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# PipEracillin Tazobactam versus mERoPENem for treatment of bloodstream infections caused by cephalosporin-resistant Enterobacteriaceae - a non-inferiority randomized controlled trial (PeterPen)

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Page 1

# PipEracillin Tazobactam versus mERoPENem for treatment of bloodstream infections caused by cephalosporin-resistant Enterobacteriaceae - a non-inferiority randomized controlled trial (PeterPen)

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Page 3

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**Key words:** extended spectrum beta-lactamase, carbapenem, beta-lactam beta-lactamase inhibitor, randomized controlled trial

**Strengths and limitations of the study**

- The question of whether combination beta-lactam beta-lactamase inhibitors are non-inferior to carbapenems for the treatment of ESBL infections remains unanswered.

- We propose an open-label, randomized controlled trial comparing piperacillin-tazobactam with meropenem for treatment of bloodstream infections with cephalosporin-resistant *Escherichia coli* and *Klebsiella*.

- Mortality at 30-days and treatment failure at day seven are the co-primary endpoints.

- A sample size of 542 patients per arm was calculated.

Page 4

## Background

Extended-spectrum β-lactamase (ESBL) producing Enterobacteriaceae, once limited to hospital-acquired infections, have now become prevalent in the community (1) and pose a serious public health threat (2). Mortality rates following ESBL bloodstream infections (BSIs) are high, with 30-day mortality ranging from 17% in *Escherichia coli* to 34% in *Klebsiella pneumoniae* ESBL BSI in a contemporary large cohort (3), reinforcing the need for optimal treatment of these infections (4). Carbapenems have traditionally been considered the treatment of choice for Enterobacteriaceae producing ESBL or AmpC due to concerns over imprecision of phenotypic susceptibility testing and the potential of an inoculum effect (5). However extensive use of carbapenems is associated with the emergence of both carbapenemase producing and non-carbapenemase producing carbapenem-resistant Gram negative bacteria (2).

Several retrospective observational studies compared treatment with carbapenems and beta-lactam beta-lactamase inhibitors (BLBLI) for BSIs caused by ESBL-producing Enterobacteriaceae. These studies differed in the pathogens evaluated (*Klebsiella* spp. vs. *E. coli* vs. all Enterobacteriaceae), the type and dose of BLBLI or carbapenem used, the site of infection primarily assessed, whether empirical or definitive treatment was evaluated, and the outcome defined. Paterson et al were the first to demonstrate significantly lower 14-day mortality with carbapenems, establishing a dogma of carbapenem's advantage in ESBL *K. pneumoniae* BSIs more than 15 years ago (6). Studies published later were inconsistent regarding the apparent efficacy of BLBLI; however, the bulk of the published observational data show no difference between empiric or definitive treatment with BLBLIs vs. carbapenems (6–10). The MERINO trial by Harris et al was the first randomized controlled trial (RCT) to

Page 5

compare piperacillin-tazobactam (PTZ) with meropenem for ESBL-producing

Enterobacteriaceae BSI (11). This multicenter non-inferiority trial enrolled adults with

ceftriaxone-resistant (presumed ESBL-producing) *E. coli* or *Klebsiella spp.* The trial

originally targeted a sample size of 454 patients was terminated prematurely on the

third interim analysis since demonstration of non-inferiority by end of enrolment was

deemed unlikely. At termination, the overall 30-day mortality among 379 patients

included in the analysis was 7.9% (30 events), with 23/187 (12.3%) deaths in those

treated with PTZ vs. 7/191 (3.7%) in those treated with meropenem (risk difference

8.6%, 97.5% one sided confidence interval -∞ to 14.5). Thus, PTZ could not be

demonstrated to be non-inferior to meropenem. Re-calculation of the risk difference

as 2-sided 95% CI shows a significant difference between groups (risk difference 8.6

(3.3% to 14.5%)). Phenotypic ESBL production was confirmed in 86% of isolates

(85% of *E. coli* and 92.5% of *Klebsiella spp.*). Most patients had a urinary tract

infection (UTI, 60.9%) and most BSIs were caused by *E. coli* (86.5%). The risk

difference (2-sided 95% CIs) among patients with UTI (RD 3.7, 95% CI -2 to 10.7,

N=230) was lower than the risk difference among patients with a non-UTI source (RD

14.1, 95% CI 3.6-24.5, N=148). The risk difference for *Klebsiella spp.* (RD 23.1, 95%

CI 8.1-42.3, N=51) was larger than that for E. coli (RD 6.3, 95% CI 0.7-12.6, N=328).

**Rationale for replication**

While the MERINO trial was the first RCT comparing PTZ to meropenem for ESBL

bacteremia, allowing estimation of effects without selection bias, there are several

reasons justifying further RCTs. The 3-fold difference in mortality between arms is

striking, and was never observed previously in a randomized comparison between

antibiotics. Such results warrant confirmation given the profound practice

implications. Several factors in the trial design favored non-inferiority, including the

Page 6

recruitment of patients with mild sepsis (median Pitt score one at randomization, with

40.7% of patients having resolved signs of infection at randomization), relatively

short duration of the intervention (median six days out of the median 13 days of

treatment for the bacteremia) and "contamination" of drug exposure between the two

groups, due to use of the comparator for empirical treatment and stepdown therapy

after the minimal duration of the intervention of four days. Considering these, the

large difference in mortality observed between groups is even more striking.

Several factors in the MERINO trial design are worth discussion.  Primarily, the

underlying assumptions which informed the non-inferiority sample size calculation.

In MERINO, the sample size calculation assumed 14% mortality for meropenem and

10% mortality for PTZ with a 5% non-inferiority margin.  This was not included in

the initial manuscript but later appeared as an erratum (12). The *a priori* assumption

that mortality would be 4% lower for PTZ allows for a smaller total sample size, but

does so reliant on an assumption which is not supported by the observational

evidence. Removing that assumption and assuming that PTZ mortality would also be

14% (with the same one-sided alpha 2.5%, 80% power, and 10% loss to follow-up)

yields a sample size of 1683.  Therefore, the MERINO trial as conducted was

terminated after recruiting 22.5% of the sample size required under a more realistic

estimate of PTZ mortality. An underpowered non-inferiority trial is at high risk of

concluding "could not demonstrate non-inferiority".

Moreover, the interim analysis at that point (379 patients with 30 deaths), might have

occurred at a time-point allowing random overestimation of the difference (13). A

systematic review comparing trials stopped early for benefit vs. trials that tested the

same interventions but completing recruitment showed that trials stopped early for

benefit exaggerate effects, especially when the number of events is small (14,15).

Page 7

Approximately half of RCTs performed subsequent to a trial being stopped for benefit, assessing the same intervention, confirmed the terminated trial's benefit while the other half found no difference or significance in the opposite direction (16).

Authors of the MERINO trial are currently investigating the reliability of VITEK and gradient strips for determination of PTZ resistance (17) as well as the association between genetic resistance mechanisms and PTZ minimal inhibitory concentrations (MICs) (18,19). The MERINO investigators assessed PTZ MICs of 321/379 isolates by broth microdilution (BMD) in a central laboratory and found that 17.8% and 6.4% were resistant to PTZ by EUCAST and CLSI criteria, respectively (18). Also blaOXA-1 genes were highly prevalent (67%) in the MERINO trial (11). This may explain the high failure rate seen with PTZ, as co-carriage of OXA-1 and CTX-M-15 (the most common ESBL gene in the MERINO trial) is associated with PTZ MICs as high as 8-16 mcg/mL (20). These MICs, although still susceptible, have a much higher chance (up to 20%) for inadequate PTZ pharmacokinetics when using the dosing strategies employed in MERINO (21).

Other reasons for replication have been raised following the trial's publication (22). These include: imbalances between treatment groups; differences between sites with respect to the effect shown; the large number of deaths due to terminal cancer; and the pharmacokinetically non-optimized administration schedule of PTZ, particularly with respect to organisms with PTZ MICs above 2mcg/L.

We are therefore left with clinical equipoise regarding the treatment of ESBL infections with carbapenems as compared to BLBLIs. Microbiological and clinical data suggest a possible benefit to carbapenems. However, many centers do not treat patients with ESBL infections routinely with a carbapenem, due to the ecological impact on these and other patients. This is especially true for centers with high

Page 8

endemicity of carbapenem-resistant Gram-negative bacteria and high rates of ESBL

infections. Accepting without reservation the superiority of carbapenems as shown in

the MERINO trial will increase their use dramatically for the treatment of all ESBL-

positive bacteremias, spilling by default also to empirical treatment and treatment of

non-bacteremic ESBL infections. The implication of switching to a primary

carbapenem strategy for ESBLs is concerning in settings where ESBLs and

carbapenem-resistant Gram-negative bacteria are frequent. At a time of increasing

drug-resistance on one hand and on the other a serious lack of new antibiotics under

development (23), it seems imprudent to embrace the MERINO findings without

further corroboration.

For these reasons, we plan a second RCT comparing PTZ to meropenem for

bacteremia caused by third-generation cephalosporin non-susceptible *E. coli* and

*Klebsiella spp*. We aim to show the non-inferiority of PTZ to meropenem. This is a

replication trial attempting to address the findings and potential shortcomings of the

MERINO trial. Learning from the MERINO experience, we hope to also improve the

standardization of microbiological methods, baseline variable data collection, and

sample size issues.

## Methods

### Design

The study is a multicenter randomized controlled non-inferiority open-label trial.

### Study hypothesis and aims

We aim to evaluate the effect of definitive treatment with meropenem vs. PTZ, both

given as extended-infusions, on the outcome of patients with bacteremia due to PTZ

Page 9

susceptible, third-generation cephalosporin-non-susceptible *E. coli* and *Klebsiella spp.* (assumed ESBL-producing Enterobacteriaceae). We aim to demonstrate that PTZ is non-inferior to meropenem.

**Setting**

The study will be conducted in three countries: in Israel at the Rambam Health Care Campus (RHCC), Rabin Medical Center (Beilinson Hospital), Tel-Aviv Sourasky Medical Center, Soroka Medical Center, Meir Medical Center, and Sheba Medical Center; in Italy at Modena University Hospital, and in Canada at the McGill University Health Centre and Jewish General Hospital of Montreal. We are currently recruiting other centers in all study countries.

**Inclusion and exclusion criteria**

We will include adults with community or hospital-acquired monomicrobial BSI with *E. coli* or *Klebsiella spp*. non-susceptible to third generation cephalosporins and susceptible to both PTZ and meropenem. Detailed inclusion and exclusion criteria are listed in Table 1.

Inclusion will be based on antibiotic susceptibility testing performed locally (Table 2). We will ask all participating laboratories to document local MICs for PTZ and meropenem for the study patients. The index culture will be kept frozen at -70ºC for subsequent antimicrobial susceptibility confirmation and genotypic ESBL testing in a reference laboratory using optimized uniform methodology. The primary analysis will be performed as randomized (based on local susceptibility testing). A secondary analysis will be performed based on the reference laboratory susceptibility test using the EUCAST and CLSI standards that will apply at the time of analysis (24,25).

Page 10

Patients in whom an exclusion criterion arises after randomization will be included in the intention to treat population.

**Patient randomization**

Patients will be randomized to PTZ or meropenem in a 1:1 ratio. Randomization will be done by a computer-generated list of random numbers allocated centrally through a web site, stratified by country; infecting organism (*E. coli* vs. *Klebsiella spp.*); source of infection (UTI vs other); and empirical antibiotics (covering antibiotics in the first 24 hours from culture taking or non-covering). The random sequence will be generated using random permuted blocks of 4 to 8.

**Intervention**

The intervention group will receive PTZ 4.5 grams q6h and the control group will receive meropenem 1 gram q8h. Dose adjustments for patients with renal insufficiency are listed in Table 3. For both treatment arms the first dose will be administered as a 30-minute bolus and the following doses will be administered as three hours prolonged infusion. If patients receive PTZ or meropenem empirically using other dosing regimens they will switch to the trial dosing regimen, without a bolus infusion if the same antibiotic is continued.

The study drug will be administered for a minimum of four to five days to complete at least seven days of antibiotic treatment. The use of other antibiotics will not be allowed in the first week of treatment.

In order to maximize the ability of additional centers to join, minimize the study infrastructure required in each center, and contain study costs for this, as yet unfunded international trial, we have chosen to perform this trial open label. For the primary endpoint of mortality, which is objective, we do not anticipate any risk of bias. For the

Page 11

second primary endpoint, and any subjective secondary endpoints, these will be adjudicated and analyzed by blinded members of the study team.

**Pharmacokinetic / pharmacodynamic considerations**

Dosing strategies of β-lactams for patients with sepsis is a matter of debate and ongoing study. Nonetheless, studies on population pharmacokinetics for PTZ show that up to 20% of patients with an isolate with an MIC of 2mcg/L treated with 4.5g q8h by intermittent infusion will not achieve the conservative pharmacokinetic target of at least 50% of the dosing interval (50% $f$T>MIC) (21,26). Increasing the frequency to q6h improves this to about 10% at 2mcg/L but this again reaches 20% at an MIC of 8mcg/L which is still considered susceptible by both EUCAST and the CLSI (24,25). Another study evaluating therapeutic drug monitoring for β-lactams showed that bolus administration of PTZ 4.5g q6h was insufficient in up to 49% of patients to achieve the study's pharmacokinetic/pharmacodynamic target (27). Taking into consideration that patients may be obese (28), have augmented renal clearance (29) and/or have febrile neutropenia (30) only reinforces the need for high-dose extended-infusion of PTZ. A recently-published systematic review and meta-analysis on continuous/prolonged vs. intermittent infusion of β-lactams has shown reduced mortality with continuous/prolonged infusion (31), lending further support for an optimized PTZ dosing schedule in future trials.

Prior to starting this trial, we conducted a survey among interested sites regarding current and recommended dosing practices. Seven of 16 centers in Israel, Italy and Canada stated they currently use either four-daily dosing of PTZ and/or extended infusion. Two thirds recommended either 4.5g PTZ q6h extended infusion or individualized dosing (using high dose extended infusion for obese, febrile neutropenia, high MIC and severe sepsis).

Page 12

As we believe that one of the MERINO shortcomings is the sub-optimal PTZ dosing

strategy; taking into consideration the previously mentioned pharmacokinetic studies

favoring a q6h extended infusion; and realizing that some PTZ susceptibility tests are

imprecise (17,32) and we could inadvertently include patients with higher MICs; we

chose a PTZ dosing of 4.5g q6h extended infusion. While we were intrigued by

individualized dosing, we believed that since this is more complicated and might not

be applied similarly across sites, the external validity of our trial might be

compromised.

We considered a meropenem dose of 1 gram TID sufficient, since this was the dose

studied in the MERINO trial for the same indications and this was the common dose

used in the study centers. Pharmacokinetic/pharmacodynamics studies support this

dosing regimen, especially when using extended infusions (33) and for the organisms

in this study which will all be carbapenem susceptible with low MICs. We chose to

give the meropenem as extended infusion so that non-inferiority would be

demonstrated against the best case administration of meropenem.

**Outcome measures**

We defined two co-primary endpoints, the first being all-cause mortality at day 30

from randomization and the second being treatment failure at day seven from

randomization. Treatment failure was defined as death, fever above 38°C in the 48

hours before the time point, symptoms attributed to the focus of infection still present,

Sequential Failure Organ Assessment (SOFA) score (34) increasing, or blood cultures

positive with the index pathogen by the time point assessed (Table 4). These

outcomes were selected according to consensus recommendations developed for

clinical trials regarding BSIs (35).

Page 13

Secondary outcomes include all-cause mortality at 14 and 90 days; treatment failure at 14 and 30 days; microbiological failure defined as positive blood cultures with index pathogen at seven and 14 days; relapsed BSI at 30 and 90 days defined as recurrent positive blood cultures with index pathogen after prior sterilization; metastatic infections with index pathogen; secondary infections; *Clostridioides difficile* associated diarrhea; hospital re-admissions; development of resistance to study drugs in clinical isolates; carriage of carbapenem-resistant Enterobacteriaceae (carbapenemase-producing and non-producing); total in-hospital days; total antibiotic days; liver function test abnormalities; allergic reactions; renal failure and other adverse events.

Subgroup analyses will be performed for the primary outcome of 30-day mortality by infecting organism (*E. coli* vs. *Klebsiella spp.*); INCREMENT score (< 11 vs. ≥11) (36); bacteremia source (UTI vs. non-UTI); covering empirical therapy given in the first 24 hours; patients not receiving the comparator drug empirically; and excluding patients with an uncontrolled focus of infection.

**Microbiological methods**

All laboratories from centers participating in the study are ISO 9001 accredited laboratories. Following growth in blood culture, isolates will be identified using automated methods (Vitek 2, BD Phoenix, Vitek MS, MALDI biotyper). Antibiotic susceptibilities will be determined according to local practices, using either automated methods, disk diffusion or gradient diffusion methods, and interpreted using either CLSI or EUCAST breakpoints as per local protocols. All isolates will be made available for future testing by a central laboratory where antibiotic susceptibility will

Page 14

be determined using BMD and interpreted according to EUCAST standards. We will

also determine and characterize the presence of ESBL and ampC genes.

**Assessment and follow up**

All patients will be followed up till day 90 post randomization. During hospitalization

patients will be visited by infectious diseases specialists as needed. Management

decisions, such as diagnostic evaluation, other medical/surgical procedures and

discharge from hospital will be left to the discretion of the treating physicians. We

will not mandate diagnostic testing further than those defined for outcome collection

and these will be done as clinically indicated.

Data will be collected from the study visits, laboratory reports and the electronic

health record. Following discharge, we will document re-admission and survival

status through the national electronic patient files in Israel, through regional databases

in Italy, and through local data and direct patient contact (text/email/phone/mail) in

Canada. Anonymous data will be entered into a central case report form (CRF)

designed in REDCap, a secure web application.

**Sample size**

For the mortality endpoint we calculated a sample size of 542 patients per arm

assuming a 12.5% mortality rate in the control group with a 5% non-inferiority

margin and a 1-sided hypothesis with 5% $\alpha$-risk and 80% power (37). The assumed

mortality rate of 12.5% was based on rates reported in contemporary observational

studies (17.3%) (7–9) and the MERINO RCT (7.9%) (11).

The sample size calculation for the treatment failure outcome assumes a 25% failure

rate at seven days in the control group. To test for non-inferiority of PTZ compared to

Page 15

meropenem with a 1-sided 5% α-risk, 80% power and a non-inferiority margin of

10% we will need 232 patients per study group.

**Monitoring and trial management**

The trial will be monitored centrally by the coordinating center at RHCC. Data entry

will be monitored continuously on RedCap, checking for timely data entry, missing

data or suspected faulty data. Inconsistencies and logical rules have been pre-defined

to allow detection of such events. We will employ a risk-based strategy, with sparse

on-site monitoring based on central inspection of the data. A steering committee has

been nominated and the trial will be followed by an independent safety monitoring

board.

**Statistical analysis**

We plan an interim analysis after recruitment of 250, 500 and 750 patients. The trial

will be stopped if an extreme difference between groups of p<0.001 will be observed

for the primary outcome of 30-day mortality. The difference was chosen based on the

MERINO trial stopping rule (11) and following the Haybittle-Peto rule (38,39) that

preserves the overall type I error rate at 0.05. The sample size of the first interim

analysis was selected based on the minimal sample size required to reach a difference

with p<0.001 presuming that the maximal difference between groups that we will

reach is the one observed in the MERINO trial (11).

The primary analysis will include all randomized patients following local

susceptibility testing. A secondary analysis will exclude patients in whom major

errors in susceptibility compared to BMD will be detected. A per protocol analysis

will include patients fulfilling inclusion based on central lab adjudication of

# Page 16

susceptibilities, without exclusion criteria and receiving the allocated intervention for at least four calendar days.

Patients' baseline characteristics will be displayed descriptively. Outcome variables will be compared using the chi-square test, Student's t-test or the Mann–Whitney U test, as appropriate. Risk differences for dichotomous outcomes will be computed with 95% confidence intervals. Non-inferiority will be fulfilled if the upper value of the 1-sided 95% CI for the risk difference of meropenem compared to PTZ will be equal or lower to the defined non-inferiority margin.

## Ethics

The ethics of recruiting patients into this study, after the MERINO trial, are embedded in the considerations we previously raised. These concern the possibility that their chance finding will not be observed in a larger repetition trial and some improvement in the study design through obtaining a larger sample size and improving PTZ pharmacokinetics. With these considerations, the study was approved by the ethics committees of the above Israeli hospitals and is awaiting approval in other hospitals. In Canada, institutional ethics approval has been provisionally granted and the study will commence after Health Canada approval has been granted for a study involving off-label use of approved pharmaceuticals.

## Patient and public involvement

We have not involved patients or the public in the trial's design and planning.

## Funding

We have not succeeded in obtaining funding for the study from the Israeli Ministry of Health. The study received the support of ESCMID through the society's Research Grant programme 2020 (30,000 Euro) which will be used for the ethics and insurance

Page 17

requirements of the Italian sites and for onsite study visits. The Canadian sites are

seeking funding through the Canadian Institutes of Health Research. Should the study

receive substantial funding we plan to recruit additional hospitals who do not have the

infrastructure to recruit into clinical trials without specific funding and to revise the

protocol, mainly with respect to the microbiological methods so as to enable local

laboratories to perform BMD and test for resistance genes in real time.

**Competing interests**

None declared.

**Authors' contributions**

All authors contributed to conception, design, trial management and planned data

analysis.

RB, FK, DY and MP contributed to trial database and randomization site design.

RB and MP wrote the first draft of the manuscript.

All authors revised the protocol critically for important intellectual content and

approved the final manuscript.

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Page 18

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Page 19

**Table 1:** Inclusion and exclusion criteria

| Inclusion criteria | Exclusion criteria |
|---|---|
| • Adults (age ≥ 18 years)<br><br>• New onset BSI due to *E. coli* or *Klebsiella spp.* in one or more blood cultures associated with evidence of infection<br><br>• The microorganism will have to be non-susceptible to third generation cephalosporins (ceftriaxone and/or ceftazidime) and susceptible to both PTZ and meropenem<br><br>• We will permit the inclusion of bacteremias due to *E. coli* or *Klebsiella spp.* with concomitant growth in blood of skin commensals considered as contaminants. | • More than 72 hours elapsed since initial blood culture taken, regardless of the time covering antibiotics were started<br><br>• Polymicrobial bacteremia defined as either growth of two or more different species of microorganisms in the same blood culture, or growth of different species in two or more separate blood cultures within the same episode of infection<br><br>• Patients with prior bacteremia or infection that have not completed antimicrobial therapy for the previous infectious episode.<br><br>• Patients with septic shock at the time of enrollment and randomization, defined as at least 2 measurements of systolic blood pressure < 90 mmHg and/or use of vasopressors (dopamine>15μg/kg/min, adrenalin>0.1μg/kg/min, |

Page 20

| | noradrenalin>0.1µg/kg/min, vasopressin any dose) in the 12 hours prior to randomization. In the absence of the use of vasopressors, a systolic blood pressure <90 would need to represent a deviation for the patient's known normal blood pressure. |
| | • BSI due to specific infections known at the time of randomization: endocarditis / endovascular infections, osteomyelitis (not resected), central nervous system infections |
| | • Allergy to any of the study drugs confirmed by history taken by the investigator |
| | • Previous enrollment in this trial |
| | • Concurrent participation in another interventional clinical trial |
| | • Imminent death (researcher's assessment of expected death within 48 hours of recruitment after discussion with treating team) |

Page 21

**Table 2:** CLSI and EUCAST breakpoint definitions for susceptibility

|  | CLSI M100-ED28: 2018. 28th Edition (25) | | EUCAST v 9 (January, 2019) (24) | |
| --- | --- | --- | --- | --- |
|  | MIC (mg/L) | Disk diffusion (mm) | MIC (mg/L) | Disk diffusion (mm) |
| Ceftriaxone | ≤1 | ≥23 | ≤1 | ≥25 |
| Ceftazidime | ≤4 | ≥21 | ≤1 | ≥22 |
| PTZ | ≤16 | ≥21 | ≤8 | ≥20 |
| Meropenem | ≤1 | ≥23 | ≤2 | ≥22 |
| Imipenem | ≤1 | ≥23 | ≤2 | ≥22 |

CLSI- clinical and laboratory standards institute; EUCAST- European committee on antimicrobial susceptibility testing; MIC- minimal inhibitory concentration

Page 22

**Table 3**: Dose adjustment for study antibiotics

|  | Meropenem | Piperacillin tazobactam |
|---|---|---|
| CrCl>50ml/min* | 1g q8h | 4.5g q6h |
| CrCl 26-50ml/min* | 1g q12h | 3.375g q6h (only if CCT<40) |
| CrCl 10-25ml/min* | 0.5g q12h | 2.25g q6h |
| CrCl<10ml/min* | 0.5g q24h | 2.25g q6h |
| Hemodialysis | 0.5g q24h (+0.5g AD) | 2.25g q8h (+0.75g AD) |
| Peritoneal dialysis | 0.5g q24h | 2.25g q8h |
| Continuous renal replacement therapy | 1g q12h | 4.5g q8h |

*CrCl should be expressed in mL/min/1.73m2, using the modification of diet in renal

disease (MDRD) formula, Cockroft and Gault equation or other means.

AD- after dialysis

In Canada, to conform with the existing product monograph and accounting for the

unavailability of the 3.375g dosage form in most hospitals the following piperacillin-

tazobactam dosing strategy will be used (as extended infusion of 3 hours).

|  | Piperacillin-tazobactam |
|---|---|
| CCT>40ml/min | 4.5g QID |
| CCT 20-40ml/min | 4.5g TID |
| CCT 10-20ml/min | 2.25g QID |
| CCT<10ml/min | 2.25g QID |
| Hemodialysis | 2.25g TID (+0.75g AD) |
| Peritoneal dialysis | 2.25g TID |
| Continuous renal replacement therapy | 4.5g TID |

Page 23

**Table 4:** Outcomes

| Outcome | Definition |
|---------|------------|
| 30-day all-cause mortality (co-primary outcome) | |
| Treatment failure at day 7 (co-primary outcome) | Composite of the following by day 7: <br> • Death <br> • Fever above 38°C in the last 48 hours <br> • Symptoms attributed to the focus of infection still present <br> • SOFA score increasing <br> • Blood cultures positive with the index pathogen |
| 14- and 90-day all-cause mortality | |
| Treatment failure at 14 & 30 days | As defined above |
| Microbiological failure at 7 & 14 days | Positive blood cultures with index pathogen at days 4-7 and 11-14 |
| Relapsed BSI at 30 & 90 days | Positive blood cultures with index pathogen following prior sterilization at days 30 and 90 |
| Metastatic focus of infection | Isolation of index pathogen from non-blood specimen related to metastatic spread of infection by day 90 |

Page 24

| Superinfection | Development of either clinically or microbiologically documented infection within 90 days according to CDC surveillance definitions of health-care associated infections for bacterial infections |
|---|---|
| Resistant infection | Clinical isolates resistant to PTZ and meropenem and any carbapenem-resistant bacteria |
| Resistant colonization | Carriage of CPE and non-CPE CRE in-hospital till day 90, detected by weekly rectal surveillance of carriage while in-hospital |
| Re-admissions | Number of hospital re-admissions until day 90 |
| CDI | *Clostridioides difficile* associated diarrhea till 90 days |
| Adverse events | • Abnormal liver enzymes and bilirubin<br>• Renal failure using the RIFLE (40) criteria by day 30 but we will not rely on urine output because it is not properly or accurately documented in many non-ICU inpatient units<br>• Leukopenia, neutropenia, thrombocytopenia<br>• Drug hypersensitivity<br>• Diarrhea<br>• Seizures |

SOFA, Sequential Organ Failure Assessment; BSI, bloodstream infection; CDC, Centers for Disease Control and Prevention; PTZ, piperacillin tazobactam; CPE, cabapenemase-producing Enterobacteriaceae; CRE, Carbapenem-resistant Enterobacteriaceae; CDI, *Clostridioides difficile* infection

Page 25

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**SPIRIT** ☑

STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

| Section/item | Item No | Description |
|---|---|---|
| **Administrative information** | | |
| Done Title | 1 | Descriptive title identifying the study design, population, interventions, and, if applicable, trial acronym |
| Done Trial registration | 2a | Trial identifier and registry name. If not yet registered, name of intended registry |
| | 2b | All items from the World Health Organization Trial Registration Data Set |
| N/R Protocol version | 3 | Date and version identifier |
| Done Funding | 4 | Sources and types of financial, material, and other support |
| Done Roles and responsibilities | 5a | Names, affiliations, and roles of protocol contributors |
| | 5b | Name and contact information for the trial sponsor |
| | 5c | Role of study sponsor and funders, if any, in study design; collection, management, analysis, and interpretation of data; writing of the report; and the decision to submit the report for publication, including whether they will have ultimate authority over any of these activities |
| | 5d | Composition, roles, and responsibilities of the coordinating centre, steering committee, endpoint adjudication committee, data management team, and other individuals or groups overseeing the trial, if applicable (see Item 21a for data monitoring committee) |
| **Introduction** | | |
| Done Background and rationale | 6a | Description of research question and justification for undertaking the trial, including summary of relevant studies (published and unpublished) examining benefits and harms for each intervention |
| | 6b | Explanation for choice of comparators |
| Done Objectives | 7 | Specific objectives or hypotheses |

Done Trial design  8  Description of trial design including type of trial (eg, parallel group, crossover, factorial, single group), allocation ratio, and framework (eg, superiority, equivalence, noninferiority, exploratory)

**Methods: Participants, interventions, and outcomes**

Done Study setting  9  Description of study settings (eg, community clinic, academic hospital) and list of countries where data will be collected. Reference to where list of study sites can be obtained

Done Eligibility criteria  10  Inclusion and exclusion criteria for participants. If applicable, eligibility criteria for study centres and individuals who will perform the interventions (eg, surgeons, psychotherapists)

Done Interventions  11a  Interventions for each group with sufficient detail to allow replication, including how and when they will be administered

11b  Criteria for discontinuing or modifying allocated interventions for a given trial participant (eg, drug dose change in response to harms, participant request, or improving/worsening disease)

11c  Strategies to improve adherence to intervention protocols, and any procedures for monitoring adherence (eg, drug tablet return, laboratory tests)

11d  Relevant concomitant care and interventions that are permitted or prohibited during the trial

Done Outcomes  12  Primary, secondary, and other outcomes, including the specific measurement variable (eg, systolic blood pressure), analysis metric (eg, change from baseline, final value, time to event), method of aggregation (eg, median, proportion), and time point for each outcome. Explanation of the clinical relevance of chosen efficacy and harm outcomes is strongly recommended

Done Participant timeline  13  Time schedule of enrolment, interventions (including any run-ins and washouts), assessments, and visits for participants. A schematic diagram is highly recommended (see Figure)

Done Sample size  14  Estimated number of participants needed to achieve study objectives and how it was determined, including clinical and statistical assumptions supporting any sample size calculations

Done Recruitment  15  Strategies for achieving adequate participant enrolment to reach target sample size

**Methods: Assignment of interventions (for controlled trials)**

Allocation:

| | | |
|---|---|---|
| Done Sequence generation | 16a | Method of generating the allocation sequence (eg, computer-generated random numbers), and list of any factors for stratification. To reduce predictability of a random sequence, details of any planned restriction (eg, blocking) should be provided in a separate document that is unavailable to those who enrol participants or assign interventions |
| Done Allocation concealment mechanism | 16b | Mechanism of implementing the allocation sequence (eg, central telephone; sequentially numbered, opaque, sealed envelopes), describing any steps to conceal the sequence until interventions are assigned |
| Done Implementation | 16c | Who will generate the allocation sequence, who will enrol participants, and who will assign participants to interventions |
| N/R Blinding (masking) | 17a | Who will be blinded after assignment to interventions (eg, trial participants, care providers, outcome assessors, data analysts), and how |
| | 17b | If blinded, circumstances under which unblinding is permissible, and procedure for revealing a participant's allocated intervention during the trial |

**Methods: Data collection, management, and analysis**

| | | |
|---|---|---|
| Done Data collection methods | 18a | Plans for assessment and collection of outcome, baseline, and other trial data, including any related processes to promote data quality (eg, duplicate measurements, training of assessors) and a description of study instruments (eg, questionnaires, laboratory tests) along with their reliability and validity, if known. Reference to where data collection forms can be found, if not in the protocol |
| | 18b | Plans to promote participant retention and complete follow-up, including list of any outcome data to be collected for participants who discontinue or deviate from intervention protocols |
| Done Data management | 19 | Plans for data entry, coding, security, and storage, including any related processes to promote data quality (eg, double data entry; range checks for data values). Reference to where details of data management procedures can be found, if not in the protocol |
| Done Statistical methods | 20a | Statistical methods for analysing primary and secondary outcomes. Reference to where other details of the statistical analysis plan can be found, if not in the protocol |
| | 20b | Methods for any additional analyses (eg, subgroup and adjusted analyses) |
| | 20c | Definition of analysis population relating to protocol non-adherence (eg, as randomised analysis), and any statistical methods to handle missing data (eg, multiple imputation) |

**Methods: Monitoring**

| | | |
|---|---|---|
| Done Data monitoring | 21a | Composition of data monitoring committee (DMC); summary of its role and reporting structure; statement of whether it is independent from the sponsor and competing interests; and reference to where further details about its charter can be found, if not in the protocol. Alternatively, an explanation of why a DMC is not needed |
| | 21b | Description of any interim analyses and stopping guidelines, including who will have access to these interim results and make the final decision to terminate the trial |
| Done Harms | 22 | Plans for collecting, assessing, reporting, and managing solicited and spontaneously reported adverse events and other unintended effects of trial interventions or trial conduct |
| Done Auditing | 23 | Frequency and procedures for auditing trial conduct, if any, and whether the process will be independent from investigators and the sponsor |

**Ethics and dissemination**

| | | |
|---|---|---|
| Done Research ethics approval | 24 | Plans for seeking research ethics committee/institutional review board (REC/IRB) approval |
| N/R Protocol amendments | 25 | Plans for communicating important protocol modifications (eg, changes to eligibility criteria, outcomes, analyses) to relevant parties (eg, investigators, REC/IRBs, trial participants, trial registries, journals, regulators) |
| Done Consent or assent | 26a | Who will obtain informed consent or assent from potential trial participants or authorised surrogates, and how (see Item 32) |
| | 26b | Additional consent provisions for collection and use of participant data and biological specimens in ancillary studies, if applicable |
| Done Confidentiality | 27 | How personal information about potential and enrolled participants will be collected, shared, and maintained in order to protect confidentiality before, during, and after the trial |
| Done Declaration of interests | 28 | Financial and other competing interests for principal investigators for the overall trial and each study site |
| Done Access to data | 29 | Statement of who will have access to the final trial dataset, and disclosure of contractual agreements that limit such access for investigators |
| N/R Ancillary and post-trial care | 30 | Provisions, if any, for ancillary and post-trial care, and for compensation to those who suffer harm from trial participation |

Done
Dissemination
policy

31a    Plans for investigators and sponsor to communicate trial results to participants, healthcare professionals, the public, and other relevant groups (eg, via publication, reporting in results databases, or other data sharing arrangements), including any publication restrictions

31b    Authorship eligibility guidelines and any intended use of professional writers

31c    Plans, if any, for granting public access to the full protocol, participant-level dataset, and statistical code

**Appendices**

N/R Informed consent materials

32    Model consent form and other related documentation given to participants and authorised surrogates

Done Biological specimens

33    Plans for collection, laboratory evaluation, and storage of biological specimens for genetic or molecular analysis in the current trial and for future use in ancillary studies, if applicable

*It is strongly recommended that this checklist be read in conjunction with the SPIRIT 2013 Explanation & Elaboration for important clarification on the items. Amendments to the protocol should be tracked and dated. The SPIRIT checklist is copyrighted by the SPIRIT Group under the Creative Commons "Attribution-NonCommercial-NoDerivs 3.0 Unported" license.

# PipEracillin Tazobactam versus mERoPENem for treatment of bloodstream infections caused by third generation cephalosporin-resistant Enterobacteriaceae – a study protocol for a non-inferiority open label randomized controlled trial (PeterPen)

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Page 1

# PipEracillin Tazobactam versus mERoPENem for treatment of bloodstream infections caused by third generation cephalosporin-resistant Enterobacteriaceae – a study protocol for a non-inferiority open label randomized controlled trial (PeterPen)

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Page 3

## Abstract

**Introduction:** The optimal treatment for ESBL-producing Enterobacteriaceae bloodstream infections has yet to be defined. Retrospective studies have shown conflicting results, with most data suggesting the non-inferiority of beta-lactam beta-lactamase inhibitor combinations compared to carbapenems. However, the recently published MERINO trial failed to demonstrate the non-inferiority of piperacillin-tazobactam to meropen. The potential implications of the MERINO trial are profound, as widespread adoption of carbapenem treatment will have detrimental effects on antimicrobial stewardship in areas endemic for ESBL and carbapenem-resistant bacteria. Therefore, we believe that it is justified to re-examine the comparison in a second randomized controlled trial prior to changing clinical practice.

**Methods and analysis:** PeterPen is a multicenter, investigator-initiated, open-label, randomized controlled non-inferiority trial, comparing piperacillin-tazobactam with meropenem for third generation cephalosporin-resistant *Escherichia coli* and *Klebsiella* bloodstream infections. The study is currently being conducted in 6 centers in Israel and 1 in Canada with other centers from Israel, Italy and Canada expected to join. The two primary outcomes are all-cause mortality at day 30 from enrollment and treatment failure at day seven (death, fever above 38°C in the last 48 hours, continuous symptoms, increasing Sequential Organ Failure Assessment (SOFA) score, or persistent blood cultures with the index pathogen). A sample size of 1084 patients was calculated for the mortality end point assuming a 12.5% mortality rate in the control group with a 5% non-inferiority margin and assuming 100% follow-up for this outcome.

Page 4

**Ethics and dissemination:** The study is approved by local and national ethics

committees as required. Results will be published and trial data will be made

available.

**Trial registration:** ClinicalTrials.gov NCT 03671967 registered 13 September 2018;

Israeli Ministry of Health trials register MOH_2018-12-25_004857 registered 25

December 2018.

**Key words:** extended spectrum beta-lactamase, carbapenem, beta-lactam beta-

lactamase inhibitor, randomized controlled trial

**Strengths and limitations of the study**

- The study addresses a question of critical importance to antibiotic stewardship.

- Assuming the sample size estimates are correct, this pragmatic randomized

  controlled trial will provide a more definitive answer.

- Susceptibilities determined by automated methods may underestimate

  piperacillin-tazobactam resistance and resistance genes will not be available in

  real-time. Hence there will be a small risk of misclassified patients.

- Antibiotic levels will not be tested to direct dosing; however extended infusion

  regimens have been chosen to match high expected predicted target attainment

  for most patients.

- The study will reflect current standard of care provided to patients

## Background

Extended-spectrum β-lactamase (ESBL) producing Enterobacteriaceae, once limited to hospital-acquired infections, have now become prevalent in the community [1] and pose a serious public health threat [2]. Mortality rates following ESBL bloodstream infections (BSIs) are high, with 30-day mortality ranging from 17% in *Escherichia coli* to 34% in *Klebsiella pneumoniae* ESBL BSI in a contemporary large cohort [3], reinforcing the need for optimal treatment of these infections [4]. Carbapenems have traditionally been considered the treatment of choice for Enterobacteriaceae producing ESBL or AmpC due to concerns over imprecision of phenotypic susceptibility testing and the potential of an inoculum effect [5]. However extensive use of carbapenems is associated with the emergence of both carbapenemase producing and non-carbapenemase producing carbapenem-resistant Gram negative bacteria [2].

Several retrospective observational studies compared treatment with carbapenems and beta-lactam beta-lactamase inhibitors (BLBLI) for BSIs caused by ESBL-producing Enterobacteriaceae. These studies differed in the pathogens evaluated (*Klebsiella* spp. vs. *E. coli* vs. all Enterobacteriaceae), the type and dose of BLBLI or carbapenem used, the site of infection primarily assessed, whether empirical or definitive treatment was evaluated, and the outcome defined. Paterson et al were the first to demonstrate significantly lower 14-day mortality with carbapenems, establishing the dogma of a carbapenem advantage in ESBL *K. pneumoniae* BSIs more than 15 years ago [6]. Studies published later were inconsistent regarding the apparent efficacy of BLBLI; however, the bulk of the published observational data show no difference between empiric or definitive treatment with BLBLIs vs. carbapenems [6–10]. The MERINO trial by Harris et al was the first randomized controlled trial (RCT) to

Page 6

compare piperacillin-tazobactam (PTZ) with meropenem for ESBL-producing

Enterobacteriaceae BSI [11]. This multicenter non-inferiority trial enrolled adults with

ceftriaxone-resistant (presumed ESBL-producing) *E. coli* or *Klebsiella spp.* The trial

originally targeted a sample size of 454 patients and was terminated prematurely on

the third interim analysis since demonstration of non-inferiority by end of enrolment

was deemed unlikely. At termination, the overall 30-day mortality among 379 patients

included in the analysis was 7.9% (30 events), with 23/187 (12.3%) deaths in those

treated with PTZ vs. 7/191 (3.7%) in those treated with meropenem (risk difference

8.6%, 97.5% one sided confidence interval -∞ to 14.5). Thus, PTZ could not be

demonstrated to be non-inferior to meropenem. Re-calculation of the risk difference

as 2-sided 95% CI shows a significant difference between groups (risk difference 8.6

(3.3% to 14.5%)). Most deaths were related to underlying cancer. Phenotypic ESBL

production was confirmed in 86% of isolates (85% of *E. coli* and 92.5% of *Klebsiella*

*spp.*). Most patients had a urinary tract infection (UTI, 60.9%) and most BSIs were

caused by *E. coli* (86.5%). The risk difference (2-sided 95% CIs) among patients with

UTI (RD 3.7, 95% CI -2 to 10.7, N=230) was lower than the risk difference among

patients with a non-UTI source (RD 14.1, 95% CI 3.6-24.5, N=148). The risk

difference for *Klebsiella spp.* (RD 23.1, 95% CI 8.1-42.3, N=51) was larger than that

for E. coli (RD 6.3, 95% CI 0.7-12.6, N=328).

**Rationale for replication**

While the MERINO trial was the first RCT comparing PTZ to meropenem for ESBL

bacteremia, allowing estimation of effects without selection bias, there are several

reasons justifying further RCTs. The 3-fold difference in mortality between arms is

striking and such a mortality difference was never observed previously in a

randomized comparison between antibiotics. Such results warrant confirmation given

## Page 7

the profound practice implications. Several factors in the trial design favored non-inferiority, including the recruitment of patients with mild sepsis (median Pitt score one at randomization, with 40.7% of patients having resolved signs of infection at randomization), relatively short duration of the intervention (median six days out of the median 13 days of treatment for the bacteremia) and "contamination" of drug exposure between the two groups, due to use of the comparator for empirical treatment and stepdown therapy after the minimal duration of the intervention of four days. Considering these, the large difference in mortality observed between groups is even more surprising.

Several factors in the MERINO trial design are worth discussion. Primarily, the underlying assumptions which informed the non-inferiority sample size calculation. In MERINO, the sample size calculation assumed 14% mortality for meropenem and 10% mortality for PTZ with a 5% non-inferiority margin. This was not included in the initial manuscript but later appeared as an erratum [12]. The *a priori* assumption that mortality would be 4% lower for PTZ allows for a smaller total sample size but does so reliant on an assumption which is not supported by the observational evidence. Removing that assumption and assuming that PTZ mortality would also be 14% (with the same one-sided alpha 2.5%, 80% power, and 10% loss to follow-up) yields a sample size of 1683. Therefore, the MERINO trial as conducted was terminated after recruiting 22.5% of the sample size required under a more realistic estimate of PTZ mortality. An underpowered non-inferiority trial is at high risk of concluding "could not demonstrate non-inferiority".

Moreover, the interim analysis at that point (379 patients with 30 deaths), might have occurred at a time-point allowing random overestimation of the difference [13]. A systematic review comparing trials stopped early for benefit vs. trials that tested the

Page 8

same interventions but completing recruitment showed that trials stopped early for

benefit exaggerate effects, especially when the number of events is small [14,15].

Approximately half of RCTs performed subsequent to a trial being stopped for

benefit, assessing the same intervention, confirmed the terminated trial's benefit while

the other half found no difference or significance in the opposite direction [16].

Authors of the MERINO trial are currently investigating the reliability of VITEK and

gradient strips for determination of PTZ resistance [17] as well as the association

between genetic resistance mechanisms and PTZ minimal inhibitory concentrations

(MICs) [18,19]. The MERINO investigators assessed PTZ MICs of 321/379 isolates

by broth microdilution (BMD) in a central laboratory and found that 17.8% and 6.4%

were resistant to PTZ by EUCAST and CLSI criteria, respectively [18]. Also

blaOXA-1 genes were highly prevalent (67%) in the MERINO trial [11]. This may

explain the high failure rate seen with PTZ, as co-carriage of OXA-1 and CTX-M-15

(the most common ESBL gene in the MERINO trial) is associated with PTZ MICs as

high as 8-16 mcg/mL [20]. These MICs, although still susceptible, have a much

higher chance (up to 20%) for inadequate PTZ pharmacokinetics when using the

dosing strategies employed in MERINO [21].

Other reasons for replication have been raised following the trial's publication [22].

These include: imbalances between treatment groups; differences between sites with

respect to the effect shown; the large number of deaths due to terminal cancer; and the

pharmacokinetically non-optimized administration schedule of PTZ, particularly with

respect to organisms with PTZ MICs above 2mcg/L.

We are therefore left with clinical equipoise regarding the treatment of ESBL

infections with carbapenems as compared to BLBLIs. Microbiological and clinical

trial data suggest a possible benefit to carbapenems. However, many centers do not

Page 9

treat patients with ESBL infections routinely with a carbapenem, due to the ecological

impact on these and other patients. This is especially true for centers with high

endemicity of carbapenem-resistant Gram-negative bacteria and high rates of ESBL

infections. Accepting without reservation the superiority of carbapenems based on the

MERINO trial will increase their use dramatically for the treatment of all ESBL-

positive bacteremias, spilling by default also to empirical treatment and treatment of

non-bacteremic ESBL infections. The implication of switching to a primary

carbapenem strategy for ESBLs is concerning in settings where ESBLs and

carbapenem-resistant Gram-negative bacteria are frequent. At a time of increasing

drug-resistance on one hand and on the other a serious lack of new antibiotics under

development [23], it seems imprudent to embrace the MERINO findings without

further corroboration.

For these reasons, we plan a second RCT comparing PTZ to meropenem for

bacteremia caused by third-generation cephalosporin non-susceptible *E. coli* and

*Klebsiella spp*. We aim to show non-inferiority of PTZ to meropenem. This is a

replication trial attempting to address the findings and potential shortcomings of the

MERINO trial. Learning from the MERINO experience, we hope to also improve the

standardization of microbiological methods, baseline variable data collection, and

sample size issues.

## Methods

### Design

The study is a multicenter randomized controlled non-inferiority open-label trial.

### Study hypothesis and aims

Page 10

We aim to evaluate the effect of definitive treatment with meropenem vs. PTZ, both given as extended-infusions, on the outcome of patients with bacteremia due to PTZ susceptible, third-generation cephalosporin-non-susceptible *E. coli* and *Klebsiella spp.* (assumed ESBL-producing Enterobacteriaceae). We aim to demonstrate that PTZ is non-inferior to meropenem.

**Setting**

The study will be conducted in three countries: in Israel at the Rambam Health Care Campus (RHCC), Rabin Medical Center (Beilinson Hospital), Tel-Aviv Sourasky Medical Center, Soroka Medical Center, Meir Medical Center, and Sheba Medical Center; in Italy at Modena University Hospital, and in Canada at the McGill University Health Centre and the Jewish General Hospital. We are currently recruiting other centers in all study countries. RHCC is the sponsor and assumes responsibility for the trial.

**Inclusion and exclusion criteria**

We will include adults with community or hospital-acquired monomicrobial BSI with *E. coli* or *Klebsiella spp.* non-susceptible to third generation cephalosporins and susceptible to both PTZ and meropenem. Detailed inclusion and exclusion criteria are listed in Table 1. Patients in whom exclusion criteria arise after randomization will be included in the intention to treat population.

Inclusion will be based on antibiotic susceptibility testing performed locally (Table 2). We will ask all participating laboratories to document local MICs for PTZ and meropenem for the study patients. The index culture will be kept frozen at -70ºC for subsequent antimicrobial susceptibility confirmation and genotypic ESBL testing in a reference laboratory using optimized uniform methodology including BMD. The

Page 11

primary analysis will be performed as randomized (based on local susceptibility

testing). A secondary analysis will be performed based on the reference laboratory

susceptibility test using the EUCAST and CLSI standards that will apply at the time

of analysis [24,25].

**Table 2:** CLSI and EUCAST breakpoint definitions for susceptibility

|  | CLSI M100-ED28: 2018. 28th Edition [25] | | EUCAST v 9 (January, 2019) [24] | |
|---|---|---|---|---|
|  | MIC (mg/L) | Disk diffusion (mm) | MIC (mg/L) | Disk diffusion (mm) |
| Ceftriaxone | ≤1 | ≥23 | ≤1 | ≥25 |
| Ceftazidime | ≤4 | ≥21 | ≤1 | ≥22 |
| PTZ | ≤16 | ≥21 | ≤8 | ≥20 |
| Meropenem | ≤1 | ≥23 | ≤2 | ≥22 |
| Imipenem | ≤1 | ≥23 | ≤2 | ≥22 |

CLSI- clinical and laboratory standards institute; EUCAST- European committee on

antimicrobial susceptibility testing; MIC- minimal inhibitory concentration

**Patient randomization**

Patients will be randomized to PTZ or meropenem in a 1:1 ratio. Randomization will

be done by a computer-generated list of random numbers allocated centrally in

REDCap [26], stratified by country; infecting organism (*E. coli* vs. *Klebsiella spp.*);

source of infection (UTI vs other); and empirical antibiotics (covering antibiotics in

the first 24 hours from culture taken or non-covering). The random sequence will be

generated using random permuted blocks of 4 to 8.

**Intervention**

Page 12

The intervention group will receive PTZ 4.5 grams q6h and the control group will

receive meropenem 1 gram q8h. Dose adjustments for patients with renal

insufficiency are listed in Table 3. For each treatment arm, the first dose will be

administered as a 30-minute bolus and the following doses will be administered as

three hours prolonged infusion. If patients receive PTZ or meropenem empirically

using other dosing regimens they will switch to the trial dosing regimen, without a

bolus infusion if the same antibiotic is continued.

The study drug will be administered for a minimum of four to five days to complete at

least seven days of antibiotic treatment. We will make a great effort to ensure that

patients will complete treatment with the assigned treatment arm. Switch to the

alternate arm antibiotic class or other antibiotics will not be permitted in the first week

of treatment, unless treatment fails or for secondary infections. Crossovers, if they

occur will be analyzed using appropriate statistical methods [27].

In order to maximize the ability of additional centers to join, minimize the study

infrastructure required in each center, and contain study costs for this, as yet unfunded

international trial, we have chosen to perform this trial open label. This is also

essential as blinding a q8h drug vs. a q6h drug mathematically challenging. For the

primary endpoint of mortality, which is objective, we do not anticipate risk of

detection bias. The second primary endpoint, and any subjective secondary endpoints,

will be adjudicated and analyzed by blinded members of the study team based on

discrete variables collected.

**Pharmacokinetic / pharmacodynamic considerations**

Dosing strategies of β-lactams for patients with sepsis is a matter of debate and

ongoing study. Nonetheless, studies on population pharmacokinetics for PTZ show

that up to 20% of patients with an isolate with an MIC of 2mcg/L treated with 4.5g

Page 13

q8h by intermittent infusion will not achieve the conservative pharmacokinetic target of at least 50% of the dosing interval (50% ƒT>MIC) [21,28]. Increasing the frequency to q6h improves this to about 10% at 2mcg/L but this again reaches 20% at an MIC of 8mcg/L which is still considered susceptible by both EUCAST and the CLSI [24,25]. Another study evaluating therapeutic drug monitoring for β-lactams showed that bolus administration of PTZ 4.5g q6h was insufficient in up to 49% of patients to achieve the study's pharmacokinetic/pharmacodynamic target [29]. Taking into consideration that patients may be obese [30], have augmented renal clearance [31] and/or have febrile neutropenia [32] only reinforces the need for high-dose extended-infusion of PTZ. A recently-published systematic review and meta-analysis on continuous/prolonged vs. intermittent infusion of β-lactams has shown reduced mortality with continuous/prolonged infusion [33], lending further support for an optimized PTZ dosing schedule in future trials. Dosing for patients with continuous renal replacement therapy by type of dialysis and flow rate; we based dosing on a contemporary literature review [34].

Prior to starting this trial, we conducted a survey among interested sites regarding current and recommended dosing practices. Seven of 16 centers in Israel, Italy and Canada stated they currently use either four-daily dosing of PTZ and/or extended infusion. Two thirds recommended either 4.5g PTZ q6h extended infusion or individualized dosing (using high dose extended infusion for obese, febrile neutropenia, high MIC and severe sepsis).

As we believe that one of the MERINO shortcomings is the sub-optimal PTZ dosing strategy; taking into consideration the previously mentioned pharmacokinetic studies favoring a q6h extended infusion; and realizing that some PTZ susceptibility tests are imprecise [17,35] and we could inadvertently include patients with higher MICs; we

Page 14

chose a PTZ dosing of 4.5g q6h extended infusion. While we were intrigued by

individualized dosing, we believed that since this is more complicated and might not

be applied similarly across sites, the external validity of our trial might be

compromised.

We considered a meropenem dose of 1 gram TID sufficient, since this was the dose

studied in the MERINO trial for the same indications and this was the common dose

used in the study centers. Pharmacokinetic/pharmacodynamics studies support this

dosing regimen, especially when using extended infusions [36] and for the organisms

in this study which will all be carbapenem susceptible with low MICs. We chose to

give the meropenem as extended infusion so that non-inferiority would be

demonstrated against the best-case administration of meropenem.

**Outcome measures**

We defined two co-primary endpoints, the first being all-cause mortality at day 30

from randomization and the second being treatment failure at day seven from

randomization. Treatment failure was defined as death, fever above 38°C in the 48

hours before the time point, symptoms attributed to the focus of infection still present,

Sequential Failure Organ Assessment (SOFA) score [37] increasing, or blood cultures

positive with the index pathogen by the time point assessed (Table 4). These

outcomes were selected according to consensus recommendations for endpoints in

clinical trials regarding BSIs [38].

Secondary outcomes include all-cause mortality at 14 and 90 days; treatment failure at

14 and 30 days; microbiological failure defined as positive blood cultures with index

pathogen at seven and 14 days; relapsed BSI at 30 and 90 days defined as recurrent

positive blood cultures with index pathogen after prior sterilization; metastatic

infections with index pathogen; secondary infections; *Clostridioides difficile*

Page 15

associated diarrhea; hospital re-admissions; development of resistance to study drugs

in clinical isolates; carriage of carbapenem-resistant Enterobacteriaceae

(carbapenemase-producing and non-producing); total in-hospital days; total antibiotic

days; liver function test abnormalities; allergic reactions; renal failure and other pre-

defined adverse events.

Subgroup analyses will be performed for the primary outcome of 30-day mortality by

infecting organism (*E. coli* vs. *Klebsiella spp.*); INCREMENT score (< 11 vs. ≥11)

[39]; bacteremia source (UTI vs. non-UTI); covering empirical therapy given in the

first 24 hours; patients not receiving the comparator drug empirically; and excluding

patients with an uncontrolled focus of infection.

**Table 4:** Outcomes

| Outcome | Definition |
|---|---|
| 30-day all-cause mortality (co-primary outcome) | |
| Treatment failure at day 7 (co-primary outcome) | Composite of the following by day 7:<br><br>• Death<br><br>• Fever above 38°C in the last 48 hours<br><br>• Symptoms attributed to the focus of infection still present<br><br>• SOFA score increasing<br><br>• Blood cultures positive with the index pathogen |

Page 16

| 14- and 90-day all-cause mortality | |
|---|---|
| Treatment failure at 14 & 30 days | As defined above |
| Microbiological failure at 7 & 14 days | Positive blood cultures with index pathogen at days 4-7 and 11-14 |
| Relapsed BSI at 30 & 90 days | Positive blood cultures with index pathogen following prior sterilization at days 30 and 90 |
| Metastatic focus of infection | Isolation of index pathogen from non-blood specimen related to metastatic spread of infection by day 90 |
| Superinfection | Development of either clinically or microbiologically documented infection within 90 days according to CDC surveillance definitions of health-care associated infections for bacterial infections |
| Resistant infection | Clinical isolates resistant to PTZ and meropenem and any carbapenem-resistant bacteria |
| Resistant colonization | Carriage of CPE and non-CPE CRE in-hospital till day 90, detected by weekly rectal surveillance of carriage while in-hospital |
| Re-admissions | Number of hospital re-admissions until day 90 |
| CDI | *Clostridioides difficile* associated diarrhea till 90 days |
| Adverse events | • Abnormal liver enzymes and bilirubin |

Page 17

| | <ul><li>Renal failure using the RIFLE [40] criteria by day 30 but we will not rely on urine output because it is not properly or accurately documented in many non-ICU inpatient units</li><li>Leukopenia, neutropenia, thrombocytopenia</li><li>Drug hypersensitivity</li><li>Diarrhea</li><li>Seizures</li></ul> |
|---|---|

SOFA, Sequential Organ Failure Assessment; BSI, bloodstream infection; CDC, Centers for Disease Control and Prevention; PTZ, piperacillin tazobactam; CPE, cabapenemase-producing Enterobacteriaceae; CRE, Carbapenem-resistant Enterobacteriaceae; CDI, *Clostridioides difficile* infection

**Microbiological methods**

All laboratories from centers participating in the study are ISO 9001 accredited laboratories. Following growth in blood culture, isolates will be identified using automated methods (Vitek 2, BD Phoenix, Vitek MS, MALDI biotyper). Antibiotic susceptibilities will be determined according to local practices, using either automated methods, disk diffusion, gradient diffusion, or a combination of these methods, and interpreted using either CLSI or EUCAST breakpoints as per local protocols. All isolates will be made available for future testing by a central laboratory where antibiotic susceptibility will be determined using BMD and interpreted according to EUCAST standards. Central laboratory personnel will be blinded to trial outcomes and to local antibiotic susceptibility test results. We will also determine and characterize the presence of ESBL and ampC genes using PCR.

Page 18

**Assessment and follow up**

Patients will be identified based on laboratory reports of Gram-negative bacteremia. All patients will be followed up till day 90 post randomization in-hospital and on re-admissions. During hospitalization patients will be visited by infectious diseases specialists as needed. Management decisions, such as diagnostic evaluation, other medical/surgical procedures and discharge from hospital will be left to the discretion of the treating physicians. Defined adverse events will be collected from the patients' charts and continuation of therapy will be similarly left of the discretion of treating physicians. We will not mandate diagnostic testing further than those defined for outcome collection and these will be done as clinically indicated. Patients will not be asked to return for study visits after discharge.

Data will be collected from the study visits, laboratory reports and the electronic health record. Following discharge, we will document re-admissions with outcome events during readmissions; and survival status through the national electronic patient files in Israel, through regional databases in Italy, and through local data and direct patient contact (text/email/phone/mail) in Canada. Anonymous data will be entered into a central case report form (CRF) designed in REDCap, a secure web application.

**Sample size**

For the mortality endpoint we calculated a sample size of 542 patients per arm assuming a 12.5% mortality rate in the control group with a 5% non-inferiority margin and a 1-sided hypothesis with 5% α-risk and 80% power [41]. The assumed mortality rate of 12.5% was based on rates reported in contemporary observational studies (17.3%) [7–9] and the MERINO RCT (7.9%) [11]. We do not assume loss to follow-up given complete 90 day follow-up in 719 patients with bloodstream infections in two previous RCTs performed by our group [42,43].

Page 19

The sample size calculation for the treatment failure outcome assumes a 25% failure rate at seven days in the control group. To test for non-inferiority of PTZ compared to meropenem with a 1-sided 5% α-risk, 80% power and a non-inferiority margin of 10% we will need 232 patients per study group.

**Monitoring and trial management**

The trial will be monitored centrally by the coordinating center at RHCC. Data entry will be monitored continuously on REDCap, checking for timely data entry, missing data or suspected faulty data. Inconsistencies and logical rules have been pre-defined to allow detection of such events. We will employ a risk-based strategy, with sparse on-site monitoring based on central inspection of the data. A steering committee has been nominated (the PIs and selected investigators representing all countries) and the trial will be followed by an independent safety monitoring board (two infectious diseases specialists and one pharmacologist, all expert in clinical trials and external to the study centers).

**Statistical analysis**

We plan an interim analysis after recruitment of 250, 500 and 750 patients. The trial will be stopped if an extreme difference between groups of $p<0.001$ will be observed for the primary outcome of 30-day mortality. The difference was chosen based on the MERINO trial stopping rule [11] and following the Haybittle-Peto rule [44,45] that preserves the overall type I error rate at 0.05. The sample size of the first interim analysis was selected based on the minimal sample size required to reach a difference with $p<0.001$ presuming that the maximal difference between groups that we will reach is the one observed in the MERINO trial [11].

Page 20

The primary analysis will include all randomized patients following local susceptibility testing. A secondary analysis will exclude patients in whom major errors in susceptibility compared to BMD will be detected. A per protocol analysis will include patients fulfilling inclusion based on central lab adjudication of susceptibilities, without exclusion criteria and receiving the allocated intervention for at least four calendar days.

Patients' baseline characteristics will be displayed descriptively. Outcome variables will be compared using the chi-square test, Student's t-test or the Mann–Whitney U test, as appropriate. Risk differences for dichotomous outcomes will be computed with 95% confidence intervals. Non-inferiority will be fulfilled if the upper value of the 1-sided 95% CI for the risk difference of meropenem compared to PTZ will be equal or lower to the defined non-inferiority margin.

**Ethics and dissemination**

The ethics of recruiting patients into this study, after the MERINO trial, are embedded in the considerations we previously raised. These concern the possibility that their chance finding will not be observed in a larger repetition trial and some improvement in the study design through obtaining a larger sample size and improving PTZ pharmacokinetics. With these considerations, the study was approved by the ethics committees of the above Israeli hospitals and is awaiting approval in other hospitals. In Canada, institutional ethics approval has been granted for the Province of Quebec and the study has received approval from Health Canada as required for studies involving off-label use of approved pharmaceuticals.

Results of the study, whether completed or not, will be analyzed and made available through publication. De-identified individual patient data collected during the trial

Page 21

will be made available for an unlimited time period following publication of trial

results. Data will be available for researchers who provide a methodologically sound

proposal and contingent on both the researchers' and our ethics committee approval

and the signing of a data sharing agreement.

**Patient and public involvement**

We have not involved patients or the public in the trial's design and planning. We plan

to conduct a survey for bacteremia survivors and the public on the acceptability the

consensus endpoints defined for BSIs [38].

**Funding**

We have not succeeded in obtaining funding for the study from the Israeli Ministry of

Health. The study received the support of ESCMID through the society's Research

Grant programme 2020 (30,000 Euro) which will be used for the ethics and insurance

requirements of the Italian sites and for onsite study visits. The Canadian sites are

seeking funding through the Canadian Institutes of Health Research. Should the study

receive substantial funding we plan to recruit additional hospitals who currently do

not have the infrastructure to recruit into clinical trials and to revise the protocol,

mainly with respect to the microbiological methods so as to enable local laboratories

to perform BMD and test for resistance genes in real time.

**Competing interests**

None declared.

**Authors' contributions**

RB, FK, CM, YG, MC, GR, LN, RBA, AT, MHS, MPC, TCL, LL, DY, MP

contributed to conception, design, trial management and planned data analysis.

RB, FK, DY and MP contributed to trial database and randomization site design.

RB and MP wrote the first draft of the manuscript.

Page 22

All authors revised the protocol critically for important intellectual content and

approved the final manuscript.

Page 23

**Table 1:** Inclusion and exclusion criteria

| Inclusion criteria | Exclusion criteria |
|---|---|
| <ul><li>Adults (age ≥ 18 years)</li><li>New onset BSI due to *E. coli* or *Klebsiella spp.* in one or more blood cultures associated with evidence of infection</li><li>The microorganism will have to be non-susceptible to third generation cephalosporins (ceftriaxone and/or ceftazidime) and susceptible to both PTZ and meropenem</li><li>We will permit the inclusion of bacteremias due to *E. coli* or *Klebsiella spp.* with concomitant growth in blood of skin commensals considered as contaminants.</li></ul> | <ul><li>More than 72 hours elapsed since initial blood culture taken, regardless of the time covering antibiotics were started</li><li>Polymicrobial bacteremia defined as either growth of two or more different species of microorganisms in the same blood culture, or growth of different species in two or more separate blood cultures within the same episode of infection</li><li>Patients with prior bacteremia or infection that have not completed antimicrobial therapy for the previous infectious episode.</li><li>Patients with septic shock at the time of enrollment and randomization, defined as at least 2 measurements of systolic blood pressure < 90 mmHg and/or use of vasopressors (dopamine>15μg/kg/min, adrenalin>0.1μg/kg/min,</li></ul> |

Page 24

| | noradrenalin>0.1μg/kg/min, vasopressin any dose) in the 12 hours prior to randomization. In the absence of the use of vasopressors, a systolic blood pressure <90 would need to represent a deviation for the patient's known normal blood pressure. <br><br> • BSI due to specific infections known at the time of randomization: endocarditis / endovascular infections, osteomyelitis (not resected), central nervous system infections <br><br> • Allergy to any of the study drugs confirmed by history taken by the investigator <br><br> • Previous enrollment in this trial <br><br> • Concurrent participation in another interventional clinical trial <br><br> • Imminent death (researcher's assessment of expected death within 48 hours of recruitment after discussion with treating team) |
|---|---|

Page 25

**Table 3**: Dose adjustment for study antibiotics

A. All sites

|  | Meropenem | Piperacillin tazobactam |
|---|---|---|
| CrCl>50ml/min* | 1g q8h | 4.5g q6h |
| CrCl 26-50ml/min* | 1g q12h | 3.375g q6h (only if CCT<40) |
| CrCl 10-25ml/min* | 0.5g q12h | 2.25g q6h |
| CrCl<10ml/min* | 0.5g q24h | 2.25g q6h |
| Hemodialysis | 0.5g q24h (+0.5g AD) | 2.25g q8h (+0.75g AD) |
| Peritoneal dialysis | 0.5g q24h | 2.25g q8h |
| Continuous renal replacement therapy | By flow rate based on recommendations in: https://doi.org/10.3389/fphar.2020.00786 | |

*CrCl should be expressed in mL/min/1.73m2, using the modification of diet in renal

disease (MDRD) formula, Cockroft and Gault equation or other means.

AD- after dialysis

B. In Canadian sites **

|  | Piperacillin-tazobactam |
|---|---|
| CCT>40ml/min | 4.5g QID |
| CCT 20-40ml/min | 4.5g TID |
| CCT 10-20ml/min | 2.25g QID |
| CCT<10ml/min | 2.25g QID |
| Hemodialysis | 2.25g TID (+0.75g AD) |
| Peritoneal dialysis | 2.25g TID |
| Continuous renal replacement therapy | As above, by flow rate |

Page 26

** In Canada, to conform with the existing product monograph and accounting for the

unavailability of the 3.375g dosage form in most hospitals the following piperacillin-

tazobactam dosing strategy will be used (as extended infusion of 3 hours).

Page 27

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SPIRIT ✓

STANDARD PROTOCOL ITEMS: RECOMMENDATIONS FOR INTERVENTIONAL TRIALS

SPIRIT 2013 Checklist: Recommended items to address in a clinical trial protocol and related documents*

| Section/item | Page | Description |
|---|---|---|
| **Administrative information** | | |
| Title | 1 | PipEracillin Tazobactam versus mERoPENem for treatment of bloodstream infections caused by third generation cephalosporin-resistant Enterobacteriaceae – a study protocol for an open-label non-inferiority randomized controlled trial (PeterPen) |
| Trial registration | 4 | NCT 03671967 |
| | | |
| Protocol version | | September 10 V1 |
| Funding | 18 | The study received the support of ESCMID through the society's Research Grant programme 2020 (30,000 Euro). |
| Roles and responsibilities | 17 | Rambam Health Care Campus (RHCC) is the sponsor of the study. The principal investigators are Bitterman Roni and Paul Mical both from RHCC. The trial is not funded. The sponsor takes responsibilities for coordinating the protocol writing, design of the trial's database, randomization scheme and data collection and randomization tools, recruiting investigators and introducing the protocol to new sites. The sponsor will perform the monitoring as specified in the protocol and will ensure all ethics regulations are followed in the trial sites. The sponsor is responsible for the trial database that will be shared with all investigators and will perform the primary analyses. All investigators will review the analysis and provide input. The roles and responsibilities of the data monitoring committee have been described separately. |
| | | |
| **Introduction** | | |
| Background and rationale | 5-9 | Optimal treatment for ESBL-producing Enterobacteriaceae bloodstream infections has yet to be defined. Retrospective studies have shown conflicting results, with the majority of data suggesting the non-inferiority of beta-lactam beta-lactamase inhibitor combinations compared to carbapenems. The recently published MERINO trial reported the contrary with an apparent survival advantage to meropenem over piperacillin-tazobactam. The potential implications of the MERINO trial are profound, as widespread adoption of carbapenem treatment will have detrimental effects on antimicrobial stewardship in areas endemic for ESBL and carbapenem-resistant bacteria.  Therefore, we believe that it is justified to re-examine the comparison in a second randomized controlled trial prior to changing clinical practice forever. |
| | | |

| Objectives | 9 | Piperacillin tazobactam is non-inferior to meropenem for treatment of third generation cephalosporin-resistant E. coli and Klebsiella BSI |
|---|---|---|
| Trial design | 9 | Open label non-inferiority randomized controlled trial |

| **Methods: Participants, interventions, and outcomes** | | |
|---|---|---|
| Study setting | 10 | Study is conducted at 6 hospitals in Israel, 1 hospital in Italy and 2 hospitals in Canada. More hospitals are joining. |
| Eligibility criteria | 10-11 | Inclusion<br>• Adults (age ≥ 18 years)<br>• New onset BSI due to *E. coli* or *Klebsiella spp*. in one or more blood cultures associated with evidence of infection<br>• The microorganism will have to be non-susceptible to third generation cephalosporins (ceftriaxone and/or ceftazidime) and susceptible to both PTZ and meropenem<br>Exclusion<br>• More than 72 hours elapsed since initial blood culture taken, regardless of the time covering antibiotics were started<br>• Polymicrobial bacteremia<br>• Patients with prior bacteremia or infection that have not completed antimicrobial therapy for the previous infectious episode.<br>• Patients with septic shock at the time of enrollment<br>• BSI due to specific infections known at the time of randomization: endocarditis / endovascular infections, osteomyelitis (not resected), central nervous system infections<br>• Allergy to any of the study drugs<br>• Previous enrollment in this trial<br>• Concurrent participation in another interventional clinical trial<br>• Imminent death |
| Interventions | 11 | The intervention group will receive PTZ 4.5 grams q6h and the control group will receive meropenem 1 gram q8h. Dose adjustments for patients with renal insufficiency are specified. The study drug will be administered for a minimum of four to five days to complete at least seven days of antibiotic treatment |
|  | 11 | Research coordinators and investigators will monitor adherence to study protocol |
|  | 11 | Concomitant antibiotics are prohibited during the first week of the trial, unless secondary infections arise post-randomization an mandate change. |
| Outcomes | 13-14 | We defined two co-primary endpoints, the first being all-cause mortality at day 30 from randomization and the second being treatment failure at day seven from randomization.<br>Secondary outcomes include all-cause mortality at 14 and 90 days; treatment failure at 14 and 30 days; microbiological failure defined as positive blood cultures with index pathogen at seven and 14 days; |

| | | relapsed BSI at 30 and 90 days defined as recurrent positive blood cultures with index pathogen after prior sterilization; metastatic infections with index pathogen; secondary infections; *Clostridioides difficile* associated diarrhea; hospital re-admissions; development of resistance to study drugs in clinical isolates; carriage of carbapenem-resistant Enterobacteriaceae (carbapenemase-producing and non-producing); total in-hospital days; total antibiotic days; liver function test abnormalities; allergic reactions; renal failure and other adverse events.<br>The mortality outcome was chosen as this is the most relevant patient-related outcome and since it is not subject to bias. Both primary outcomes were selected based on a consensus statement of endpoints for such a trial. |
|---|---|---|
| Participant timeline | | Randomization has to occur within 72 hours from the time index blood culture was taken. We minimized need for mandatory tests and have limited it to blood chemistry, CBC and blood gases obtained on randomization and select time points (only as long as patient is hospitalized). |
| Sample size | 15-16 | For the mortality endpoint we calculated a sample size of 542 patients per arm assuming a 12.5% mortality rate in the control group with a 5% non-inferiority margin and a 1-sided hypothesis with 5% α-risk and 80% power. The assumed mortality rate of 12.5% was based on rates reported in contemporary observational studies (17.3%) and the MERINO RCT (7.9%).<br>The sample size calculation for the treatment failure outcome assumes a 25% failure rate at seven days in the control group. To test for non-inferiority of PTZ compared to meropenem with a 1-sided 5% α-risk, 80% power and a non-inferiority margin of 10% we will need 232 patients per study group. |
| Recruitment | | Recruitment will be initiated by researchers and based on laboratory-derived reports. Each center has flexibility can modify according to the common practice in place. |
| **Methods: Assignment of interventions (for controlled trials)** | | |
| Allocation: | | |
|     Sequence generation | 11 | Computer-generated list of random numbers |
|     Allocation concealment mechanism | 11 | Allocated centrally through a web site |
|     Implementation | 11 | Done by researches in each center |
| Blinding (masking) | 14-15 | There will be no blinding (except for staff at central laboratory for post-hoc analysis) |
| | | |
| **Methods: Data collection, management, and analysis** | | |
| Data collection methods | 15 | Data will be collected from the study visits, laboratory reports and the electronic health record. Following discharge, we will document re-admission and survival status through the national electronic patient |

| | | files in Israel, through regional databases in Italy, and through local data and direct patient contact (text/email/phone/mail) in Canada. |
|---|---|---|
| | | All data will be collected for patients deviating from trial protocol |
| Data management | 15 | Anonymous data will be entered into a central case report form (CRF) designed in REDCap, a secure web application. |
| Statistical methods | 16-17 | The primary analysis will include all randomized patients following local susceptibility testing. A secondary analysis will exclude patients in whom major errors in susceptibility compared to BMD will be detected. A per protocol analysis will include patients fulfilling inclusion based on central lab adjudication of susceptibilities, without exclusion criteria and receiving the allocated intervention for at least four calendar days.<br>Patients' baseline characteristics will be displayed descriptively. Outcome variables will be compared using the chi-square test, Student's t-test or the Mann–Whitney U test, as appropriate. Risk differences for dichotomous outcomes will be computed with 95% confidence intervals. Non-inferiority will be fulfilled if the upper value of the 1-sided 95% CI for the risk difference of meropenem compared to PTZ will be equal or lower to the defined non-inferiority margin. |
| | 14 | Subgroup analyses will be performed for the primary outcome of 30-day mortality by infecting organism (*E. coli* vs. *Klebsiella spp.*); INCREMENT score (< 11 vs. ≥11); bacteremia source (UTI vs. non-UTI); covering empirical therapy given in the first 24 hours; patients not receiving the comparator drug empirically; and excluding patients with an uncontrolled focus of infection. |
| | | |
| **Methods: Monitoring** | | |
| Data monitoring | 16 | The trial will be monitored centrally by the coordinating center at RHCC. Data entry will be monitored continuously on RedCap, checking for timely data entry, missing data or suspected faulty data. Inconsistencies and logical rules have been pre-defined to allow detection of such events. We will employ a risk-based strategy, with sparse on-site monitoring based on central inspection of the data. |
| | 16 | We plan an interim analysis after recruitment of 250, 500 and 750 patients. The trial will be stopped if an extreme difference between groups of p<0.001 will be observed for the primary outcome of 30-day mortality. The difference was chosen based on the MERINO trial stopping rule and following the Haybittle-Peto rule that preserves the overall type I error rate at 0.05. The sample size of the first interim analysis was selected based on the minimal sample size required to reach a difference with p<0.001 presuming that the maximal difference between groups that we will reach is the one observed in the MERINO trial. |
| Harms | 16 | All adverse events data will be collected into RedCap. Serious adverse events will also be reported to local IRB. |
| Auditing | 16 | Monitoring will be mainly remote with minimal on-site monitoring |

| **Ethics and dissemination** | | |
|---|---|---|
| Research ethics approval | 17 | IRB approval has been obtained in the specified centers and other centers are still seeking. |
| Protocol amendments | | Protocol amendments will be distributed to participating sites by coordinating center. |
| Consent or assent | | Each center will obtain informed consent according to local regulations |
| | | |
| Confidentiality | 15 | All data will be shared anonymously |
| Declaration of interests | 18 | Principal investigators have no conflicts of interest |
| Access to data | | The coordinating center (RHCC) will have access to the final combined data set. |
| Ancillary and post-trial care | | Insurance for trial participants is obtained according to local regulations |
| Dissemination policy | 18 | De-identified individual patient data collected during the trial will be made available for an unlimited time period following publication of trial results. Data will be available for researchers who provide a methodologically sound proposal and contingent on both the researchers' and our ethics committee approval and the signing of a data sharing agreement |
| | | We do not intend to use professional writers. |
| | | |
| **Appendices** | | |
| Informed consent materials | 17 | Local consent forms have been generated by each participating center. |
| Biological specimens | 10 | Blood sample for drug monitoring will be collected and stored for future analysis. The index bacteria isolate will be stored by each center for further analysis. |