3

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
V_1	VASO (Frac)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 \times (1+(\theta_5 \times VASO))$	1769.333 [b]	+0.002
	VENT (Frac)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 \times (1+(\theta_5 \times VENT))$	1769.331[b]	0.000

[a] The model could not estimate parameters at the initial evaluation. [b] The model could not throughout estimate parameters. *A decrease in OFV ≥ 3.84 indicates that the covariate has a significant effect on the PK parameter (p-value ≤ 0.05). Lin, Linear model; Pow, Power model; Exp, Exponential model; Frac, Fractional change model.

Table 11 Change in OFV during forward addition step 4

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model added with 3 linear relations: CL_{Cr} and CL, MAP and CL, ABW and V1		$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 + (\theta_5 \times ABW\text{-}56)$	1759.001	
CL	TBW (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times TBW\text{-}57)$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.021	-0.980
	TBW (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (TBW/57)^{\theta 4}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.010	-0.991
	TBW (Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times TBW/57)}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.547	-0.454
	ABW (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times ABW\text{-}56)$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.384	-0.617
	ABW (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (ABW/56)^{\theta 4}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.451	-0.550
	ABW (Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times ABW/56)}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1758.816	-0.185
	TBIL (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68) + (\theta_4 \times TBIL\text{-}0.8)$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1757.391	-1.610
	TBIL (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68) \times (TBIL/0.8)^{\theta 4}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1757.417	-1.584
	TBIL (Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68) \times e^{(\theta 4 \times TBIL/0.8)}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1757.207	-1.794

Table 11 Change in OFV during forward addition step 4

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Base model added with 3 linear relations: CL_{Cr} and CL, MAP and CL, ABW and V1		$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 + (\theta_5 \times ABW\text{-}56)$	1759.001	
CL	RFLU (Lin)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) + (\theta_4 \times RFLU\text{-}1300)$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	NA[a]	
	RFLU (Pow)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (RFLU/1300)^{\theta 4}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	NA[a]	
	RFLU(Exp)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times e^{(\theta 4 \times RFLU/1300)}$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1757.104	-1.897
	VASO (Frac)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (1+(\theta_4 \times VASO))$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1759.006[b]	+0.005
	VENT (Frac)	$CL = ((\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)) \times (1+(\theta_4 \times VENT))$ $V_1 = \theta_5 + (\theta_6 \times ABW\text{-}56)$	1759.003[b]	+0.002
V1	RFLU (Lin)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = (\theta_4 + (\theta_5 \times ABW\text{-}56)) + (\theta_6 \times RFLU\text{-}1300)$	NA[a]	
	RFLU (Pow)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = (\theta_4 + (\theta_5 \times ABW\text{-}56)) \times (RFLU/1300)^{\theta 6}$	NA[a]	
	RFLU(Exp)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = (\theta_4 + (\theta_5 \times ABW\text{-}56)) \times e^{(\theta 6 \times RFLU/1300)}$	1757.874	-1.127

Table 11 Change in OFV during forward addition step 4

Parameters	**Added covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
	VASO (Frac)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = (\theta_4 + (\theta_5 \times ABW\text{-}56)) \times (1+(\theta_6 \times VASO))$	1759.002[b]	+0.001
	VENT (Frac)	$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = (\theta_4 + (\theta_5 \times ABW\text{-}56)) \times (1+(\theta_6 \times VENT))$	1759.002[b]	0.000

[a] The model could not estimate parameters at the initial evaluation. [b] The model could not throughout estimate parameters. Lin, Linear model; Pow, Power model; Exp, Exponential model; Frac, Fractional change model.

Table 12 Change in OFV during backward deletion of the full model

Parameters	**Removed covariates (Patterns)**	**Models**	**OFV**	**ΔOFV**
Full model		$CL = (\theta_1 + (\theta_2 \times CL_{Cr}\text{-}55)) + (\theta_3 \times MAP\text{-}68)$ $V_1 = \theta_4 + (\theta_5 \times ABW\text{-}56)$	1759.001	
CL	CL_{Cr} (Lin)	$CL = \theta_1 + (\theta_2 \times (MAP\text{-}68))$ $V_1 = \theta_3 + (\theta_4 \times (ABW\text{-}56))$	1793.132	34.131*
CL	MAP (Lin)	$CL = \theta_1 + (\theta_2 \times (CL_{Cr}\text{-}55))$ $V_1 = \theta_3 + (\theta_4 \times (ABW\text{-}56))$	1775.148	16.147*
V1	ABW (Lin)	$CL = (\theta_1 + (\theta_2 \times (CL_{Cr}\text{-}55))) + (\theta_3 \times MAP\text{-}68)$	1769.331	10.330*

*A decrease in OFV ≥ 6.64 indicates that the covariate has a significant effect on the PK parameter (p-value ≤ 0.01). Lin, Linear model.

Table 13 Population PK parameter estimates of the base model, final model and bootstrap

	Base model	**Final model**	**Bootstrap**		
Parameters	**Estimates**	**Estimates**	**Estimates**	**95% CI**	
CL (L/h)	5.65	5.37	5.34	4.83	5.87
V_1 (L)	9.59	9.35	9.34	6.69	12.20
V_2 (L)	7.55	7.77	7.78	5.06	11.30
Q (L/h)	20.10	21.30	22.31	6.00	47.50
IIV of CL(CV%)	46.3	28.5	27.5	21.1	33.2
IIV of V_1 (CV%)	64.0	55.4	55.3	36.3	73.3
RV (CV%)	22.1	22.3	22.1	17.4	27.1

CI, confidence interval; CV, coefficient of variation.

4.2.3. Model evaluation

The goodness-of-fit plots proved the correctness of the final model, as shown in figure 9. The final model could provide predicted concentrations which corresponded to observed concentrations (data points were scattered around the identity line), as presented in figure 9a and b. In addition, weighted residuals (the differences between observed and predicted concentrations) were scattered around the horizontal zero line and were within ±1 along with all predicted concentrations and time, as shown in figure 9c and d. The goodness-of-fit plots confirmed that the final model was correctly specified. Moreover, the evaluation based on the simulation; bootstrap and visual predictive check (VPC) were performed. The results of 1,000 bootstrap runs (the final model was implemented with 1,000 modified data files) are shown in table 13. All PK parameter estimates were similar to bootstrap estimates and within 95%CI of bootstrap runs, this finding confirmed that the final model provided accurate and precise PK parameter estimates. In addition, the VPC showed that the observed percentiles remained within the 95% CI of corresponding predicted percentiles, as displayed in figure 10. These results confirmed that the final model had the sufficient performance to further simulate concentration-time profiles.

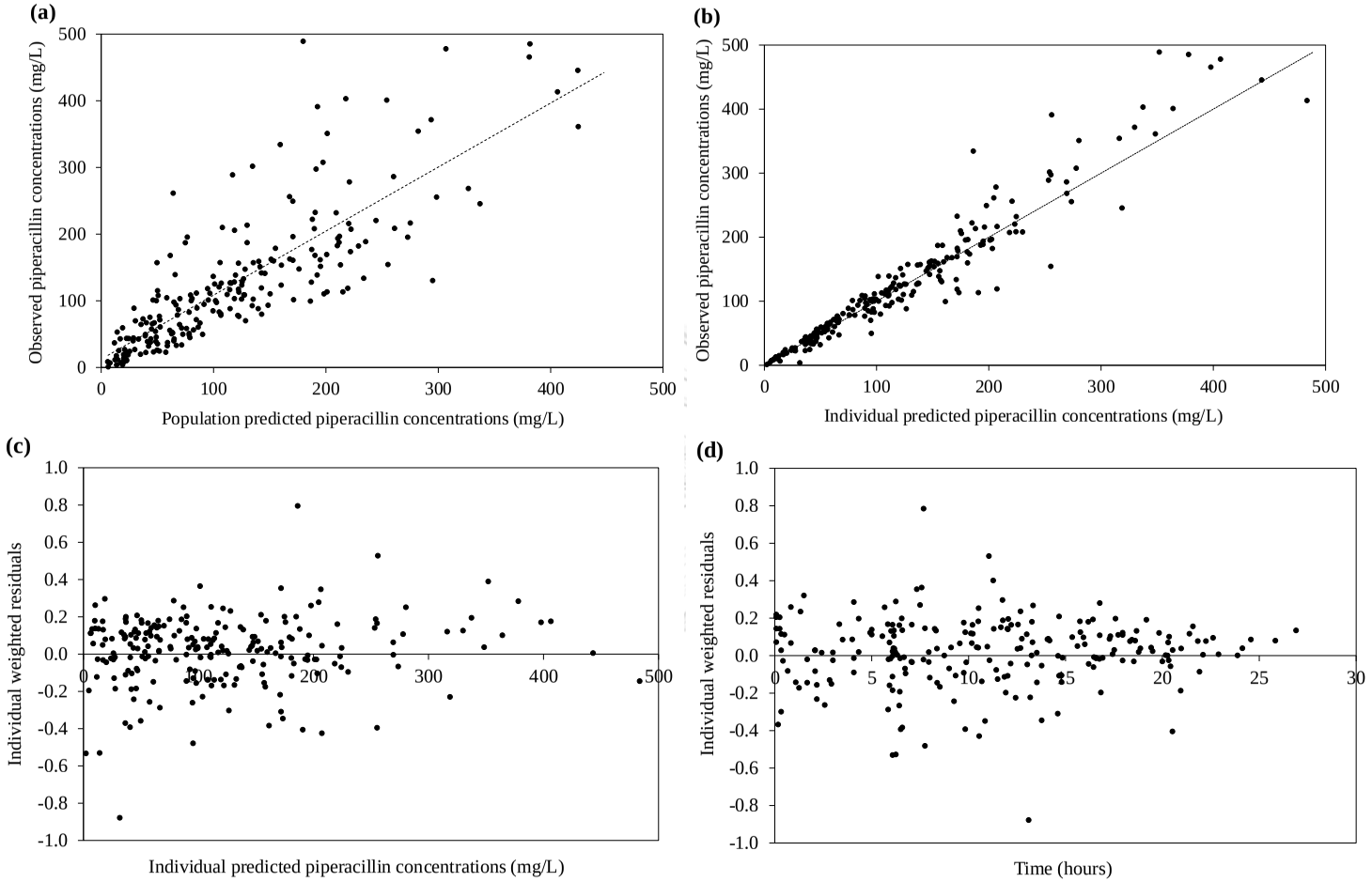

Figure 9 The goodness-of-fit plots of the final model.
(a) Observed versus population predicted piperacillin concentrations; (b) Observed versus individual predicted piperacillin concentrations; (c) Individual weighted residuals versus individual predicted piperacillin concentrations; (d) Individual weighted residuals versus time. Dotted lines, identity lines.

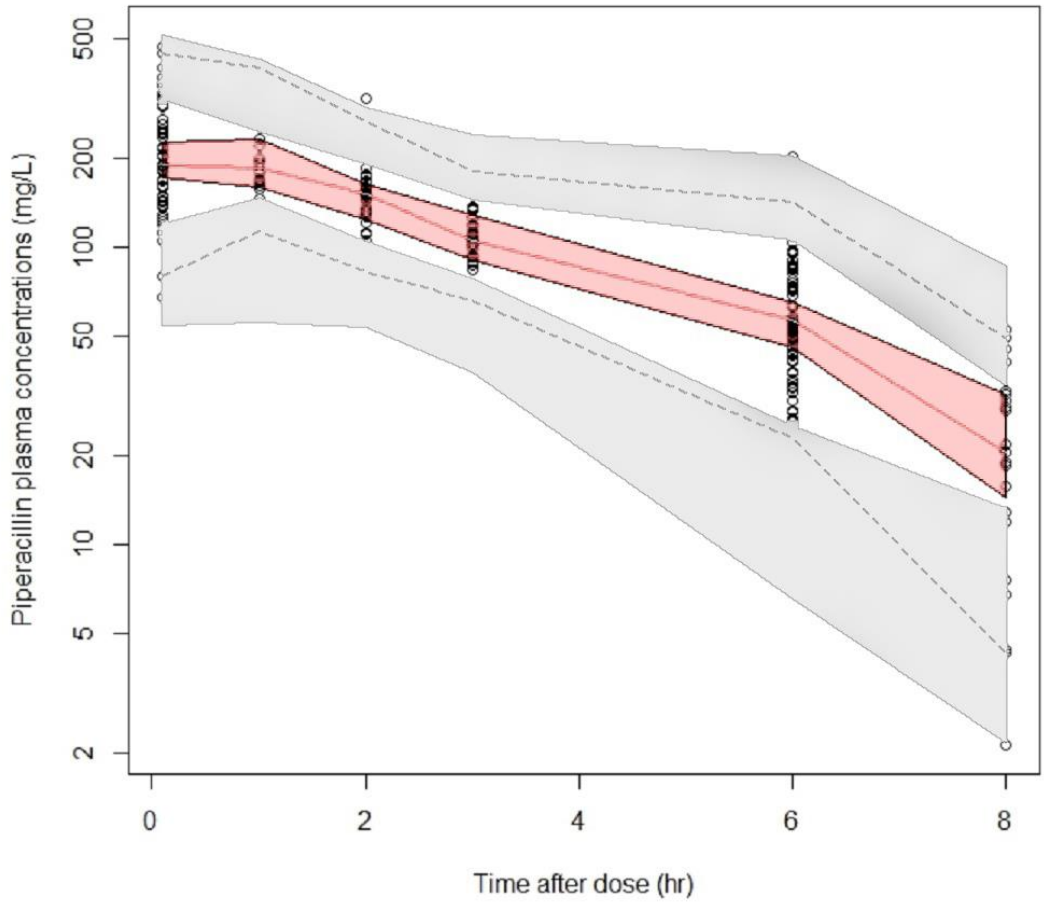

Figure 10 Visual predictive check of the final model.
Individual points represent observed data. Dashed lines represent 5th, 50th and 95th percentiles of observed data. The red area represents 95% CI of the 50th percentile of predicted data. The black areas represent 95% CI of the 5th and 95th percentiles of predicted data.

4.3. Pharmacodynamic assessment using Monte Carlo simulation (MCS)

4.3.1. Probability of target attainment (PTA)

All subsequent Monte Carlo simulations were based on the validated final model. Population PK parameter estimates and variabilities were used to simulate 10,000 virtual patients for each dosage regimens and renal functions. PTA (PK/PD target: 90% $fT_{>MIC}$) at various MIC when using 40 different dosage regimens in patients with 4 different renal function groups are shown in figure 11 to 14 and table 14.

Regarding patients with CL_{Cr} 60 to 120 mL/min and pathogens with MIC 16 mg/L (for the highest MIC of susceptible pathogens), there were 9 dosage regimens which provided ≥ 90% PTA; piperacillin 6 g continuous infusion (CI) with loading dose (LD) 2 g or 4 g *(6 g/day with LD)*, 8 g CI with LD 2 or 4 g *(8 g/day with LD)*, 12 g CI without or with LD 2 or 4 g *(12 mg/day)*, and 4 g every 6 hours by infusion over 4 hours and 16 g CI *(16 mg/day)*, as shown in figure 11c, 11d, 11e and 11f, respectively.

Concerning patients with CL_{Cr} 40 to 60 mL/min and pathogens with MIC 16 mg/L, 11 dosage regimens could provide ≥ 90% PTA; piperacillin 6 g CI with LD 2 g or 4 g *(6 g/day with LD)*, 8 g CI with LD 2 or 4 g *(8 g/day with LD)*, 12 g CI without or with LD 2 or 4 g *(12 mg/day)*, and 4 g every 6 hours by infusion over 2, 3, or 4 hours and 16 g CI *(16 mg/day)*, as presented in figure 12c, 12d, 12e and 12f, respectively.

As regards patients with CL_{Cr} 20 to 40 mL/min and pathogens with MIC 16 mg/L, there were 19 dosage regimens which provided ≥ 90%; piperacillin 6 g CI with LD 2 g or 4 g *(6 g/day with LD)*, 2 g every 6 hours by all infusion with LD 4 g and 8 g CI with LD 2 or 4 g *(8 g/day with LD)*, 4 g every 8 hours by infusion over 4 hours and 12 g CI without or with LD 2 or 4 g *(12 mg/day)*, and 4 g every 6 hours by all infusion time and 16 g CI *(16 mg/day)*, as shown in figure 13c, 13d, 13e and 13f, respectively.

For patients with CL_{Cr} < 20 mL/min and pathogens with MIC 16 mg/L, 22 dosage regimens could provide ≥ 90%; piperacillin 2 g every 6 hours by infusion over 2 or 3 hours *(8 g/day without LD)*, 6 g CI with LD 2 g or 4 g *(6 g/day with LD)*, 2 g every 6 hours by all infusion with LD 4 g and 8 g CI with LD 2 or 4 g

(8 g/day with LD), 4 g every 8 hours by infusion over 3 or 4 hours and 12 g CI without or with LD 2 or 4 g *(12 mg/day)*, and 4 g every 6 hours by all infusion time and 16 g CI *(16 mg/day)*, as presented in figure 14b, 14c, 14d, 14e and 14f, respectively.

4.3.2. Cumulative fraction of response (CFR)

CFR of 3 major pathogens including *P. aeruginosa, K. pneumoniae, E. coli* when using 40 dosage regimens in patients with 4 different renal function groups are described in table 15 to 18.

Concerning patients with 60 to 120 mL/min, there was no dosage regimen which provided CFR ≥ 90% for the *P. aeruginosa* infection. For the *K. pneumoniae* infection, piperacillin 12 g CI with LD 2 g or 4 g provided CFR of 90%. Contrarily, there were several dosage regimens to achieve CFR ≥ 90% for the *E. coli* infection; for example, 4 g every 8 hours by infusion over 3 or 4 hours or 12 g CI without/with LD (12 g/day), 4 g every 6 hours by infusion over 0.5, 1, 2, 3, 4 hours or 16 g CI (16 g/day), as shown in table 15.

When considering patients with 40 to 60 mL/min, there was no dosage regimen which provided CFR ≥ 90% for the *P. aeruginosa* infection. For the suspected *K. pneumoniae* infection, piperacillin 8 g or 12 g CI with LD 2 g or 4 g provided CFR ≥ 90%. Inversely, almost all dosage regimens achieved CFR ≥ 90% for the suspected *E. coli* infection, excluding piperacillin 2 g every 8 hours by infusion over 0.5, 1, 2 hours without LD, as presented in table 16.

In case of patients with 20 to 40 mL/min, there was no dosage regimen which provided CFR ≥ 90% for the *P. aeruginosa* infection. As for the suspected *K. pneumoniae* infection, piperacillin 6, 8 or 12 g CI with LD 2 g or 4 g could reach CFR ≥ 90%. Almost all dosage regimens could achieve CFR ≥ 90%, excluding piperacillin 2 g every 8 hours by infusion 0.5 or 1 hours without LD for the *E. coli* infection, as shown in table 17.

Regarding patients with < 20 mL/min, there was no dosage regimen which provided CFR ≥ 90% for the *P. aeruginosa* infection. In part of the *K. pneumoniae* infection, piperacillin 6, 8, or 12 g CI with LD 2 g or 4 g could achieve CFR ≥ 90. All dosage regimens could reach CFR ≥ 90% for the *E. coli* infection, as presented in table 18.

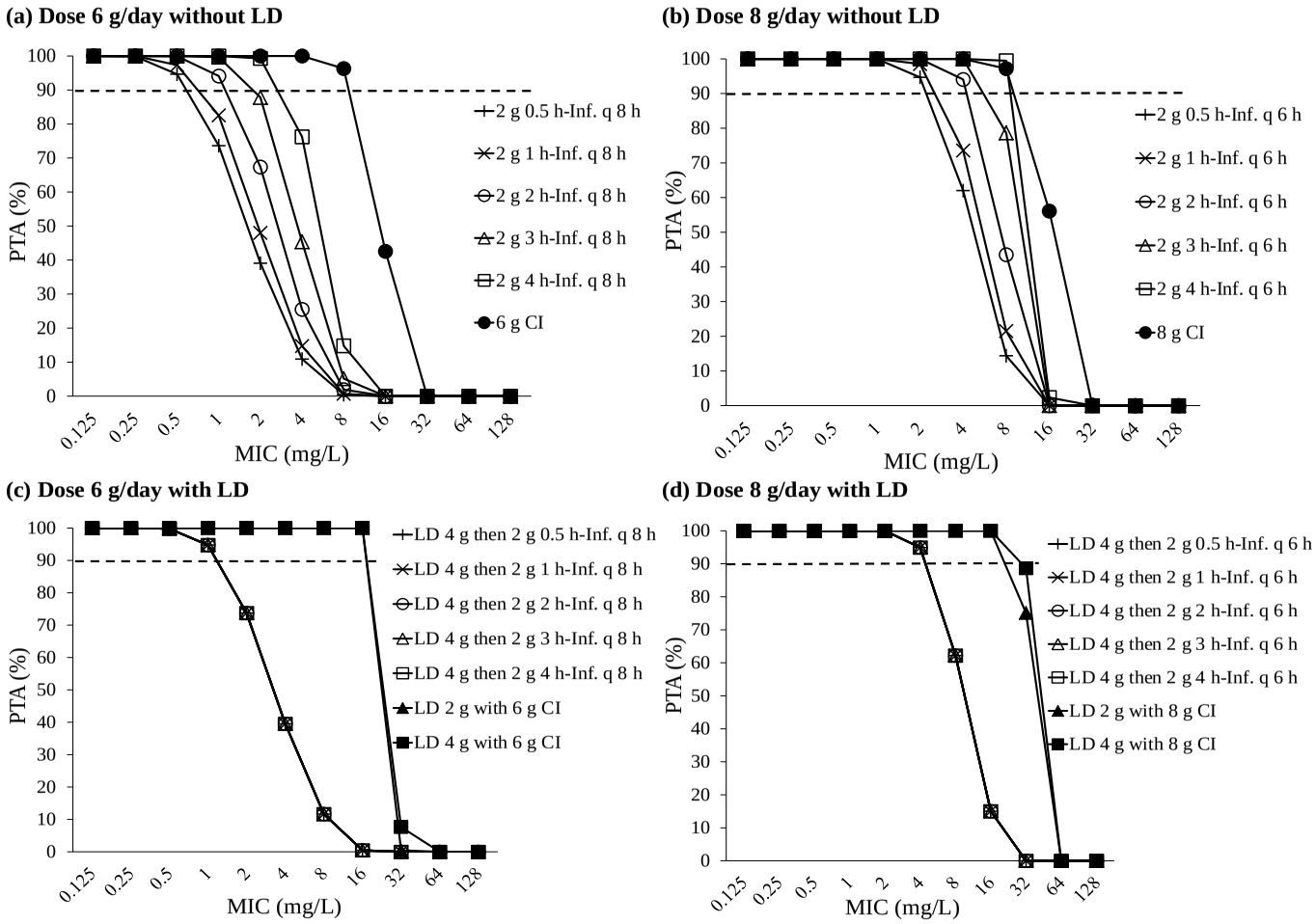

Figure 11 PTA versus MIC profiles in patients with CL_{Cr} 60 to 120 mL/min.

The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h.

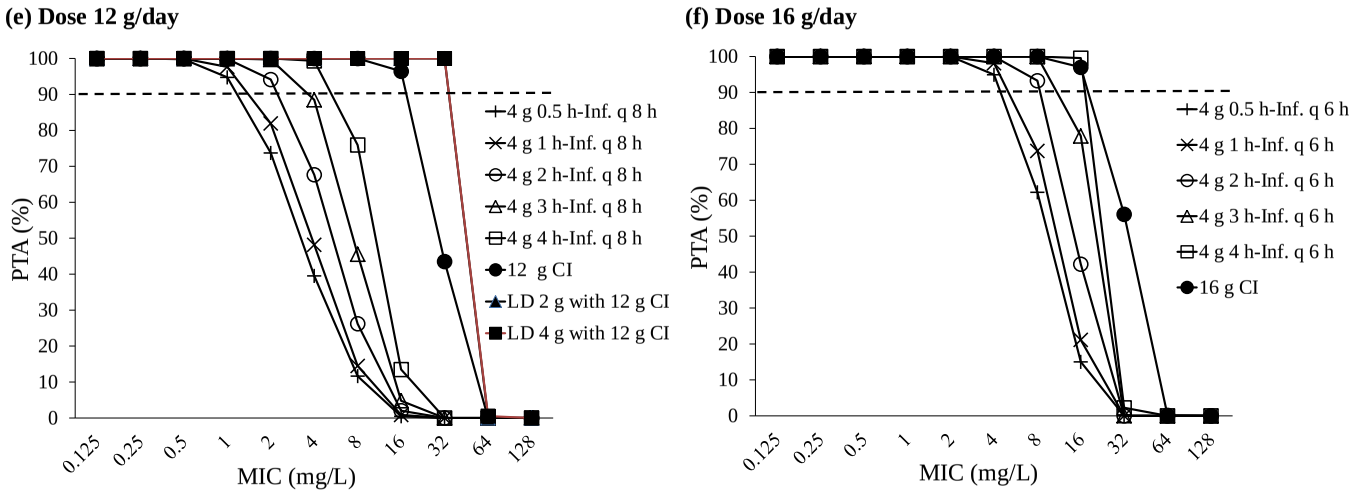

Figure 11 PTA versus MIC profiles in patients with CL_{Cr} 60 to 120 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h. (continue)

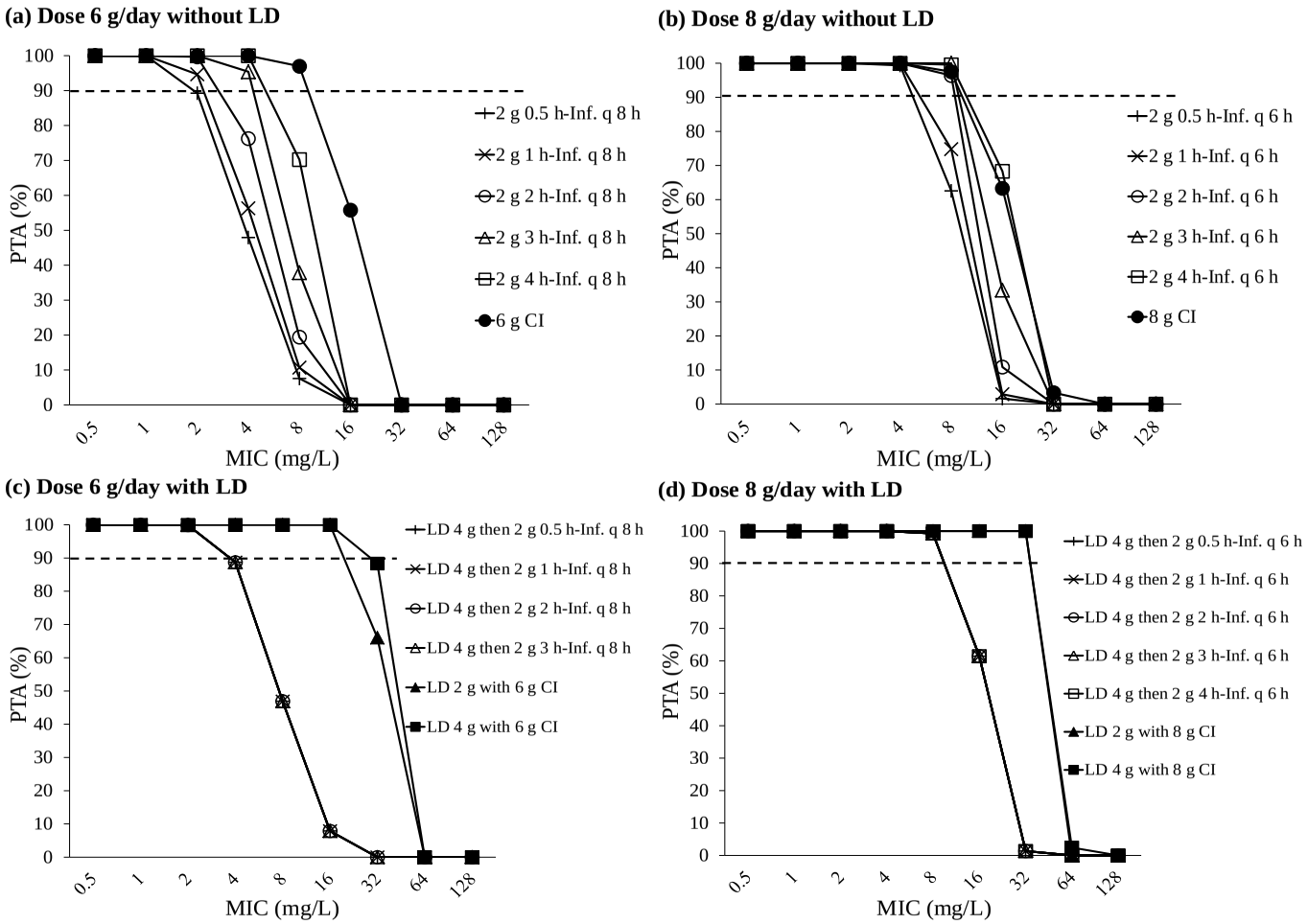

Figure 12 PTA versus MIC profiles in patients with CL_{Cr} 40 to 60 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h.

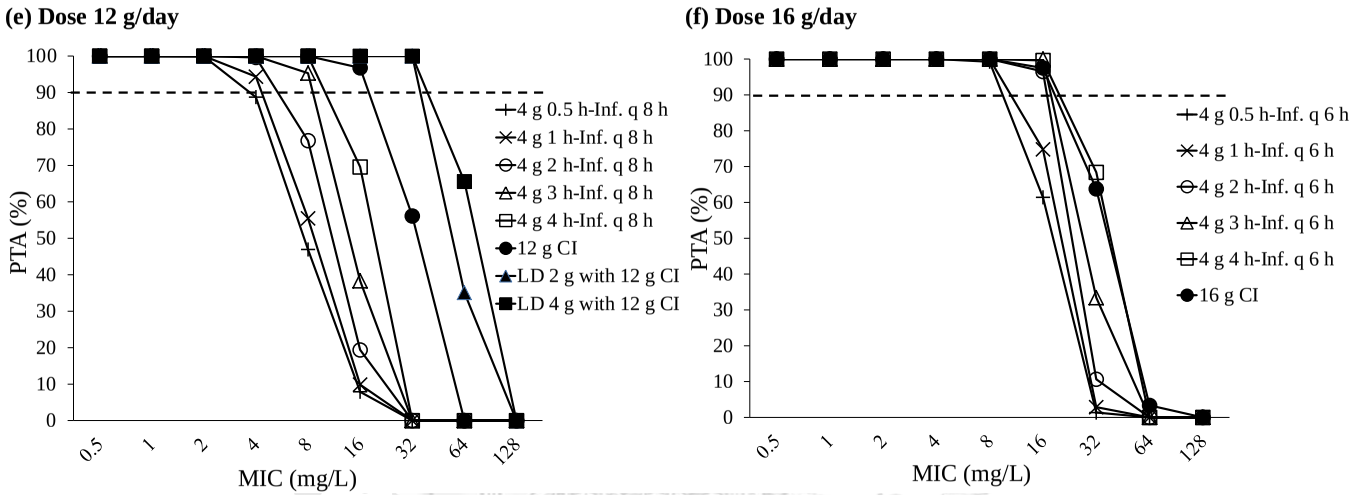

Figure 12 PTA versus MIC profiles in patients with CLCr 40 to 60 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h. (continue)

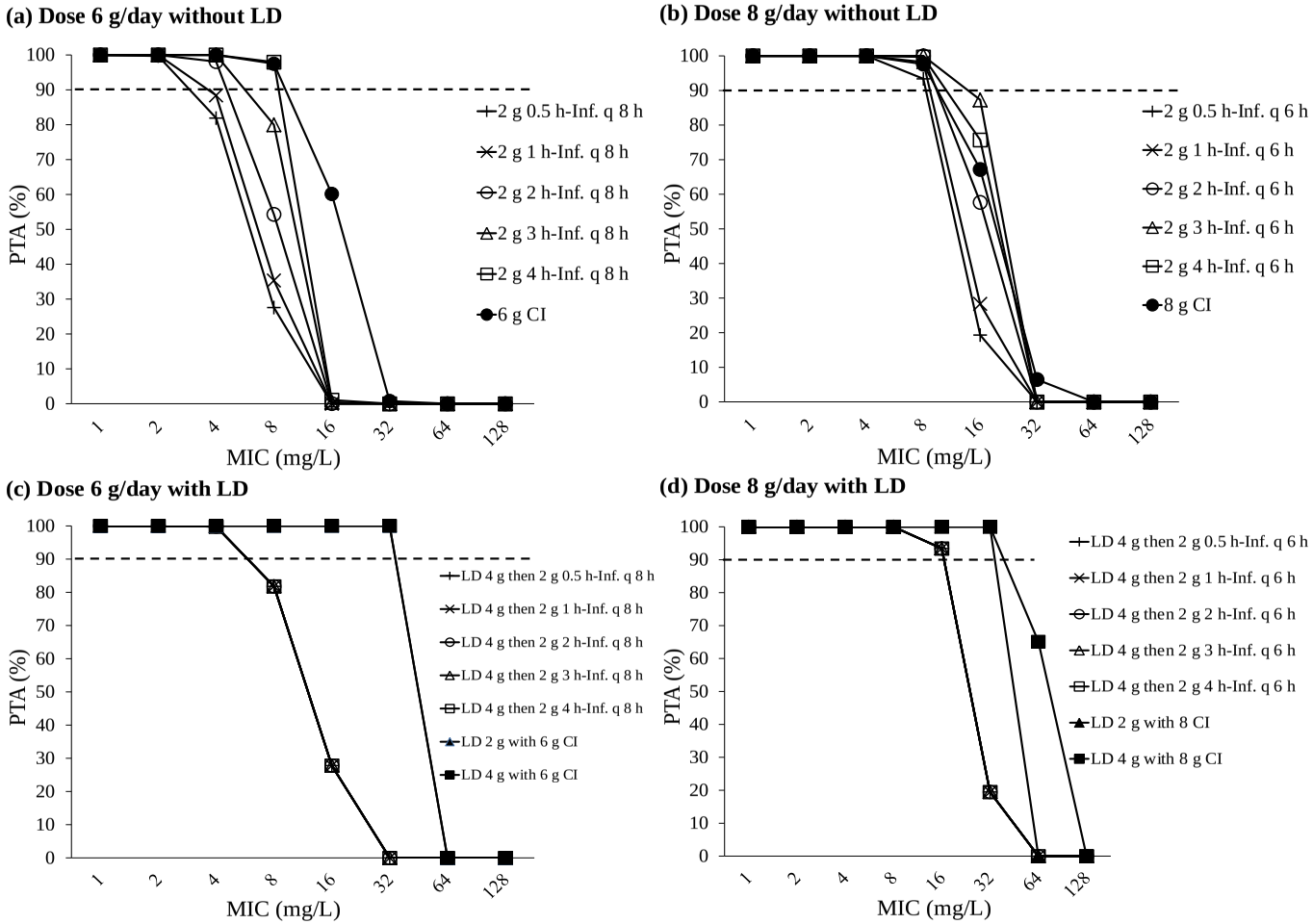

Figure 13 PTA versus MIC profiles in patients with CL_{Cr} 20 to 40 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h.

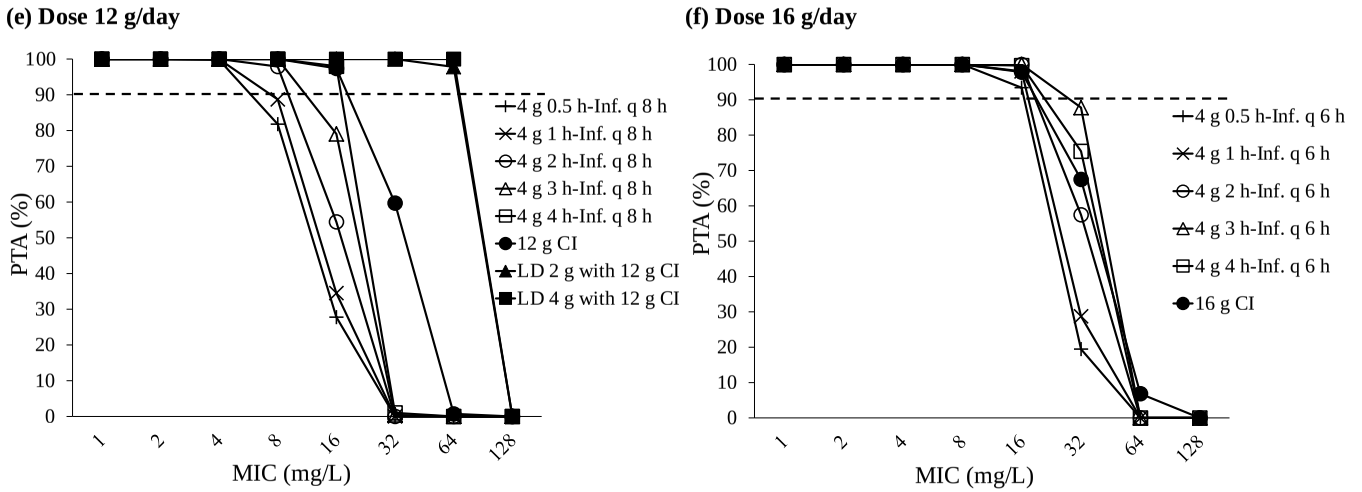

Figure 13 PTA versus MIC profiles in patients with CL_{Cr} 20 to 40 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h. (continue)

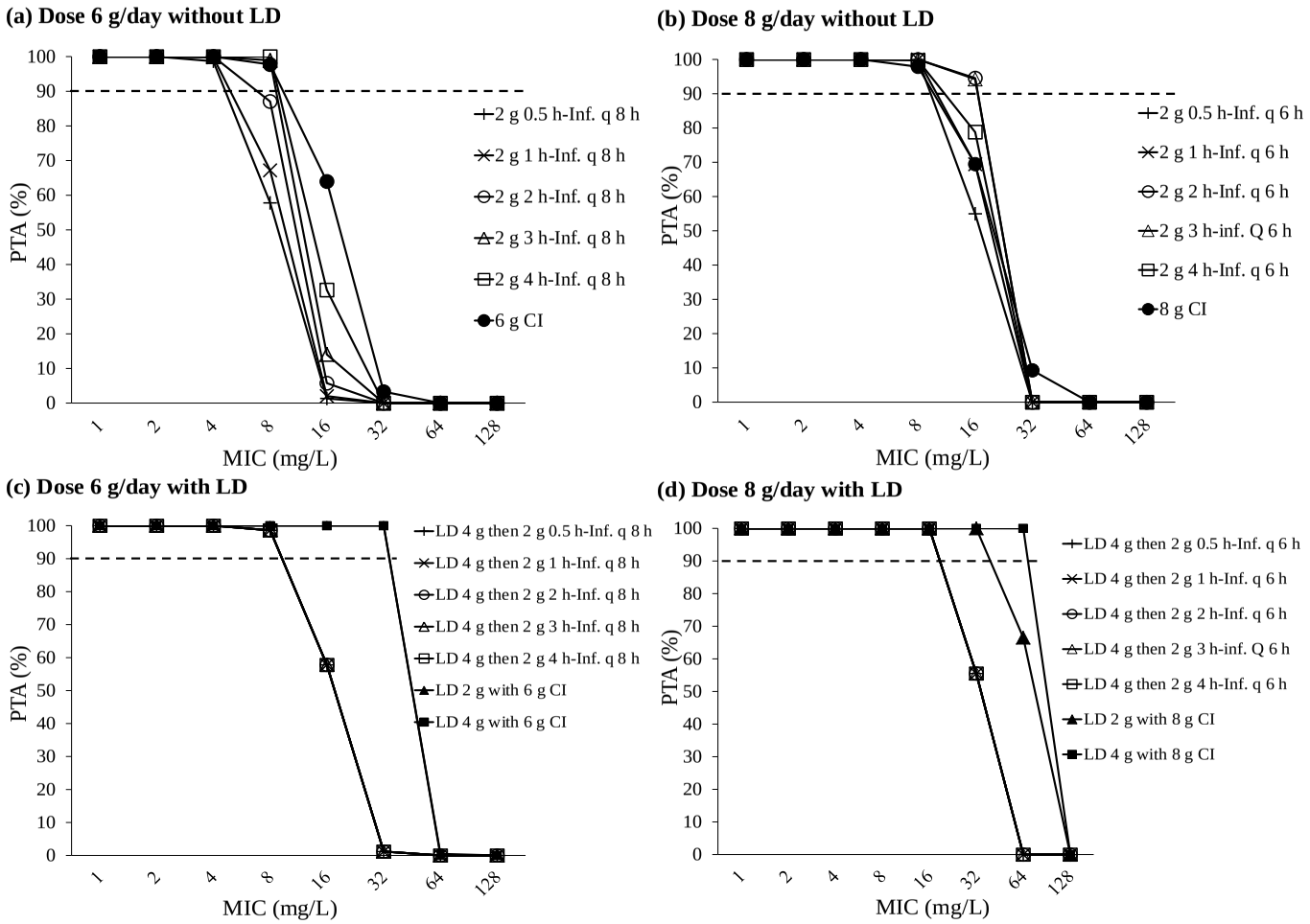

Figure 14 PTA versus MIC profiles in patients with CL_{Cr} < 20 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h.

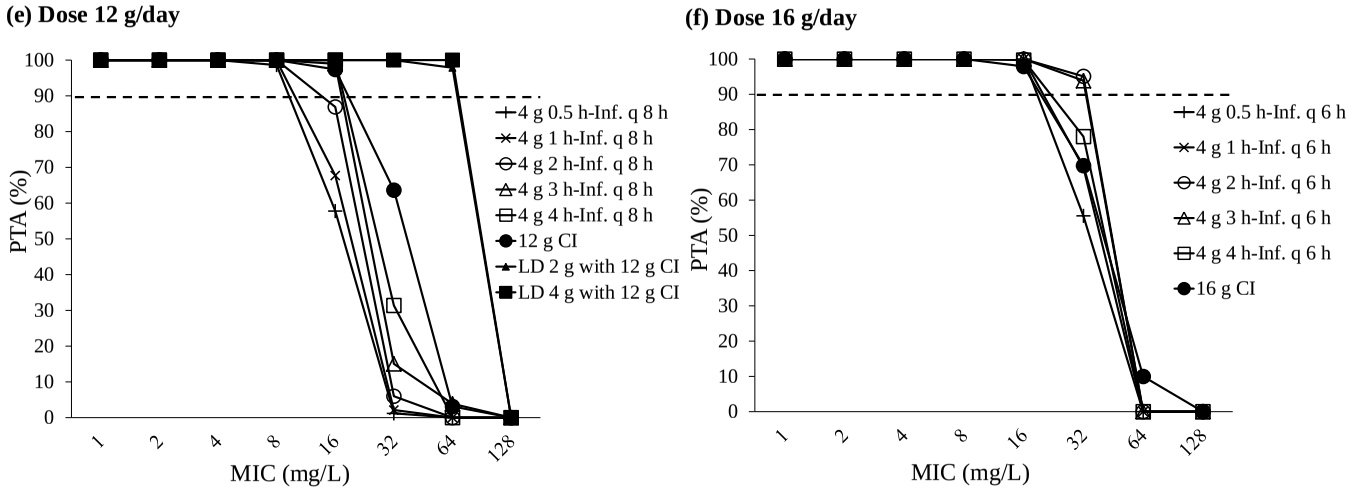

Figure 14 PTA versus MIC profiles in patients with CL_{Cr} < 20 mL/min.
The graphs present the total dose/day of (a) 6 g without LD, (b) 8 g without LD, (c) 6 g with LD, (d) 8 g with LD, (e) 12 g, (f) 16 g. Dashed lines represent 90% of the virtual patients reached the 90%. $fT_{>MIC}$, LD was added by infusion over 0.5 h (continue).

Table 14 PTA with the following CL_{Cr} and MIC of various dosage regimens

Dosage regimens	**MIC (mg/L)**	**PTA (%) with the following CL_{Cr} (mL/min)**			
		60-120	**40-60**	**20-40**	**<20**
Dose 6 g/day without LD					
2 g 0.5 h-inf. q 8 h	0.5	95	100	100	100
	1	74	100	100	100
	2	39	89	100	100
	4	11	48	82	99
	8	0	8	28	58
	16	0	0	0	1
2 g 1 h-inf. q 8 h	0.5	97	100	100	100
	1	83	100	100	100
	2	48	95	100	100
	4	15	56	88	100
	8	1	11	35	67
	16	0	0	0	2
2 g 2 h-inf. q 8 h	0.5	100	100	100	100
	1	94	100	100	100
	2	67	100	100	100
	4	26	76	98	100
	8	2	19	54	87
	16	0	0	0	6
2 g 3 h-inf. q 8 h	0.5	100	100	100	100
	1	100	100	100	100
	2	88	100	100	100
	4	45	95	100	100
	8	5	38	80	99
	16	0	0	0	14
2 g 4 h-inf. q 8 h	0.5	100	100	100	100
	1	100	100	100	100
	2	99	100	100	100
	4	76	100	100	100
	8	15	70	98	100
	16	0	0	1	33
6 g CI	0.5	100	100	100	100
	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	96	97	97	98
	16	43	56	60	64

Table 14 PTA with the following CL_{Cr} and MIC of various dosage regimens (continue)

Dosage regimens	**MIC (mg/L)**	**PTA (%) with the following CL_{Cr} (mL/min)**			
		60-120	**40-60**	**20-40**	**<20**
Dose 6 g/day with LD					
LD 4 g then 2 g q 8 h	1	95	100	100	100
(All infusion time)	2	74	100	100	100
	4	40	89	100	100
	8	12	47	82	99
	16	0	8	28	58
LD 2 g with 6 g CI	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
LD 4 g with 6 g CI	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
Dose 8 g/day without LD					
2 g 0.5 h-inf. q 6 h	2	95	100	100	100
	4	62	99	100	100
	8	14	63	93	100
	16	0	2	19	55
2 g 1 h-inf. q 6 h	2	99	100	100	100
	4	74	100	100	100
	8	22	75	98	100
	16	0	3	28	69
2 g 2 h-inf. q 6 h	2	100	100	100	100
	4	94	100	100	100
	8	44	96	100	100
	16	0	11	58	95

Table 14 PTA with the following CL_{Cr} and MIC of various dosage regimens (continue)

Dosage regimens	**MIC (mg/L)**	**PTA (%) with the following CL_{Cr} (mL/min)**			
		60-120	**40-60**	**20-40**	**<20**
Dose 8 g/day without LD					
2 g 3 h-inf. q 6 h	2	100	100	100	100
	4	100	100	100	100
	8	79	100	100	100
	16	0	33	87	94
2 g 4 h-inf. q 6 h	2	100	100	100	100
	4	100	100	100	100
	8	99	100	100	100
	16	2	68	76	79
8 g CI	2	100	100	100	100
	4	100	100	100	100
	8	97	98	98	98
	16	56	63	67	69
Dose 8 g/day with LD					
LD 4 g then 2 g q 6 h	4	95	100	100	100
(All infusion times)	8	62	99	100	100
	16	15	61	93	100
LD 2 g with 8 g CI	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
LD 4 g with 8 g CI	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100

Table 14 PTA with the following CL_{Cr} and MIC of various dosage regimens (continue)

Dosage regimens	**MIC**	**PTA (%) with the following CL_{Cr} (mL/min)**			
		60-120	**40-60**	**20-40**	**<20**
Dose 12 g/day					
4 g 0.5 h-inf. q 8 h	1	95	100	100	100
	2	74	100	100	100
	4	40	89	100	100
	8	12	47	82	99
	16	0	8	28	58
4 g 1 h-inf. q 8 h	1	98	100	100	100
	2	82	100	100	100
	4	48	94	100	100
	8	14	55	89	100
	16	1	10	35	68
4 g 2 h-inf. q 8 h	1	100	100	100	100
	2	94	100	100	100
	4	68	100	100	100
	8	26	77	98	100
	16	2	19	54	87
4 g 3 h-inf. q 8 h	1	100	100	100	100
	2	100	100	100	100
	4	89	100	100	100
	8	46	95	100	100
	16	5	38	79	99
4 g 4 h-inf. q 8 h	1	100	100	100	100
	2	100	100	100	100
	4	99	100	100	100
	8	76	100	100	100
	16	13	70	98	100
12 g CI	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	100	100	100	100
	16	96	97	97	97

Table 14 PTA with the following CL_{Cr} and MIC of various dosage regimens (continue)

Dosage regimens	**MIC**	**PTA (%) with the following CL_{Cr} (mL/min)**			
		60-120	**40-60**	**20-40**	**<20**
Dose 12 g/day					
LD 2 g with 12 g CI	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
LD 4 g with 12 g CI	1	100	100	100	100
	2	100	100	100	100
	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
Dose 16 g/day					
4 g 0.5 h-inf. q 6 h	4	95	100	100	100
	8	62	99	100	100
	16	15	61	93	100
4 g 1 h-inf. q 6 h	4	98	100	100	100
	8	74	100	100	100
	16	21	75	98	100
4 g 2 h-inf. q 6 h	4	100	100	100	100
	8	93	100	100	100
	16	42	97	100	100
4 g 3 h-inf. q 6 h	4	100	100	100	100
	8	100	100	100	100
	16	78	100	100	100
4 g 4 h-inf. q 6 h	4	100	100	100	100
	8	100	100	100	100
	16	100	100	100	100
16 g CI	4	100	100	100	100
	8	100	100	100	100
	16	97	98	98	98

Table 15 CFR in patients with CL_{Cr} 60 to 120 mL/min

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 6 g/day without LD			
2 g 0.5 h-inf. q 8 h	11	28	47
2 g 1 h-inf. q 8 h	14	33	54
2 g 2 h-inf. q 8 h	20	44	66
2 g 3 h-inf. q 8 h	29	57	79
2 g 4 h-inf. q 8 h	42	69	87
6 g CI	69	84	94
Dose 6 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 8 h	27	51	71
LD 4 g then 2 g 1 h-inf. q 8 h	27	51	71
LD 4 g then 2 g 2 h-inf. q 8 h	27	51	71
LD 4 g then 2 g 3 h-inf. q 8 h	27	51	71
LD 4 g then 2 g 4 h-inf. q 8 h	27	51	71
LD 2 g with 6 g CI	76	87	96
LD 4 g with 6 g CI	77	87	96
Dose 8 g/day without LD			
2 g 0.5 h-inf. q 6 h	37	64	84
2 g 1 h-inf. q 6 h	42	69	87
2 g 2 h-inf. q 6 h	53	76	90
2 g 3 h-inf. q 6 h	61	80	92
2 g 4 h-inf. q 6 h	65	82	93
8 g CI	71	85	94
Dose 8 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 6 h	58	79	92
LD 4 g then 2 g 1 h-inf. q 6 h	58	79	92
LD 4 g then 2 g 2 h-inf. q 6 h	58	79	92
LD 4 g then 2 g 3 h-inf. q 6 h	58	79	92
LD 4 g then 2 g 4 h-inf. q 6 h	58	79	92
LD 2 g with 8 g CI	77	87	96
LD 4 g with 8 g CI	82	89	97

Table 15 CFR in patients with CL_{Cr} 60 to 120 mL/min (continue)

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 12 g/day			
4 g 0.5 h-inf. q 8 h	27	51	71
4 g 1 h-inf. q 8 h	31	56	76
4 g 2 h-inf. q 8 h	41	66	85
4 g 3 h-inf. q 8 h	52	75	90
4 g 4 h-inf. q 8 h	62	81	92
12 g CI	79	88	96
LD 2 g with 12 g CI	82	90	97
LD 4 g with 12 g CI	82	90	97
Dose 16 g/day			
4 g 0.5 h-inf. q 6 h	58	79	92
4 g 1 h-inf. q 6 h	62	81	92
4 g 2 h-inf. q 6 h	69	84	94
4 g 3 h-inf. q 6 h	74	86	95
4 g 4 h-inf. q 6 h	76	87	96
16 g CI	79	88	96

Table 16 CFR in patients with CL_{Cr} 40 to 60 mL/min

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 6 g/day without LD			
2 g 0.5 h-inf. q 8 h	30	58	80
2 g 1 h-inf. q 8 h	34	63	83
2 g 2 h-inf. q 8 h	43	70	87
2 g 3 h-inf. q 8 h	52	76	90
2 g 4 h-inf. q 8 h	60	80	92
6 g CI	71	85	94
Dose 6 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 8 h	53	75	90
LD 4 g then 2 g 1 h-inf. q 8 h	53	75	90
LD 4 g then 2 g 2 h-inf. q 8 h	53	75	90
LD 4 g then 2 g 3 h-inf. q 8 h	53	75	90
LD 4 g then 2 g 4 h-inf. q 8 h	53	75	90
LD 2 g with 6 g CI	80	89	97
LD 4 g with 6 g CI	82	89	97
Dose 8 g/day without LD			
2 g 0.5 h-inf. q 6 h	58	79	92
2 g 1 h-inf. q 6 h	61	80	92
2 g 2 h-inf. q 6 h	66	82	93
2 g 3 h-inf. q 6 h	69	84	94
2 g 4 h-inf. q 6 h	73	86	95
8 g CI	72	85	95
Dose 8 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 6 h	72	85	95
LD 4 g then 2 g 1 h-inf. q 6 h	72	85	95
LD 4 g then 2 g 2 h-inf. q 6 h	72	85	95
LD 4 g then 2 g 3 h-inf. q 6 h	72	85	95
LD 4 g then 2 g 4 h-inf. q 6 h	72	85	95
LD 2 g with 8 g CI	82	90	97
LD 4 g with 8 g CI	82	90	97

Table 16 CFR in patients with CL_{Cr} 40 to 60 mL/min (continue)

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 12 g/day			
4 g 0.5 h-inf. q 8 h	53	75	90
4 g 1 h-inf. q 8 h	56	78	91
4 g 2 h-inf. q 8 h	63	81	93
4 g 3 h-inf. q 8 h	69	84	94
4 g 4 h-inf. q 8 h	73	86	95
12 g CI	79	88	96
LD 2 g with 12 g CI	84	90	97
LD 4 g with 12 g CI	85	91	98
Dose 16 g/day			
4 g 0.5 h-inf. q 6 h	72	85	95
4 g 1 h-inf. q 6 h	74	86	95
4 g 2 h-inf. q 6 h	77	87	96
4 g 3 h-inf. q 6 h	78	88	96
4 g 4 h-inf. q 6 h	80	89	97
16 g CI	80	89	96

Table 17 CFR in patients with CL_{Cr} 20 to 40 mL/min

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 6 g/day without LD			
2 g 0.5 h-inf. q 8 h	46	72	88
2 g 1 h-inf. q 8 h	50	74	89
2 g 2 h-inf. q 8 h	56	78	91
2 g 3 h-inf. q 8 h	62	81	92
2 g 4 h-inf. q 8 h	65	82	93
6 g CI	71	85	94
Dose 6 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 8 h	65	82	93
LD 4 g then 2 g 1 h-inf. q 8 h	65	82	93
LD 4 g then 2 g 2 h-inf. q 8 h	65	82	93
LD 4 g then 2 g 3 h-inf. q 8 h	65	82	93
LD 4 g then 2 g 4 h-inf. q 8 h	65	82	93
LD 2 g with 6 g CI	82	90	97
LD 4 g with 6 g CI	82	90	97
Dose 8 g/day without LD			
2 g 0.5 h-inf. q 6 h	66	83	93
2 g 1 h-inf. q 6 h	68	83	94
2 g 2 h-inf. q 6 h	72	85	94
2 g 3 h-inf. q 6 h	75	87	95
2 g 4 h-inf. q 6 h	74	86	95
8 g CI	73	85	95
Dose 8 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 6 h	77	87	96
LD 4 g then 2 g 1 h-inf. q 6 h	77	87	96
LD 4 g then 2 g 2 h-inf. q 6 h	77	87	96
LD 4 g then 2 g 3 h-inf. q 6 h	77	87	96
LD 4 g then 2 g 4 h-inf. q 6 h	77	87	96
LD 2 g with 8 g CI	82	90	97
LD 4 g with 8 g CI	85	91	98

Table 17 CFR in patients with CL_{Cr} 20 to 40 mL/min (continue)

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 12 g/day			
4 g 0.5 h-inf. q 8 h	65	82	93
4 g 1 h-inf. q 8 h	67	83	94
4 g 2 h-inf. q 8 h	71	85	94
4 g 3 h-inf. q 8 h	74	86	95
4 g 4 h-inf. q 8 h	76	87	95
12 g CI	80	89	96
LD 2 g with 12 g CI	87	92	98
LD 4 g with 12 g CI	87	92	98
Dose 16 g/day			
4 g 0.5 h-inf. q 6 h	77	87	96
4 g 1 h-inf. q 6 h	78	88	96
4 g 2 h-inf. q 6 h	80	89	96
4 g 3 h-inf. q 6 h	82	89	97
4 g 4 h-inf. q 6 h	81	89	97
16 g CI	80	89	97

Table 18 CFR in patients with CL_{Cr} < 20 mL/min

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 6 g/day without LD			
2 g 0.5 h-inf. q 8 h	57	78	91
2 g 1 h-inf. q 8 h	59	79	92
2 g 2 h-inf. q 8 h	63	81	93
2 g 3 h-inf. q 8 h	66	83	93
2 g 4 h-inf. q 8 h	69	84	94
6 g CI	72	85	95
Dose 6 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 8 h	71	85	94
LD 4 g then 2 g 1 h-inf. q 8 h	71	85	94
LD 4 g then 2 g 2 h-inf. q 8 h	71	85	94
LD 4 g then 2 g 3 h-inf. q 8 h	71	85	94
LD 4 g then 2 g 4 h-inf. q 8 h	71	85	94
LD 2 g with 6 g CI	82	90	97
LD 4 g with 6 g CI	82	90	97
Dose 8 g/day without LD			
2 g 0.5 h-inf. q 6 h	71	85	94
2 g 1 h-inf. q 6 h	73	86	95
2 g 2 h-inf. q 6 h	76	87	95
2 g 3 h-inf. q 6 h	76	87	95
2 g 4 h-inf. q 6 h	74	86	95
8 g CI	73	86	95
Dose 8 g/day with LD			
LD 4 g then 2 g 0.5 h-inf. q 6 h	80	89	96
LD 4 g then 2 g 1 h-inf. q 6 h	80	89	96
LD 4 g then 2 g 2 h-inf. q 6 h	80	89	96
LD 4 g then 2 g 3 h-inf. q 6 h	80	89	96
LD 4 g then 2 g 4 h-inf. q 6 h	80	89	96
LD 2 g with 8 g CI	85	91	98
LD 4 g with 8 g CI	87	92	98

Table 18 CFR in patients with CL_{Cr} < 20 mL/min (continue)

Dosage regimens	**CFR with the following pathogens (%)**		
	P. aeruginosa	***K. pneumoniae***	***E. coli***
Dose 12 g/day			
4 g 0.5 h-inf. q 8 h	71	85	94
4 g 1 h-inf. q 8 h	73	86	95
4 g 2 h-inf. q 8 h	75	87	95
4 g 3 h-inf. q 8 h	77	88	96
4 g 4 h-inf. q 8 h	78	88	96
12 g CI	80	89	96
LD 2 g with 12 g CI	87	92	98
LD 4 g with 12 g CI	87	92	98
Dose 16 g/day			
4 g 0.5 h-inf. q 6 h	80	89	96
4 g 1 h-inf. q 6 h	80	89	97
4 g 2 h-inf. q 6 h	82	90	97
4 g 3 h-inf. q 6 h	82	90	97
4 g 4 h-inf. q 6 h	81	89	97
16 g CI	81	89	97

4.4. Clinical outcomes

Of 48 patients, 36 patients (75%) were eligible to be assessed the clinical improvement. As for 12 ineligible patients, 6 patients were treated with piperacillin/tazobactam no more than 48 hours. Six patients presented high body temperature from pathophysiology of the concomittent diseases (tumor fever and burn). The clinical improvement rate was 55.6% (20/36). Of 20 patients with clinical improvement, 17 patients exposed pathogens susceptible to piperacillin/tazobactam, most of them showed improved fever within 3 days. There were 4 patients who showed no pathogen in repeated cultures. Antibiotic de-escalation was implemented with 7 patients who had clinical improvement, as shown in Table 19. For 16 patients with clinical failure, 6 patients did not know causative pathogens and 6 patients exposed pathogens not susceptible to piperacillin/tazobactam. Almost all patients had persistent fever and received step-up treatment. Regarding the association between $\%fT_{>MIC}$ and mortality rate, there were 25 patients (52%) with available susceptibility test to piperacillin/ tazobactam (MIC of pathogens could be identified). The 28-day all-cause mortality rate was 28% (7/25). Patient characteristics between alive patients and dead patients are shown in table 20. There was no significant difference in any factors between 2 groups.

Table 19 Clinical responses

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
4	Pyelonephritis	*E.coli* susceptible to pip/taz	Improved fever (Day 1: BT 38.6 °C, Day3: BT 37.3 °C)	No repeat	Deescalate to ciprofloxacin	Improved
8	Septicemia	*E.coli* ESBL no known susceptibility	Improved fever (Day 1: BT 38.0 °C, Day7: BT 37.4 °C)	Day 2: H/C: no growth	No change, Continued 10 days	Improved
9	Urinary tract infection and Septicemia	U/C:*P. aeruginosa* susceptible to pip/taz, H/C: *Streptococcus pneumoniae*	Improved fever (Day 1: BT 38.9 °C, Day3: BT 36.8 °C)	No repeat	Deescalate to levofloxacin	Improved
10	Pneumonia	*K. pneumoniae* and *A. baumanii* susceptible to pip/taz	Improved fever (Day 1: BT 42.0 °C, Day3: BT 36.5 °C)	No repeat	No change, Continued 7 days	Improved
11	Septicemia	*E.coli* susceptible to pip/taz	Improved fever (Day 1: BT 40.2 °C, Day3: BT 38.9 °C)	Day 4: H/C: no growth	Deescalate to ciprofloxacin	Improved

pip/taz, piperacillin/tazobactam; BT, body temperature; U/C, Urine culture; H/C, Hemo culture.

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
13	Urinary tract infection	*E. coli* susceptible to pip/taz	Improved fever (Day 1: BT 38.5 °C, Day3: BT 37.3 °C) Decreased costovertebral angle tenderness	No repeat	No change, Continued 13 days	Improved
15	Pneumonia	*K. pneumoniae* ESBL susceptible to pip/taz	Improved fever (Day 1: BT 38.7 °C, Day5: BT 37.5 °C)	No repeat	No change, Continued 15 days	Improved
16	Urinary tract infection	*K. pneumoniae*, and *E. coli* susceptible to pip/taz	Improved fever (Day 1: BT 38.5 °C, Day3: BT 37.2 °C)	No repeat	Deescalate to ciprofloxacin	Improved
17	Urinary tract infection	*E. coli* ESBL susceptible to pip/taz	Improved fever (Day 1: BT 38.4 °C, Day3: BT 37.4 °C)	No repeat	No change, Continued 14 days	Improved
19	Septicemia	*E. coli* ESBL susceptible to pip/taz	Improved fever (Day 1: BT 37.8 °C, Day3: BT 37.4 °C)	No repeat	No change, Continued 6 days	Improved

pip/taz, piperacillin/tazobactam; BT, body temperature

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
21	Pneumonia	No known	Improved fever (Day 1: BT 38.9 °C, Day3: BT 37.1 °C)	No repeat	No change, Continued 5 days	Improved
23	Pneumonia	*E. coli* susceptible to pip/taz	Improved fever (Day 1: BT 38.6 °C, Day3: BT 37.8 °C)	No repeat	Deescalate to ceftazidime	Improved
28	Pneumonia and Urinary tract infection	*E. coli* susceptible to pip/taz	Improved fever (Day 1: BT 39.5 °C, Day4: BT 37.1 °C)	No repeat	No change, Continued 7 days	Improved
37	Pneumonia	*K. pneumoniae* ESBL and Enterococcus cloaceae susceptible to pip/taz	Improved fever (Day 1: BT 38.7°C, Day3: BT 37.8 °C) No dyspnea	No repeat	Deescalate to ceftriaxone	Improved
38	Pneumonia	*P. aeruginosa* and *K. pneumoniae* ESBL susceptible to pip/taz	Improved tachypnea (Day 1 : RR 28/min, Day 3: RR 24/min) No dyspnea Sputum decreased	No repeat	No change, Continued 8 days	Improved

pip/taz, piperacillin/tazobactam; BT, body temperature

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
40	Septicemia	*E. coli* ESBL susceptible to pip/taz	Improved fever (Day 1: BT 38.0 °C, Day10: BT 37.6 °C)	Day 3: H/C: no growth	No change, Continued 15 days	Improved
45	Septicemia	*P. aeruginosa* susceptible to pip/taz	Improved fever (Day 1: BT 39.4 °C, Day3: BT 37.8 °C)	No repeat	Deescalate to ciprofloxacin	Improved
46	Peritonitis	No known	Improved fever (Day 1: BT 38.5 °C, Day4: BT 36.8 °C) Abdominal tenderness decreased	No repeat	No change, Continued 5 days	Improved
48	Urinary tract infection and Septicemia	E. coli ESBL susceptible to pip/taz	No pain in lower abdomain No turbid urine	Day 3: U/C: no growth	No change, Continued 3 days	Improved
49	Pneumonia	Enterobacter aerogenes susceptible to pip/taz	Improved fever (Day 1: BT 37.7°C, Day3: BT 36.5 °C)	No repeat	No change, Continued 5 days	Improved

pip/taz, piperacillin/tazobactam; BT, body temperature; U/C, Urine culture; H/C, Hemo culture.

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
1	No known	No known	Day 1: BT 39.0 °C Blood pressure drop	No repeat	Step up to imipenem/cilastatin	Failured
2	No known	No known	Worsening fever (Day 1: BT 38.5 °C, Day3: BT 39.5 °C)	No repeat	Step up to imipenem/cilastatin and colistin	Failured
6	Pneumonia	No known	Persistent fever (Day 1: BT 39.0 °C, Day3: BT 39.1°C)	No repeat	Step up to imipenem/cilastatin	Failured
20	Pneumonia	No known	Persistent fever (Day 1: BT 39.8 °C, Day5: BT 39.9°C)	No repeat	Step up to sulperazone and colistin	Failured
22	Pneumonia	No known	Persistent fever (Day 1: BT 40.8 °C, Day2: BT 40.7°C)	No repeat	Step up to imipenem/cilastatin	Failured

BT,body temperature

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
24	Pyelonephritis	*K. pneumoniae* ESBL not susceptible to pip/taz	Worsening fever (Day 1: BT 38.8 °C, Day3: BT 39.8 °C)	No repeat	Step up to imipenem/cilastatin	Failured
26	Pneumonia	*P. aeruginosa* not susceptible to pip/taz	Day 1: BT 38.8 °C	No repeat	Step up to imipenem/cilastatin	Failured
27	Pneumonia	*S. aureus* not susceptible to Pip/Taz and *P. aeruginosa* susceptible to pip/taz	Persistent fever (Day 1: BT 38.1 °C, Day2: BT 38.1 °C)	No repeat	Step up to imipenem/cilastatin	Failured
29	Pneumonia	*K. pneumoniae* susceptible to pip/taz	Persistent fever (Day 1: BT 38.8 °C, Day8: BT 38.1 °C)	No repeat	Step up to imipenem/cilastatin	Failured
30	Pneumonia	*A. baumanii* (MDR)	Worsening fever (Day 1: BT 37.1 °C, Day4: BT 38.3 °C)	No repeat	Step up to imipenem/cilastatin	Failured

pip/taz, piperacillin/tazobactam; BT, body temperature

Table 19 Clinical responses (continue)

No.	Suspected infections	Pathogens	Assessment criteria			Clinical responses
			(i) Clinical signs and symptoms	(ii) Repeat cultures	(iii) Changes in antibiotic therapy	
32	Pneumonia	Enterobacter cloaceae no known susceptibility	Day 1: BT 38.1 °C Blood pressure drop	No repeat	Step up to imipenem/cilastatin	Failured
33	Urinary tract infection	*E. coli* ESBL not susceptible to pip/taz	Day 1: BT 40.5°C	No repeat	Step up to imipenem/cilastatin	Failured
36	Pneumonia	*K. Pneumoniae* ESBL and *Enterobacter cloaceae* susceptible to pip/taz	Worsening fever (Day 1: BT 38.9 °C, Day3: BT 39.9 °C)	No repeat	No change	Failured
39	Pneumonia	*A. baummanii* not susceptible to pip/Taz	Persistent fever (Day 1: BT 38.2 °C, Day3: BT 38.1 °C)	No repeat	No change	Failured
43	No known	No known	Day 1: BT 37.8°C Blood pressure drop	No repeat	Step up to imipenem/cilastatin	Failured
50	Urinary tract infection	*E. coli* ESBL susceptible to pip/taz	Persistent fever (Day 1: BT 38.2 °C, Day2: BT 38.2 °C) Blood pressure drop	No repeat	Step up to meropenem	Failured

piperacillin/tazobactam; BT, body temperature

Table 20 Patient characteristics between alive and dead patients

Variables	**Alive patients (N=18)**	**Dead patients (N=7)**	***p-value*[a]**
Age (years)	70 (62-87)	60 (46-79)	0.485
Total body weight (kg)	53.5 (45.0-57.3)	56 (51.8-66.1)	0.317
Body mass index (kg/m^2)	19.8 (18.9-21.5)	20.1 (19.3-23.0)	0.785
Creatinine clearance (mL/min)	56.0 (41.7-84.6)	54.2 (35.6-94.0)	0.949
APACHE II score	22 (18-23)	22 (20-27)	0.466
Mean arterial pressure (mmHg)	67 (61-70)	63 (46-68)	0.108
$\%fT_{>MIC}$ (%)	99.7 (98.7-99.7)	99.7 (99.5-99.7)	0.442
Pathogens susceptible to PIP/TAZ, n (%)	15 (83)	7 (100)	0.250
Dosage regimens, n(%)			
4/0.5 g 0.5-h infusion q 6 h	16 (88)	6 (86)	0.826
4/0.5 g 0.5-h infusion q 8 h	1 (6)	0 (0)	0.524
4/0.5 g then 2/0.25 g 0.5-h infusion q 6 h	1 (6)	1 (14)	0.470
Concomitant diseases, n(%)			
Cancer	7 (39)	3 (43)	0.856
Myocardial infarction	1 (6)	2 (29)	0.112
Concomitant antibiotics, n(%)			
PIP/TAZ monotherapy	16 (88)	7 (100)	0.358
Vancomycin	1 (6)	0 (0)	0.524
Metronidazole	1 (6)	0 (0)	0.524

Data are presented as median (interquartile range) or number (percentage). [a] Mann-Whitney U test for continuous variables and Pearson Chi-square test for categorical variables; [b] p-value < 0.05 (2-sided)

CHAPTER V DISCUSSION

This work aimed to characterize population pharmacokinetic (PK) behaviors, investigate the probability of target attainment (PTA) of various piperacillin dosage regimens and explore the cumulative fraction of response (CFR) against pathogens commonly found in critically ill patients with the early phase of sepsis. There have been two previous works which studied the population PKs and pharmacodynamics (PDs) of piperacillin in patients with the early phase of sepsis.(24, 27) Obrink-Hansen et al. collected blood samples during the third administration of 4-g of piperacillin (3-min infusion) every 8 hours in 15 patients with septic shock. Of these, 67% (10 patients) had acute kidney injury (AKI).(27) Roberts et al. studied on Days 1 (first-dose) and days 2 (steady-state) of therapy in 16 patients with sepsis and normal renal function, with 8 patients receiving intermittent bolus and 8 patients receiving continuous infusion (CI).(24) In this present study, we explore population PKs of piperacillin in 48 patients during their first 24 hours of sepsis. Fourteen patients (29%) had septic shock and 14 patients (29%) had unstable renal function.

Based on our final population PK model, the PK behavior of piperacillin in the early phase of sepsis was best described by the two-compartment model, consistent with two previous studies.(24, 27) The piperacillin total volume of distribution (V_d) in our population (17.12 L) is smaller than that of Roberts et al. (25.0 L) but V_d normalized to total body weight (TBW) is quite similar (0.30 vs 0.33 L/kg). This finding suggests that the difference in V_d is probably a result of the difference in body size (median TBW 56.6 vs 75.7 kg).(24) However, the total V_d in our population is much larger than the result of Obrink-Hansen et al. (11.20 L, 0.14 L/kg).(27) The plausible explanation is that all patients in Obrink-Hansen et al. had septic shock and obtained vasopressor therapy which can cause vasoconstriction and may limit the drug distribution, subsequently. In addition, the patients in the present study had higher the acute physiology and chronic health evaluation II (APACHE II) score (median score of 22 vs 19), indicating higher severity of disease which may relate to higher V_d.

Interestingly we found that piperacillin central volume of distribution (V_1) (9.35 L) is larger than piperacillin peripheral volume of distribution (V_2) (7.77 L) similar to that of Obrink-Hansen et al. (7.3 and 3.9 L, respectively).(27) The large V_1

of piperacillin, a hydrophilic antibiotic, could be due to the increased capillary permeability and fluid resuscitation during the early phase of sepsis. Contrarily, Roberts et al. reported a smaller V_1 (V_1 7.2 L and V_2 17.8 L).(24) One explanation for this discrepancy could be that Roberts et al. included the data during steady state. It should also be noted that they did not directly measure free piperacillin concentrations.

The median piperacillin total clearance (CL) was 5.37 L/h which is lower than the results of Roberts et al. (17.20 L/h)(24) but higher than that of Obrink-Hansen et al. (3.60 L/h)(27). piperacillin is eliminated primarily via renal clearance. These differences in CL could be results of the differences in renal function of the studied populations of each study. Roberts et al. included patients with normal renal function(24), while AKI occurred in 67% of all patients in Obrink-Hansen's study(27). Due to the narrow range of renal function, Roberts et al. did not find creatinine clearance (CL_{Cr}) as a covariate to predict CL(24) while Obrink-Hansen et al. indicated that plasma creatinine was a significant covariate to CL(27).

When we performed the covariate exploration step, initially we did graphical examination of the relationship of CL and different indicators of renal function including plasma creatinine, CL_{Cr} calculated by Cockcroft-Gault's, by Jellife's and by MDRD equations. The best potential predictor was CL_{Cr} calculation by using the Cockcroft-Gault equation if a patient's renal function was stable or the Jelliffe equation if unstable. We think it is a rational approach because 14% of our patients had unstable renal function. Apparently, addition of this CL_{Cr} as a covariate of CL in forward addition step provided the most significantly improvement of the model (a change in objective function value (ΔOFV) of -25.41). Interestingly; the second most significant predictor was mean arterial pressure (MAP), an average blood pressure during a single cardiac cycle, as a covariate of CL. An increase in MAP might associate with increasing blood perfusion to organs including kidney and liver, resulting in rising CL. From the final model, if MAP increases 10 mmHg, CL will raise 0.5 L/h, thus piperacillin dosing increment might be necessary.

In this study, 6.3% of our patients were overweight, 16.7% were obese and 10.4% were morbidly obese. We explored the relationship of V_1 and CL vs 3 types of weight; TBW, ideal body weight (IBW) and adjusted body weight (ABW), and we

found that ABW is the third most significant predictor as a covariate for V_1. Addition of ABW reduced the interindividual variability (IIV) of V_1 from 64.0% to 55.4%. Although many previous studies reported that TBW has been a significant covariate for V_d (19, 23-26), ABW has also been suggested to be a plausible size descriptor for beta-lactams.(22, 23) Since piperacillin is hydrophilic, using ABW for overweight patients seems to be a reasonable approach. Our model indicated that an increase in ABW of 1 kg would increase the V_1 of 0.26 L. This finding suggests that obese patients may need higher-than-usual loading dose (LD). Roberts et al found that TBW could reduce IIV of CL but not statistically significance (ΔOFV of -2.35).(24) Our study also found that weight was not a significant covariate for CL but it should be noted that weight were used in CL_{Cr} calculation.

Regarding the PK/PD target, an *in vitro* study found that 75% $fT_{>MIC}$ could provide bactericidal activity of piperacillin.(10) However, higher PK/PD targets (90 to 100% $fT_{>MIC}$) have been recommended for microbiological success and prevention of bacterial regrowth in patients with serious bacterial infections.(13) From the results of the preliminary analysis pertaining to 90 and 100% $fT_{>MIC}$, 90% $fT_{>MIC}$ was chosen as the target in this study. During the early phase of sepsis, this study found that a standard dosage regimen; 4 g 30 min-infusion every 6 h could achieve PTA ≥ 90% (a target of 90% $fT_{>MIC}$) at MIC 16 mg/L in patients with CL_{Cr} 10 to 40 mL/min but could not achieved the target in patients with CL_{Cr} 40 to 120 mL/min. While another standard dosage regimen; 4 g 30 min-infusion every 8 h could not achieve the target in patients with all different levels of CL_{Cr}. Likewise, Obrink-Hansen et al. found that during the early phase of septic shock, 4 g 3 min-infusion every 8 h could not provide PTA ≥ 90% for both targets of 100% $fT_{>MIC}$ and 50% $fT_{>4MIC}$ at MIC 16 mg/L in patients with all groups of plasma creatinine (80, 150, 250 μmol/L).(27) Similarly, Robert et al also reported that during the first dose, 4 g 20 min-infusion every 6 h and 8 h could not provide PTA ≥ 90% (a target of 50% $fT_{>MIC}$) at MIC 16 mg/L in patients with sepsis and normal renal function.(24)

To overcome the non-target attainment problem, prolonged infusion has been suggested to increase the PTA. The results of this study show that prolonged infusion could achieve PTA ≥ 90% more than short infusion in patients with CL_{Cr} 40 to 120

mL/min similar to previous studies.(24, 27) However, using every-6-hours dosing in patients with CL_{Cr} < 20 mL/min, the 2 and 3 hours-infusion time provided PTA ≥ 90% while the 4 hours-infusion time could not achieve PTA ≥ 90%. This finding can imply that the longer infusion time did not show the better benefit to attain the PTA target in patients with CL_{Cr} < 20 mL/min who received every-6-hours dosing consistent with the study of Abdul-Aziz et al. They reported that beta-lactam exposure is more likely to be adequate in patients with significant renal impairment, regardless of the drug administration method.(95) Moreover, to achieve PTA ≥ 90% at MIC 16 mg/L, this study found that LD 2 or 4 g is necessary for continuous infusion of dose 6 and 8 mg/day, without the LD, these dosage regimens could achieve the target at only MIC 8 mg/L.

Regarding CFR, all dosage regimens provided CFR ≥ 90% for the *E. coli* infection similar to previous studies.(75) When considering the *K. pneumoniae* infection, 6, 8 and 12 g CI with LD could be useful options for patients with CL_{Cr} < 40 mL/min, 40 to 60 mL/min, and 60 to 120 mL/min respectively. Different from Alobaid et al. using a target of 50%$fT_{>MIC}$, besides continuous infusion, they also found that short and extended infusion could provide PTA achievement.(75) There was no dosage regimen provided CFR ≥ 90% for *P. aeruginosa* consistent with previous studies.(26, 75) Likewise, Udy et al. also found that 4 g 20 min-infusion every 6 h could not provide CFR ≥ 90% (both targets of 50% or 100% $fT_{>MIC}$) for *P. aeruginosa* in septic patients with CL_{Cr} 10 to 300 mL/min.(26) Alobaid et al. reported that 4 g every 8 or 6 h (all modes of administration) could not achieve CFR ≥ 90% (50% $fT_{>MIC}$) for *P. aeruginosa* in critically ill patients with CL_{Cr} 30, 50, 150 mL/min.(75) It should be noted that maximum daily dose (16 g of piperacillin/day) in this study were based on manufacturer's licensing dose of piperacillin/tazobactam, however, piperacillin alone may be used at higher dose to attain the target. Alternative treatment should be considered to achieve effective treatment. Vojtová et al. documented that combined therapy between piperacillin/tazobactam and amikacin showed the relative safety and usefulness in the treatment of infections caused by *P. aeruginosa* at intensive care unit (96) but such treatment should be carefully used in

patients with renal impairment. Another approach proposed is the treatment with carbapenem antibiotics.(97)

Regarding clinical outcomes, there was no predictor shown to be significant. Differently, Robert et al. reported that APACHE II score was significantly related to hospital mortality in patients with severe sepsis (n=632).(36) Likewise, Fan et al. found that APACHE II score of 29.5 or higher was the significant predictor for 14-day mortality rate in critically ill patients (n=367).(40) The difference in these findings may result from the smaller sample size in this study. In addition, $\%fT_{>MIC}$ was not be found to be a significant factor to predict mortality rate, it might result from the homogenous data in both groups of patients.

The limitation of this study should be noted. This study explored the likelihood of target attainment using EUCAST data to improve generalizability for empirical dosing, although it limits conclusions about sufficiency of drug exposure in any specific patients. In addition, due to the relatively small sample size in each subgroup of renal function used in this simulation, expanding sample size in the further study may have revealed some interesting findings.

In conclusions, this work has shown that subtherapeutic concentrations can occur in patients with normal renal function, prolonged infusion has been a beneficial tool to enhance target attainment. However, in patients with renal impairment, the target could be achieved by using shorter infusion. Regarding the susceptibility of pathogens, standard dosage regimens should be considered that they might not provide enough target attainment for pathogens with MIC ≥16 mg/L, prolonged infusion may be needed to increase concentrations for target achievement.

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APPENDIX

APPENDIX A: The acute physiology and chronic health evaluation (APACHE) II severity of disease classification system

PHYSIOLOGIC VARIABLE	HIGH ABNORMAL RANGE					LOW ABNORMAL RANGE			
	+4	+3	+2	+1	0	+1	+2	+3	+4
TEMPERATURE — rectal (°C)	○ ≥ 41°	○ 39°-40.9°		○ 38.5°-38.9°	○ 36°-38.4°	○ 34°-35.9°	○ 32°-33.9°	○ 30°-31.9°	○ ≤ 29.9°
MEAN ARTERIAL PRESSURE — mm Hg	○ ≥ 160	○ 130-159	○ 110-129		○ 70-109		○ 50-69		○ ≤ 49
HEART RATE (ventricular response)	○ ≥180	○ 140-179	○ 110-139		○ 70-109		○ 55-69	○ 40-54	○ ≤ 39
RESPIRATORY RATE — (non-ventilated or ventilated)	○ ≥50	○ 35-49		○ 25-34	○ 12-24	○ 10-11	○ 6-9		○ ≤ 5
OXYGENATION: A-aDO$_2$ or PaO$_2$ (mm Hg) a. FIO$_2$ ≥ 0.5 record A-aDO$_2$	○ ≥ 500	○ 350-499	○ 200-349		○ <200				
b. FIO$_2$ < 0.5 record only PaO$_2$					○ PO$_2$ > 70	○ PO$_2$ 61-70		○ PO$_2$ 55-60	○ PO$_2$ < 55
ARTERIAL pH	○ ≥7.7	○ 7.6-7.69		○ 7.5-7.59	○ 7.33-7.49		○ 7.25-7.32	○ 7.15-7.24	○ < 7.15
SERUM SODIUM (mMol/L)	○ ≥180	○ 160-179	○ 155-159	○ 150-154	○ 130-149		○ 120-129	○ 111-119	○ ≤110
SERUM POTASSIUM (mMol/L)	○ ≥7	○ 6-6.9		○ 5.5-5.9	○ 3.5-5.4	○ 3-3.4	○ 2.5-2.9		○ < 2.5
SERUM CREATININE (mg/100 ml) (Double point score for **acute** renal failure)	○ ≥3.5	○ 2-3.4	○ 1.5-1.9		○ 0.6-1.4		○ < 0.6		
HEMATOCRIT (%)	○ ≥ 60		○ 50-59.9	○ 46-49.9	○ 30-45.9		○ 20-29.9		○ < 20
WHITE BLOOD COUNT (total/mm3) (in 1,000s)	○ ≥40		○ 20-39.9	○ 15-19.9	○ 3-14.9		○ 1-2.9		○ < 1
GLASGOW COMA SCORE (GCS): Score = 15 minus actual GCS									
[A] Total ACUTE PHYSIOLOGY SCORE (APS): Sum of the 12 individual variable points									
Serum HCO$_3$ (venous-mMol/L) [Not preferred, use if no ABGs]	○ ≥52	○ 41-51.9		○ 32-40.9	○ 22-31.9		○ 18-21.9	○ 15-17.9	○ < 15

[B] AGE POINTS:
Assign points to age as follows:

AGE(yrs)	Points
≤ 44	0
45-54	2
55-64	3
65-74	5
≥ 75	6

[C] CHRONIC HEALTH POINTS
If the patient has a history of severe organ system insufficiency or is immuno-compromised assign points as follows:
a. for nonoperative or emergency postoperative patients — 5 points
or
b. for elective postoperative patients — 2 points

DEFINITIONS
Organ insufficiency or immuno-compromised state must have been evident **prior** to this hospital admission and conform to the following criteria:

LIVER: Biopsy proven cirrhosis and documented portal hypertension; episodes of past upper GI bleeding attributed to portal hypertension; or prior episodes of hepatic failure/encephalopathy/coma.

CARDIOVASCULAR: New York Heart Association Class IV.

RESPIRATORY: Chronic restrictive, obstructive, or vascular disease resulting in severe exercise restriction, i.e., unable to climb stairs or perform household duties; or documented chronic hypoxia, hypercapnia, secondary polycythemia, severe pulmonary hypertension (>40mmHg), or respirator dependency.

RENAL: Receiving chronic dialysis.

IMMUNO-COMPROMISED: The patient has received therapy that suppresses resistance to infection, e.g., immuno-suppression, chemotherapy, radiation, long term or recent high dose steroids, or has a disease that is sufficiently advanced to suppress resistance to infection, e.g., leukemia, lymphoma, AIDS.

APACHE II SCORE
Sum of [A] + [B] + [C]

[A] APS points

[B] Age points

[C] Chronic Health points

Total APACHE II

APPENDIX B: The sequential organ failure assessment (SOFA) scoring system

SOFA score	1	2	3	4
Respiratory system				
Pao_2/Fio_2 (mmHg)	<400	<300	<220	<100
Sao_2/Fio_2	221-301	142-220	67-141	<67
Coagulation				
Platelet x $10^3/mm^3$	<150	<100	<50	<20
Liver system				
Bilirubin (mg/dL)	1.2-1.9	2.0-5.9	6.0-11.9	>12.0
Cardiovascular system				
Hypotension	MAP<70	Dopamine ≤ 5 or Dobutamine (any)	Dopamine > 5 or Norepinephrine ≤ 0.1	Dopamine > 15 or Norepinephrine > 0.1
Central nervous system				
Glasgow Coma Score	13-14	10-12	6-9	<6
Renal system				
Creatinine (mg/dL) or Urine output (mL/day)	1.2-1.9	2.0-3.4	3.5-4.9 or <500	>5.0 or <200

[a] vasoactive mediations administered for at least 1 hour (dopamine and norepinephrine μg/kg/min). Pao_2/Fio_2, ratio of partial pressure of oxygen and fraction of inspired oxygen; Sao_2, peripheral arterial oxygen saturation; MAP, mean arterial pressure.

APPENDIX C: Jelliffe equation for patients with unstable renal function

For male patients:

CL_{Cr} (mL/min/1.73 m^2) = {[(IBW x (29.3-(0.203xAge))) x (1.035-(0.0337 x S_{Cr}))] – [4 x IBW* x (S_{Cr1}-S_{Cr2}) / Δt]} / (14.4 x S_{Cr})

For female patients:

CL_{Cr} (mL/min/1.73 m^2) = {[(IBW x (25.1-(0.175xAge))) x (1.035-(0.0337 x S_{Cr}))] – [4 x IBW* x (S_{Cr1}-S_{Cr2}) / Δt]} / (14.4 x S_{Cr})

* Use IBW if TBW > 130% of IBW

APPENDIX D: Evaluation of FOCEI and SAEM estimation methods

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ORIGINAL RESEARCH ARTICLE

Evaluation of FOCEI and SAEM Estimation Methods in Population Pharmacokinetic Analysis Using NONMEM® Across Rich, Medium, and Sparse Sampling Data

Waroonrat Sukarnjanaset[1] · Thitima Wattanavijitkul[1] · Sutep Jarurattanasirikul[2]

Abstract

Background and Objectives First-order conditional estimation with interaction (FOCEI) is one of the most commonly used estimation methods in nonlinear mixed effects modeling, while the stochastic approximation expectation maximization (SAEM) is the newer estimation algorithm. This work aimed to compare the performance of FOCEI and SAEM methods when using NONMEM® with the classical one- and two-compartment models across rich, medium, and sparse data.

Methods One- and two-compartment models of the previous studies were used to simulate data in three scenarios: rich, medium, and sparse data. For each scenario, there were 100 data sets, containing 100 individuals in each data set. Every data set was estimated with both FOCEI and SAEM methods. The simulation and estimation were performed using NONMEM®. The completion rates, percentage of relative estimation errors (%RERs), root mean square errors (RMSEs), and runtimes were considered to assess the completion, accuracy, precision, and speed of estimation, respectively.

Results Both FOCEI and SAEM methods provided comparable completion rates, median %RERs (ranged from − 9.03 to 3.27% for FOCEI and − 9.17 to 3.27% for SAEM) and RMSEs (ranged from 0.0004 to 1.244 for FOCEI and 0.0004 to 1.131 for SAEM) for most parameters in both models across three scenarios. The run times were much shorter with FOCEI (ranged from 0.18 to 0.98 min) compared to SAEM method (ranged from 4.64 to 12.03 min).

Conclusions For the classical one- and two-compartment models, FOCEI method exhibited comparable performance similar to SAEM method but with significantly shorter runtimes across rich, medium, and sparse sampling scenarios.

Key Points

A classical estimation method, FOCEI using the approximate maximum likelihood, is widely used, while a newer method, SAEM which uses the exact maximum likelihood, could be more reliable.

With default options, FOCEI performed similarly to SAEM but with significantly shorter runtimes across sparse, medium and rich clinical data scenarios.

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1 Introduction

Nonlinear mixed effect modeling with the software NONMEM® is regarded as the gold standard in population pharmacokinetic/pharmacodynamic (PK/PD) analysis. Currently, several estimation methods are available in NONMEM® including first-order conditional estimation

(FOCE), Laplace, iterative two-stage, Monte Carlo expectation–maximization (EM), and Monte Carlo Bayesian methods [1–3]. Among the classical methods, first-order conditional estimation with interaction (FOCEI) is the most commonly used in clinical literature.

As described in NONMEM® user's guide, FOCEI involves first-order approximation around individual estimates of the random effects. The interaction between inter-individual and residual variability is also taken into account. The stochastic approximation expectation maximization (SAEM) is the newer estimation algorithm. It was originally implemented in the MONOLIX® software and later in NONMEM® version 7. While FOCEI uses the approximate maximum likelihood, SAEM uses the exact maximum likelihood [4–6]. SAEM takes part in two steps including expectation and maximization steps. In the expectation step, the observed data and the previous predicted parameter values are used to expect the likelihood using Monte Carlo simulation and stochastic approximation, while in the maximization steps, the new parameter values are computed to maximize the likelihood based on the expectation step [5, 7, 8]. Theoretically, SAEM method should be less susceptible to locking into a local minimum and provides more accurate results than the classical methods.

There are a few systematic comparisons of different estimation methods implemented in NONMEM® which were likely to focus on relatively complex models [4, 9–11]. Gibiansky et al. [4] compared speed and accuracy of estimation methods available in NONMEM® using simulated examples of a target-mediated drug disposition model with rich sampling. They found that the SAEM algorithm provided estimates similar to those of FOCEI but with shorter runtimes. Plan et al. [9] also compared the performance of several algorithms implemented in different programs for dose–response models. Each data set was analysed twice, with true initial condition and with altered initial conditions. They found that both FOCE and SAEM run in NONMEM performed well, but SAEM required adequately truthful initial estimates or appropriately tuned to obtain accurate parameter values. Johansson et al. [10] investigated performance of different estimation methods available in NONMEM® 7 for a various set of PD models. They did not show any clear different performance of FOCE and SAEM methods in terms of bias and precision, but FOCE was faster. On the other hand, Liu et al. [11] evaluated three EM methods [SAEM, Monte Carlo importance sampling parametric EM (IMP), quasi-random parametric EM (QREM)] and FOCE for their accuracy and speed when solving complex population physiologically based pharmacokinetic models. They showed that EM methods were faster and more stable than FOCE method for complex models and sparse data.

Currently, most of the published population PK analyses in clinical studies, both rich and sparse data, still use FOCEI and the pharmacokinetic behavior of most drugs can be described by the classical one- or two-compartment model. Therefore, we are interested in comparing the performance (accuracy, precision, completed estimations, and runtimes) of FOCEI and SAEM estimation methods in population PK analysis using NONMEM® when implemented with the classical one- and two-compartment models across rich, medium, and sparse sampling data. Piperacillin, a broad-spectrum β-lactam antibiotic frequently used in critically ill patients, was selected as the model drug. Data sets were simulated based on previously published clinical PK studies.

2 Methods

2.1 Data and Models

Two previously published results modeled with one [12] and two [13] compartment models were used for the comparison (Table 1). Chen et al. [12] collected data from 50 adult patients with nosocomial infections (6–7 blood samples per patient) and they found that a one-compartment model adequately described piperacillin pharmacokinetics. Bulitta et al. [13] analysed pharmacokinetic data from eight adult cystic fibrosis patients (21 blood samples per patient) and a two-compartment model well described the data. We simulated concentration–time profiles in three different scenarios for each model: rich sampling data (12 samples per subject for one-compartment model; 21 samples per subject for two-compartment model as previously described [13]), medium sampling data (7 samples per subject), and sparse sampling data (3 samples per subject). Optimal sampling times were chosen using PFIM Interface 4.0 by assigning the numbers of sampling timepoints of each dosing interval were 12, 7, and 3 points for the rich (for the one-compartment model), medium, and sparse data, respectively. The simulated data below the limit of quantification were removed from the data sets. In each scenario, one hundred data sets were simulated, each consisting of 100 individuals. ADVAN1 TRANS2 and residual variability with proportional model were used to simulate the one-compartment models, while ADVAN3 TRANS4 and residual variability with combined (additive and proportional) model were used to simulate the two-compartment models. Every simulated data set was separately estimated with both FOCEI and SAEM estimation methods with default options to obtain parameter estimates (see Electronic Supplementary material for the control stream used). Initial estimates remained the same across all trials.

Table 1 Summary of pharmacokinetic model parameter values used in the simulation and estimation

Parameters	One-compartment model [12]		Two-compartment model [13]	
	Parameter values for the simulation	Initial estimates for the estimation	Parameter values for the simulation	Initial estimates for the estimation
Cl (L/h)	13.74	10	11.3	10
V_d or V_1 and V_2 (L)	21.69	20	6.49 and 3.12	10 and 5
Q (L/h)	NA	NA	12.8	15
ω^2_{Cl} (%CV)	0.097 (31.1)	0.01	0.011 (10.4)	0.01
ω^2_{Vd} or ω^2_{V1} and ω^2_{V2} (%CV)	0.144 (38.0)	0.01	0.068 (26.0) and 0.117 (34.2)	0.01 and 0.01
ω^2_{Q} (%CV)	NA	NA	NA	NA
σ^2_{prop}	0.00868	0.01	0.0174	0.01
σ^2_{add}	NA	NA	3.534	1

Cl clearance, V_d volume of distribution, V_1 central volume of distribution, V_2 peripheral volume of distribution, Q intercompartmental clearance, ω^2 variance of inter-individual variability, σ^2_{prop} variance of residual variability with proportional model, σ^2_{add} variance of residual variability with additive model, *NA* not available

2.2 Hardware and Software

The simulation and estimation were conducted using NONMEM® version 7.3 (ICON Development Solutions, Ellicott city, MD, USA) under Windows 7 Enterprise 64-bit operating system. The NONMEM® runs were executed by PDx-Pop® version 5.2 (ICON Development Solutions, Ellicott city, MD, USA). Sampling timepoints were chosen using PFIM Interface 4.0 (PFIM group, INSERM and Université Paris Diderot, Paris, France).

2.3 Performance Evaluation

2.3.1 Completion Rates

The completion rates were calculated from the proportion of data sets that produced parameter estimates with each algorithm, however; when the completion rates were less than 50%, the accuracy and precision were not evaluated.

2.3.2 Accuracy and Precision

The true parameter values for simulation were used as expected parameter estimates.

Both fixed effect [clearance (Cl), volume of distribution (V_d), central volume of distribution (V_1), peripheral volume of distribution (V_2), and intercompartmental clearance (Q)] and random effect [variance of inter-individual variability (ω^2), variance of residual variability (σ^2)] parameter estimates were evaluated. The percentage of relative estimation error (%RER) and root mean square error (RMSE) was calculated to evaluate the accuracy and precision of parameter estimates by each method. The mathematical definitions of these evaluations are shown in Eqs. 1 and 2:

$$\%\mathrm{RER}({}^{a}_{p}\theta i) = \left(\frac{{}^{a}_{p}\theta i - {}_{p}\theta}{{}_{p}\theta}\right) \times 100 \quad (1)$$

$$RMSE({}^{a}_{p}\theta) = \sqrt{\frac{1}{N}\sum_{i-1}^{N}({}^{a}_{p}\theta i - {}_{p}\theta)^2} \quad (2)$$

where ${}^{a}_{p}\theta i$ represents the estimate of parameter p in data set i obtained with algorithm a, and ${}_{p}\theta$ represents the true value of parameter (the value used in the simulations).

2.3.3 Runtimes

Runtimes were recorded as the computation (CPU) time needed to estimate parameters for each data set with each algorithm.

3 Results

Among 600 simulated data sets of 100 individuals each, less than 10% (median 2.5%, range 0–8%) of total observations of each data set were below the limit of quantification and were excluded from the analysis. The numbers of simulated concentrations in rich data set were 8–12 samples per subject based on the one-compartment model and 15–20 samples per subject based on the two-compartment model. The medium and sparse sampling data contained 4–7 samples per subject and 1–3 samples per subject, respectively. There were not more than two subjects (2%) with a single timepoint in each sparse data set.

Table 2 Completion rates of FOCEI and SAEM estimation methods

Model	Completion rate (%)					
	Sparse data		Medium data		Rich data	
	FOCEI	SAEM	FOCEI	SAEM	FOCEI	SAEM
One-compartment model	100	100	100	100	100	100
Two-compartment model	21	21	100	100	100	100

FOCEI first-order conditional estimation with interaction, *SAEM* stochastic approximation expectation maximization

The completion rates for each method and model across 3 scenarios are shown in Table 2. Both FOCEI and SAEM provided the same completion rates for all scenarios. Full completion rate (100%) was obtained for all scenarios except sparse data sets from two-compartment model (21% completion rate), and therefore, the accuracy and precision were not assessed for this scenario because of failing to meet the 50% completion criterion.

Figures 1 and 2 illustrate box plots of %RER for parameter estimates of the one- and two-compartment models, respectively. For the one-compartment model, both FOCEI and SAEM methods comparably estimated all five parameters across three scenarios. They provided accurate parameter estimates, although a few marked deviations were found in some random effect parameters particularly with the sparse data. In addition, ranges of %RER of residual variability when implemented with the sparse data were apparently larger than those when implemented with medium and rich data. Similarly, for the two-compartment model, both FOCEI and SAEM exhibited comparable estimations from the medium or rich data. Both estimation methods could accurately estimate parameters, although a few noticeable deviations were found in some parameters particularly the random effect parameters. In addition, FOCEI and SAEM methods provided comparable RMSEs across three scenarios (Table 3).

The median runtimes for each method and model across three scenarios are shown in Table 4. FOCEI had much shorter runtimes than SAEM. Runtimes approximately ranged from 0.18 to 0.98 min and from 4.64 to 12.03 min when using FOCEI and SAEM, respectively.

4 Discussion

This study evaluated the performance of FOCEI and SAEM estimation methods in population PK analysis when implemented with the classical one- and two-compartment models across the rich, medium, and sparse data. We chose piperacillin as the model drug, because it is commonly used in critically ill patients, and there are available published models from patient data. The final models with median values of covariates were used in this comparison.

FOCEI and SAEM provided full completion rate (100%) for most scenario except when implemented with the sparse data of two-compartment model, the completion rates were equally low (21%). This may suggest that data sets of 100 individual with sparse sampling (1–3 sample per subject) are not sufficient to identify two-compartment kinetics. One major feature of SAEM method is the adjustability of the sampling process, but we used default condition in this comparison. SAEM method may perform better with appropriately tuned condition [9]. Liu et al. found that SAEM successfully executed when implemented with the sparse data and complex PBPK models [11]. It may result from the higher number of burn-in iterations (NBURN = 4000) in Liu et al., while NBURN of this study was set to 1000 by default with the same number of accumulation iterations (NITER = 1000) and the same option of Markov Chain Monte Carlo Bayesian Metropolis–Hastings algorithm (ISAMPLE = 2).

FOCEI and SAEM performed similarly in terms of bias and precision. Both methods provided comparably accurate and precise parameter estimates of the one- and two-compartment models for all studied scenarios. The %RER of random effect parameter estimates was larger than % RER of fixed effect parameters. Similar to a previous study with rich sampling data, fixed effect parameter estimates of both FOCEI and SAEM methods with naïve options were similarly close to their true values, while random effect parameter estimates had more deviation [4]. Although SAEM, using exact likelihood algorithm, has been documented that it would be a useful method for the sparse data, the results of this study demonstrated that SAEM did not show higher accuracy and precision than FOCEI. One possible explanation would be the default options implemented may not support SAEM in achieving accurate and precise estimations. Interestingly, %RER for V_d and ω^2 of V_d from the one-compartment model in rich data scenario was higher than medium data scenario. This finding may have resulted from the large inter-individual variability of V_d (38%) used for the simulation [12]. This relatively high

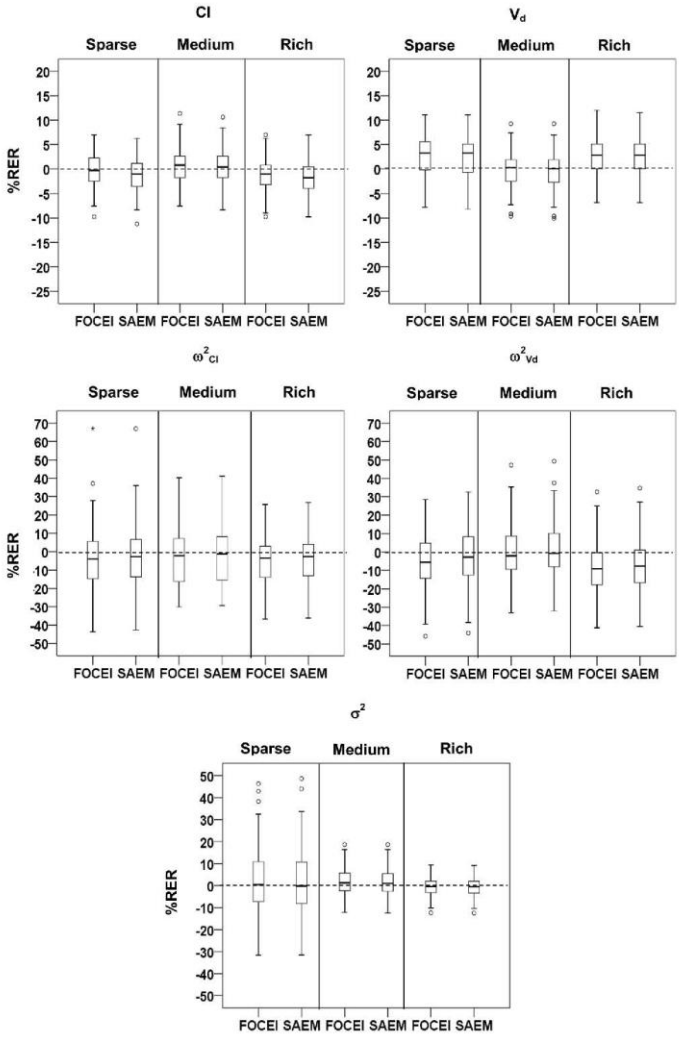

Fig. 1 %RER for five parameters from the one-compartment model across three scenarios. The box plots represent the median (middle bar), the interquartile range (box limits), and the outliers (circles and stars). *Cl* clearance, *RER* relative estimation error, V_d volume of distribution, ω^2 variance of inter-individual variability, σ^2 variance of residual variability, *FOCEI* first-order conditional estimation with interaction, *SAEM* stochastic approximation expectation maximization

variability in V_d may create the simulated data which were difficult to estimate related parameters accurately.

In these comparisons, using default options, FOCEI took much shorter time than SAEM, consistent with the most results of the previous studies [9–11]. Johansson et al. [10] showed that FOCE was the method with the shortest runtime for all various PD models. Likewise, Plan et al. [9] presented that runtimes were shortest with FOCE and LAPLACE for dose–response models. Liu et al. [11] also showed that FOCE method was faster than EM methods for classical 1-compartment model using ADVAN2 TRAN2, but not for complex models.

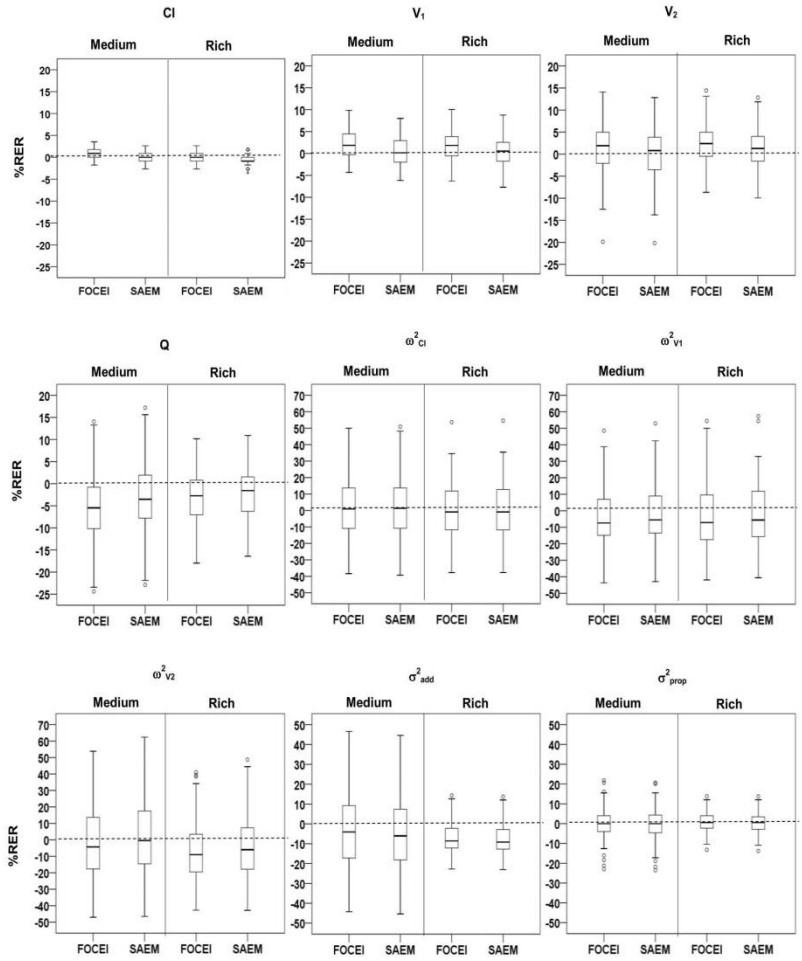

Fig. 2 %RER for nine parameters from the two-compartment model across two scenarios (%RERs from the sparse data were not shown, because the completion rates were less than 50%). The box plots represent the median (middle bar), the interquartile range (box limits), and the outliers (circles). *Cl* clearance, *RER* relative estimation error, V_1 Central volume of distribution, V_2 peripheral volume of distribution, Q intercompartmental clearance, ω^2 variance of inter-individual variability, σ^2_{add} variance of residual variability with additive model, σ^2_{prop} variance of residual variability with proportional model, *FOCEI* first-order conditional estimation with interaction, *SAEM* stochastic approximation expectation maximization

Table 3 Root mean square errors of parameter estimates from FOCEI and SAEM estimation methods

Parameter	Root mean square error					
	Sparse data		Medium data		Rich data	
	FOCEI	SAEM	FOCEI	SAEM	FOCEI	SAEM
One-compartment model						
Cl (L/h)	0.466	0.492	0.484	0.474	0.478	0.485
V_d (L)	1.016	1.004	0.769	0.785	0.913	0.896
ω^2_{Cl}	0.016	0.016	0.015	0.015	0.014	0.013
ω^2_{Vd}	0.023	0.023	0.022	0.022	0.022	0.021
σ^2_{prop}	0.001	0.001	0.001	0.001	0.0004	0.0004
Two-compartment model						
Cl (L/h)	NA	NA	0.176	0.129	0.131	0.137
V_1 (L)	NA	NA	0.244	0.209	0.223	0.199
V_2 (L)	NA	NA	0.190	0.185	0.155	0.141
Q (L/h)	NA	NA	1.244	1.131	0.801	0.733
ω^2_{Cl}	NA	NA	0.002	0.002	0.002	0.002
ω^2_{V1}	NA	NA	0.013	0.013	0.013	0.013
ω^2_{V2}	NA	NA	0.028	0.030	0.023	0.024
σ^2_{add}	NA	NA	0.710	0.717	0.367	0.376
σ^2_{prop}	NA	NA	0.001	0.001	0.001	0.001

FOCEI first-order conditional estimation with interaction, *SAEM* stochastic approximation expectation maximization, *Cl* clearance, V_d volume of distribution, V_1 central volume of distribution, V_2 peripheral volume of distribution, Q intercompartmental clearance, ω^2 variance of inter-individual variability, σ^2_{add} variance of residual variability with additive model, σ^2_{prop} variance of residual variability with proportional model, *NA* not available (the completion rates were less than 50%)

Table 4 Runtimes of FOCEI and SAEM estimation methods

Model	Runtime (mins)					
	Sparse data		Medium data		Rich data	
	FOCEI	SAEM	FOCEI	SAEM	FOCEI	SAEM
One-compartment model	0.18 (0.15–0.27)	4.64 (4.47–5.61)	0.23 (0.20–0.33)	4.81 (4.66–5.65)	0.29 (0.26–0.42)	5.01 (4.61–6.35)
Two-compartment model	0.26 (0.24–0.35)	5.59 (5.39–7.36)	0.51 (0.46–0.70)	7.15 (6.97–9.14)	0.98 (0.90–1.10)	12.03 (11.59–14.12)

Values are expressed as median (range)

FOCEI first-order conditional estimation with interaction, *SAEM* stochastic approximation expectation maximization

5 Conclusions

In this study, the performance of FOCEI and SAEM methods was evaluated using simulated data from previously published population pharmacokinetics models of piperacillin in patients. For the classical one- and two-compartment models with default options, FOCEI exhibited comparable performance similar to SAEM but with significantly shorter runtimes across sparse, medium, and rich data scenarios. Each of them showed noticeable, but comparable, bias in some parameters of both models. Practically, FOCEI would be the appropriate method for the classical one- and two-compartment models.

Compliance with Ethical Standards

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Conflict of interest The authors declare no conflicts of interest.

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APPENDIX E: Antimicrobial wild type distributions of microorganisms, version 5.26. 2018 from European Committee on Antimicrobial Susceptibility Testing (EUCAST)

MIC	Pathogens		
	Escherichia coli	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*
0.002	0	0	0
0.004	0	0	0
0.008	8	2	5
0.016	6	0	1
0.032	9	3	0
0.064	47	11	4
0.125	105	24	23
0.25	221	46	37
0.5	2978	444	453
1	16376	2539	886
2	20910	7785	3147
4	5495	5041	10479
8	1905	1802	5692
16	1233	1076	3595
32	801	560	1879
64	526	397	1506
128	752	1280	3135
256	187	336	863
512	89	209	161

APPENDIX F: Ethic Approval

เอกสารรับรองของคณะกรรมการจริยธรรมการวิจัยในมนุษย์
คณะแพทยศาสตร์ มหาวิทยาลัยสงขลานครินทร์

เอกสารเพื่อแสดงว่าคณะกรรมการจริยธรรมการวิจัยในมนุษย์ ได้พิจารณาและรับรองเอกสารที่เกี่ยวข้องกับโครงการวิจัยนี้ ดังนี้

รหัสโครงการ	:	56-501-14-1
ชื่อโครงการ (ภาษาไทย)	:	การศึกษาเภสัชจลนศาสตร์เชิงประชากรและเภสัชพลศาสตร์ของยา piperacillin/tazobactam ในระยะเริ่มต้นของผู้ที่อยู่ในภาวะพิษเหตุติดเชื้อขั้นวิกฤต
ชื่อโครงการ (ภาษาอังกฤษ)	:	Population pharmacokinetics and pharmacodynamics study of piperacillin/tazobactam during early phase in critically ill patients with severe sepsis
หัวหน้าโครงการวิจัย	:	ศาสตราจารย์นายแพทย์สุเทพ จารุรัตนศิริกุล
หน่วยงานที่สังกัด	:	ภาควิชาอายุรศาสตร์ คณะแพทยศาสตร์ มหาวิทยาลัยสงขลานครินทร์
เอกสารที่รับรอง	:	1. แบบเสนอโครงการวิจัย (Full Board Review) 2. เอกสารชี้แจงข้อมูลแก่ผู้เข้าร่วมโครงการวิจัย 3. หนังสือแสดงเจตนายินยอมเข้าร่วมการวิจัย 4. แบบบันทึกข้อมูล 5. ประวัติผู้วิจัย

คณะกรรมการจริยธรรมการวิจัยในมนุษย์ คณะแพทยศาสตร์ มหาวิทยาลัยสงขลานครินทร์ ดำเนินการให้การรับรองโครงการวิจัยตามแนวทางหลักจริยธรรมการวิจัยในมนุษย์ที่เป็นสากล ได้แก่ Declaration of Helsinki, The Belmont Report, CIOMS Guidelines และ The international Conference on Harmonization in Good Clinical Practice (ICH-GCP)

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(รองศาสตราจารย์นายแพทย์บุญสิน ตั้งตระกูลวนิช)
รองประธานคณะกรรมการพิจารณาจริยธรรมการวิจัยในมนุษย์

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PUBLICATION	1. Jaruratanasirikul S, Thengyai S, Wongpoowarak W, Wattanavijitkul T, Tangkitwanitjaroen K, Sukarnjanaset W, et al. Population pharmacokinetics and Monte Carlo dosing simulations of meropenem during the early phase of severe sepsis and septic shock in critically ill patients in intensive care units. Antimicrob Agents Chemother. 2015;59(6):2995-3001. 2. Jaruratanasirikul S, Wongpoowarak W, Wattanavijitkul T, Sukarnjanaset W, Samaeng M, Nawakitrangsan M, et al. Population Pharmacokinetics and Pharmacodynamics Modeling To Optimize Dosage Regimens of Sulbactam in Critically Ill Patients with Severe Sepsis Caused by Acinetobacter baumannii. Antimicrob Agents Chemother. 2016;60(12):7236-44. 3. Sukarnjanaset W, Wattanavijitkul T, Jarurattanasirikul S. Evaluation of FOCEI and SAEM estimation methods in population pharmacokinetic analysis using nonmem((r)) across rich, medium, and sparse sampling data. Eur J Drug Metab Pharmacokinet. 2018.