Statistical Analysis Plan

Study Title:	REducinG Antibiotics tReatment Duration for Ventilator-Associated Pneumonia (REGARD-VAP)
Brief Title:	Defining Antibiotic Treatment Duration for Ventilator - Associated Lung Infection (REGARD_VAP)
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Chief Investigator:	Ben Cooper, Professor Mahidol-Oxford Tropical Medicine Research Unit, Mahidol University and University of Oxford

REducin**G** **A**ntibiotics t**R**eatment **D**uration for **V**entilator-**A**ssociated **P**neumonia

(REGARD-VAP)

Statistical Analysis Plan

Version 1.0

23 February 2023

Based on REGARD-VAP trial protocol version 5.1

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1. INTRODUCTION

This document details the proposed presentation and analysis for the Reducing antibiotics treatment duration for ventilator-associated pneumonia trial (REGARD-VAP). Any primary reporting of the REGARD-VAP study should follow the strategy set out in this document. Subsequent analyses of a more exploratory nature will not be bound by this strategy, though they are expected to follow the broad principles laid down here. The principles are not intended to curtail exploratory analysis nor to prohibit accepted practices (for example, data transformation prior to analysis), but they are intended to establish the rules that will be followed, as closely as possible, when analysing and reporting the trial.

The analysis strategy will be available on request when the principal papers are submitted for publication in a journal. Suggestions for subsequent analyses by journal editors or referees will be considered carefully, and carried out as far as possible in line with the principles of this analysis strategy; if reported, the source of the suggestion will be acknowledged. Any deviations from the statistical analysis plan will be described and justified in the final report of the trial. The analysis should be carried out by an identified statistician, who should ensure the integrity of the data during their processing. Examples of such procedures include quality control and evaluation procedures.

1.1. Key personnel

- Trial statistician: Mo Yin, mdcmy@nus.edu.sg
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- Data safety and monitoring board:
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2. BACKGROUND INFORMATION

2.1. Objectives

Primary objective

To assess the effect of short (7 days or fewer depending on clinical response) versus long (8 days or more depending on physicians' preference) antibiotic treatment strategies on mortality and pneumonia recurrence within 60 days of enrolment in adults with ventilator-associated pneumonia (VAP) in Asia.

Secondary objectives

To compare the following endpoints according to treatment allocation within 60 days of enrolment:

a) Ventilator-associated events
b) Duration of mechanical ventilation
c) Duration of hospitalisation
d) Acquisition of multidrug resistant infection or colonisation during the hospitalisation
e) Number of days of exposure to antibiotics during hospitalisation
f) Number and types of extrapulmonary infections during hospitalisation (determined from cultures taken from sterile sites)
g) Adverse events, i.e. antibiotic related diarrhoea, allergic reactions, readmissions

2.2. Study Design

The study design is a randomised, partially double-blinded (blinding may be achieved up to 7 days) controlled trial to assess the efficacy of short versus longer antibiotics course in adults with VAP. The trial has a stepwise noninferiority–superiority hypothesis, i.e. if non-inferiority of short duration, compared with long duration, is shown, statistical tests for superiority will be performed using closed testing methods without requiring adjustment of the significance level for multiple comparisons.

Period of external pilot	19 February 2018 to 23 May 2018
Date of start of recruitment	25 May 2018
Date of end of recruitment	16 December 2022
Date of end of follow-up	14 February 2023
Date of expected analysis	14 March 2023 to 14 May 2023
target number of participants	460 (230 per arm)

Participating study sites include:

1) National University Hospital, Singapore
2) Tan Tock Seng Hospital, Singapore
3) Sunpasitthiprasong Hospital, Ubon Ratchathani, Thailand
4) Srinagarind Hospital, Khon Kaen, Thailand
5) Patan Academy of Health Science, Patan Hospital, Kathmandu, Nepal
6) Civil Hospital, Kathmandu, Nepal
7) Khon Kaen Hospital, Khon Kaen, Thailand
8) Cajuru University Hospital, Pontifical Catholic University of Paraná, Brazil

2.3. Eligibility

Adult patients (≥18 years old) in participating wards who have been on mechanical ventilation for more than 48 hours who satisfy the following criteria are eligible for the study.

Inclusion Criteria

a. Patients 18 years and older

b. Invasive mechanical ventilation ≥ 48 hours

c. Satisfy the US Centers for Disease Control and Prevention National Healthcare Safety Network VAP diagnostic criteria [1]

- At least one of the following:
  1. temperature > 38 °C
  2. white blood cell count ≥ 12,000 cells/mm^3 or ≤ 4,000 cells/mm^3
  3. altered mental status with no other causes in > 70 year-olds; AND
- Two or more chest imaging tests demonstrating at least one of the following:
  1. new and progressive OR progressive and persistent infiltrate
  2. new and persistent OR progressive and persistent consolidation
  3. new and persistent OR progressive and persistent cavitation, AND
- At least two of the following:
  1. new onset of purulent sputum, or change in character of sputum, or increased respiratory secretions, or increased in suctioning requirements
  2. new onset or worsening tachypnea or dyspnea
  3. rales or bronchial breath sounds
  4. worsening gas exchange defined by oxygen desaturations (e.g., PaO2/FiO2 <240), increased oxygen requirements or increased ventilation demand

Exclusion Criteria

a. Poor likelihood of survival as defined by a Sepsis-related Organ Failure Assessment score (SOFA score) of >11 points [2]

b. Immunocompromised patients (HIV with CD4 <200 cells/mm^3, corticosteroids> 0.5 mg/kg per day for > 30 days, received chemotherapy in the past 3 months, solid organ or hematopoietic cell transplant)

c. Patients receiving antibiotic therapy for any other defined extra-pulmonary infections that warrant a duration of antibiotics longer than 7 days or complications of pneumonia such as lung abscess or empyema that warrant a duration of antibiotics longer than 7 days (excluding anti-tuberculosis treatment, antifungal medications, antibiotics meant for chronic suppression of chronic infections or chronic obstructive lung disease)

d. Patients who have been treated for VAP for more than 7 days from screening

e. Vulnerable population including prisoners and refugees

2.4. Treatment interventions

Randomisation

Randomisation will be done via stratified block randomisation by the study sites to ensure participants with similar characteristics such as sex and age are distributed equally in the intervention and control groups. Randomisation will be done with a computer program with a seed to allow reproducibility. Randomisation will be done with a 1:1 ratio. To prevent predictability of the random sequence, generation of the randomisation sequence is performed by an independent statistician and details of the randomisation generation is unavailable to all investigators. Randomisation will be allocated using sequentially numbered opaque envelopes. Fitness criteria for randomisation must be met prior to randomisation.

Antibiotic choice

Antibiotic treatment for VAP will be tailored to the susceptibility of the pathogen(s) and in accordance to the 2016 IDSA/ATS VAP guideline[3]. Primary physicians are encouraged to convert initial empirical regimen to narrow- spectrum therapy based on culture results. In culture-negative cases, empirical antibiotic choice should be made depending on local

antibiogram. The study team will not intervene on the antibiotics choices as this is beyond the objectives of the study.

Antibiotic duration

Number of days of antibiotics is calculated from the first day of appropriate coverage according to the susceptibility of at least 1 of the pathogen(s) recovered from respiratory cultures taken within 48h of screening or VAP symptom onset.

For those participants randomised to the intervention (short duration) arm, site investigators should assess the patients daily. Antibiotics should be stopped at

a) day 3 to 7, if all respiratory cultures during the same episode of VAP are negative, given that the criteria below are satisfied,

b) day 5 to 7, if any of the respiratory cultures are positive and positivity is attributable to the current episode of VAP, given that the fitness criteria below are satisfied,

a. body temperature was ≤ 38.3°C (core body temperature measured orally or rectally) or 38.0°C (axillary) for 48 hours, and

b. lack of hemodynamic instability (systolic blood pressure ≥ 90 mm Hg without inotropic support or no requirement of inotropic support to maintain systolic blood pressure above 90 mm Hg).

In the short duration arm, all antibiotics should be withdrawn by day 7 (short duration) according to the randomisation assignment, except for those participants with treatment failure in the case of persistent VAP or a new-onset infection of a different source prior to the last day of assigned duration of antibiotics. Persistent VAP or treatment failure is defined as the lack of improvement of hemodynamic stability or ventilation requirements without an alternative cause other than the same episode of VAP. These patients will continue to be monitored for the above-described “fitness criteria” and antibiotics should be stopped when they eventually meet the criteria. Recurrent VAP is defined as an additional episode of VAP satisfying the CDC clinical and radiological criteria of VAP[1] following commencement of the primary VAP treatment within 60($\pm$5) days of enrolment. Opinion from 2 respiratory experts blinded to the randomisation will be sought to diagnose persistence and recurrences. Patients who are diagnosed with an alternative source of

infection or a complication from VAP such as lung abscess or empyema that warrant a duration of antibiotics longer than 7 days prior to the last day of assigned duration of antibiotics are not considered treatment failures. These patients will not meet fitness criteria for randomisation and be discontinued from the study and subsequent analysis. In cases where patients are randomised to short duration and satisfy the above-described "stop criteria" but did not have their antibiotics stopped by day 7, they are considered protocol deviations. They will be analysed in the intention to treat analysis according to arm they are randomised to.

Duration of antibiotics in the control group will be at least 8 days with the exact duration decided by the managing physicians.

Adverse events will be recorded throughout the study period, including *C. difficile* infection, acute kidney injury, hepatitis, drug allergies, haematological and other complications, readmissions.

Blinding

Patients will be blinded to the study arm they are randomised to, as they will not be informed of the treatment duration and are likely to be sedated and unaware of the treatment regimens. Investigators will be blinded during the assessment of the participants for clinical stability based on the above-described criteria to minimise observer bias. Once conditions for stopping antibiotics are satisfied, the investigator will be unblinded and contact the primary physicians to stop antibiotics. The physicians will remain blinded until they are informed that the participant is suitable to stop antibiotics. Independent assessors, who are assigned to determine pneumonia recurrences, will be blinded from the randomisation arms. This will be achieved by blinding these independent assessors to study details which could be used to determine the allocation arm, including antibiotic treatment, for participants with potential recurrences from.

2.5. Non-inferiority margin and sample size

The study is designed as a hierarchical non-inferiority /superiority trial where, if non-inferiority can be established, superiority will be assessed. The primary outcome is the composite endpoint of mortality and recurrence at 60(±5) days of the short duration versus

the long duration of antibiotic treatment for VAP. A meta-analysis showed that mortality attributable to VAP ranges from 13.6 to 42.8% in Southeast Asia[4]. Considering that our primary outcome is a composite binary outcome of mortality and recurrence of VAP, we estimate this to be 55%. We derived an absolute non-inferiority margin of 12% with the fixed-margin method, preserving at least 50% of the efficacy of standard treatment in VAP. Using a group sequential design adopting the boundaries proposed by Fleming- Harrington-O'Brien (R=0.8)[5], a maximum of 412 patients will be required to achieve a power of 80% to conclude non-inferiority between the two groups with an one-sided α risk of 5%. As we anticipate a loss to follow-up of up to 10%, we plan to enrol a maximum of 460 patients.

2.6. Hypotheses and Definition of Primary and Secondary Outcomes

Primary end point

The primary end point of the study is the difference in the proportion of participants with the composite endpoint of death and VAP recurrence rate within 60(±5) days of enrolment.

Death refers to all-cause mortality. VAP recurrences are episodes which satisfy the US CDC VAP criteria, and verified by two independent respiratory or infectious diseases physicians blinded to the randomisation arm.

The first statistical test for the primary end point will be for non-inferiority. Here, the null hypothesis (H_0): $\theta \geq \delta$ will be tested against the alternative (H_1): $\theta < \delta$, where θ is the treatment effect difference between the proportion of participants who have the primary outcome in the short and long treatment strategy groups, and δ is the fixed non-inferiority margin of 12%.

In the event that short is non-inferior to the long treatment strategy, standard two-sided superiority testing will be performed. That is, the null hypothesis (H_0): $\theta = 0$ will be tested against the alternative (H_1): $\theta > 0$, where θ is the treatment effect difference between the proportion of participants who have the primary outcome in the short and long treatment strategy groups.

Secondary end points

All statistical tests for the secondary endpoints are standard two-sided superiority tests.

a) Ventilator-associated events

Definition: Events which satisfy the US CDC VAP criteria [1] but were not verified by two independent respiratory or infectious diseases physicians.

H_0: There is no difference in the proportion of participants who have ventilator-associated events in the short and long treatment strategy groups, i.e. $\theta^{VAE} = 0$;

H_1: There is a difference in the proportion of participants who have ventilator-associated events in the short and long treatment strategy groups, i.e. $\theta^{VAE} \neq 0$;

where θ^{VAE} is the treatment effect difference between the proportion of participants in the short and long treatment strategy groups.

b) Duration of mechanical ventilation

Definition: Total duration of mechanical ventilation during the 60 days of follow-up after enrolment.

H_0: There is no difference in the total duration of mechanical ventilation in the short and long treatment strategy groups, i.e. $t^v_s = t^v_l$;

H_1: There is a difference in the total duration of mechanical ventilation in the short and long treatment strategy groups, i.e. $t^v_s \neq t^v_l$;

where t^v_s is the duration of mechanical ventilation in the short and t^v_l is the duration of mechanical ventilation in the long treatment strategy groups.

c) Duration of hospitalisation

Definition: Total duration of hospitalisation during the 60 days of follow-up after enrolment.

H_0: There is no difference in the total duration of hospitalisation in the short and long treatment strategy groups, i.e. $t^h_s = t^h_l$;

H_1: There is a difference in the total duration of hospitalisation in the short and long treatment strategy groups, i.e. $t^h_s \neq t^h_l$;

where t^h_s is the duration of mechanical ventilation in the short strategy group and t^h_l is the duration of mechanical ventilation in the long treatment strategy group.

d) Acquisition of multidrug resistant infection or colonisation during the hospitalisation

Definition: Growth of multidrug resistant bacteria[6] from clinically indicated or screening cultures from any source during 60 days of follow-up.

H_0: There is no difference in the proportion of participants who colonised with multidrug resistant bacteria in the short and long treatment strategy groups, i.e. $\theta^{MDR} = 0$;

H_1: There is a difference in the proportion of participants who have ventilator-associated events in the short and long treatment strategy groups, i.e. $\theta^{MDR} \neq 0$;

where θ^{MDR} is the treatment effect difference between the proportion of participants in the short and long treatment strategy groups.

e) Total number of days of exposure to antibiotics

Definition: Total duration of antibiotic use during the 60 days of follow-up after enrolment.

H_0: There is no difference in the total duration of antibiotic use in the short and long treatment strategy groups, i.e. $t^a_s = t^a_l$;

H_1: There is a difference in the total duration of antibiotic use in the short and long treatment strategy groups, i.e. $t^a_s \neq t^a_l$;

where t^a_s is the duration of antibiotic use in the short and t^a_l is the duration of mechanical ventilation in the long treatment strategy groups.

f) Number and types of extrapulmonary infections during hospitalisation (determined from cultures taken from sterile sites)

Definition: Number and types of bloodstream, bone and joint, intracranial, intraabdominal infections during 60 days of follow-up.

H_0: There is no difference in the number of sterile site infections in the short and long treatment strategy groups, i.e. $x_s = x_l$;

H_1: There is a difference in the number of sterile site infections in the short and long treatment strategy groups, i.e. $x_s \neq x_l$;

where x_s is the number of sterile site infections in the short and x_l is the number of sterile site infections in the long treatment strategy groups.

g) Adverse events, i.e. antibiotic related diarrhoea, allergic reactions

Definition: Number and types of adverse events during 60 days of follow-up, which include antibiotic related diarrhoea, allergic reactions, readmissions

H_0: There is no difference in the number of adverse events in the short and long treatment strategy groups, i.e. $x^{AE}_s = x^{AE}_l$;

H_1: There is a difference in the number of adverse events in the short and long treatment strategy groups, i.e. $x^{AE}_s \neq x^{AE}_l$;

where x^{AE}_s is the number of adverse events in the short and x^{AE}_l is the number of adverse events in the long treatment strategy groups.

2.7. Outcomes Assessment Schedule

Baseline assessments are performed prior to randomisation on day 0. Other important assessment time points are detailed below.

Time point (Day)	Activity
0	Enrolment, baseline assessments.
Antibiotic prescription	
0-7	Antibiotic choice(s) is / are checked against respiratory endotracheal tube cultures. If current antibiotic choice(s) is / are not appropriate, primary physicians are advised to switch antibiotic choice according

	to antibiotic susceptibility report (when there is positive growth) or local microbiology antibiogram (when there is no positive growth). Randomisation to short or long treatment strategy. If randomisation is to short arm, all antibiotics for the participant should be stopped on the day of randomisation (other than, when applicable, anti-tuberculosis antibiotics, prophylaxis e.g. peri or post operative or in immunocompromised hosts, gut prokinetic antibiotics)
0 - discharge	Weekly follow-up for new or changes in antibiotic prescription for all participants.
Outcome assessment	
0 - discharge	New or changes in antibiotic prescription or new respiratory culture reports will trigger screening for pneumonia recurrence. The US CDC VAP diagnostic criteria will be checked during the preceding week. When these diagnostic criteria are met, two independent respiratory or infectious diseases physicians who are blinded to the randomisation arm will be consulted to verify if the episode is a recurrent episode.
0 - 60	Mortality is assessed weekly. Secondary outcomes such as adverse events, discharge / readmissions, intubation / extubation, clinical cultures, biochemistry laboratory tests are recorded weekly.

2.8. Data Management Responsibility

An appointed study monitor from the MORU clinical trial support group and a project coordinator will regularly visit the study sites for quality control. All study sites will be assessed prior to initiation of the study for capacity to conduct the randomised controlled trial, during the study and upon completion to ensure data quality. After site initiation, monitoring visits will perform monitoring on informed consent forms, CRF for completeness and accuracy of data and sample storage. There will be a minimum of three monitoring

visits per study site: after 3-5 participants enrolled, after 50% of the target sample size enrolled for a particular site and upon study completion. Monitoring reports will be made available to the study sites and investigators after each visit.

3. QUALITY CONTROL AND DATA VALIDATION

Throughout the trial, data checks will be performed in conjunction with data collection and data entry. Prior to any analysis, the Trial Statistician will perform additional data checks and validations, investigating the data for outliers and inconsistent dates. All apparent outliers will be checked against paper records and either confirmed as valid observations or corrected.

For the final analysis, a manual 100% data entry check of the following variables will be performed by local research staff:

- Dates for enrolment, randomisation, antibiotic durations, discharge, readmissions, intubation and extubations
- Primary outcomes including mortality and pneumonia recurrences
- Secondary outcomes including microbiological cultures, antibiotic susceptibility reports, adverse events

Following this, at least two rounds of queries will be raised by the central data manager based at MORU using the MACRO software to identify possible inconsistencies and missing data.

4. DATA SAFETY MONITORING COMMITTEE AND INTERIM ANALYSES

The data safety and monitoring committee (DSMB) consists of members independent from the study investigator team. The DSMB will review the analysis plan, interim analyses results, and their approval will be required before protocol and analysis plan amendments. The DSMB will receive reports regarding severe adverse events on a monthly basis, and will review the final study report. The DSMB will be empowered to advise stopping or suspending the trial, independent of the study investigators.

DSMB meetings should be held with every interim analysis and extra meetings may be convened at the request of the investigators, sponsor, or DSMB members to discuss emerging data that is a cause for concern. A full interim analysis including all available data from all sites will be reviewed by the DSMB after approximately 25% (125) participants from sites have been recruited and completed their follow-up to review the safety and ethics of the REGARD-VAP trial. Hence there will be four interim analyses will be performed on the primary endpoint.

A trial steering committee will also be constituted and will decide on the continuation of the trial and will report to the central ethics committee. The trial will be terminated if superiority of either short or long treatment durations is shown. We will use the group sequential design adopting the boundaries proposed by Fleming- Harrington- O'Brien (R=0.8)[5] to terminate the trial prematurely once the Z value exceeds the defined boundaries for superiority.

5. DESCRIPTIVE ANALYSIS

5.1. Representativeness of Study Sample and Patient Throughput

A complete CONSORT flow diagram will be included in the trial report, clearly stating the number of patients screened, eligible, randomised and followed-up throughout the trial. Information on reasons for ineligibility will be given; information on randomisations and follow-up will be presented by treatment arm and detail how many participants received their allocated intervention.

5.2. Baseline Comparability of Randomised Groups

For all information collected at baseline, numbers (with percentages) for binary and categorical variables (including sex) and means (and 95% confidence intervals), or medians (with the interquartile range and range) for continuous variables (including baseline patient reported outcomes and age) will be presented overall and by treatment group. There will be no tests of statistical significance or confidence intervals for differences between

randomised groups on any baseline variable because, by definition of randomisation, these arise only due to chance.

5.3. Comparison of Losses to Follow-up

The numbers (with percentages) of losses to follow-up (defaulters and withdrawals) over the 60 days of the study follow-up will be reported and compared between the short and long groups with absolute differences (95% CI). Any deaths (and their causes) will be reported separately within the section on SAEs and complications.

5.4. Description of Available Data

The availability of data for baseline assessments as well as for primary and secondary endpoints will be described for all appropriate trial time points. Data items are defined as available if the case report form has been completed and if the information provided can be used in the analysis. Summaries will be provided overall and by trial arm, and the number of available data items will be presented together with the number of data item expected and a percentage indicating the rate of data compliance for each endpoint and time point (i.e. investigating what percentage of expected data is actually available).

5.5. Description of Adherence with Intervention

Adherence with randomised intervention will be carefully assessed since poor adherence may increase type 1 error.

Adherence in the short arm is defined as all antibiotics being stopped 7 days or fewer from the start date of appropriate antibiotics or VAP respiratory symptom onset date, whichever is later. For example, for a participant who started having respiratory symptoms on 1st March 2020, and who was prescribed an appropriate antibiotic (e.g. meropenem) on 2nd March 2020, day 0 for which antibiotic duration is based on is 2nd March 2020. This participant will be considered adherent if all antibiotics (other than, when applicable, anti-tuberculosis antibiotics, prophylaxis e.g. peri or post operative or in immunocompromised hosts, gut prokinetic antibiotics) are stopped by 9th March 2020. Antibiotics are allowed to

be restarted subsequently for clinical indications. The percentage of participants in the short arm for whom antibiotics are restarted will be reported in the final report.

Adherence in the long arm is defined as antibiotics being continued for more than 7 days from the start date of appropriate antibiotics or VAP respiratory symptom onset date, whichever is later. For example, the same participant who started having respiratory symptoms on 1st March 2020, and who was prescribed an appropriate antibiotic (e.g. meropenem) on 2nd March 2020, day 0 for which antibiotic duration is based on is 2nd March 2020. This participant will be considered adherent if any antibiotics (other than, when applicable, anti-tuberculosis antibiotics, prophylaxis e.g. peri or post operative or in immunocompromised hosts, gut prokinetic antibiotics) are continued on or after 10th March 2020.

Antibiotic durations for the index episode of VAP, calculated using the above descriptions, will be presented in a survival plot to show the distribution of the data. This demonstrates how successful the trial intervention is and no statistical analysis will be performed. Total antibiotic duration, classified by short and long arms, during 60-day follow-up will be analysed as a secondary outcome.

The proportion of participants who are non-adherent will be presented in the CONSORT diagram with a breakdown of the actual reasons.

5.6. Bacterial Pathogens and Antibiotics Used for Treatment

For each index episode of VAP for which the study participants were enrolled for, the associated bacterial pathogen and the initial antibiotic treatment prescribed which were considered appropriate will be presented. These antibiotic regimens will be presented according to bacterial pathogen types and phenotypic resistance patterns (frequency and percentages). Due to the large number of antibiotic treatment choices and pathogen combinations, there will be no subgroup analysis for these combinations.

5.7. Reliability of Outcome Assessment

As it is not possible to completely blind the treating physicians to the antibiotic treatment duration, we have therefore put in measures for an objective measure of study endpoints. Firstly, all participants are followed up on a weekly interval at the same frequency. Secondly, as the primary outcome measure, all-cause mortality is used. As for pneumonia recurrence, independent assessors are blinded to the randomisation arms by only being provided relevant medical notes which do not contain information on the allocated intervention. Cause of mortality will be determined by the local physicians directly.

6. PATIENT GROUPS FOR ANALYSIS

The following patient populations will be utilised in the analysis:

Intention-to-treat (ITT)

The intention-to-treat population includes all study participants who have been randomised during the conduct of the study

Per-protocol

The per-protocol population includes all study participants who fulfill eligibility criteria specified in the inclusion/exclusion criteria, fitness criteria for randomisation and who received 7 days or fewer of appropriate antibiotics in the short arm, and 8 days or more of appropriate antibiotics in the long arm (Section 5.5).

7. ANALYSES TO ADDRESS PRIMARY AIMS

All analyses will be formed with R[7]. All packages used in the analysis will be cited according with the corresponding version numbers. All analysis code will be published in an open access online repository with the final manuscript.

All primary and secondary outcomes of the study populations will be analysed using both unadjusted and adjusted methods in both the PP and ITT populations. The purpose of using both adjusted analyses on the intention-to-treat and per-protocol populations to determine non-inferiority is to minimise the inflation of type 1 error associated with non-adherence in non-inferiority trials[8].

Multivariable regressions will be performed with inverse probability weighting, using baseline patient characteristics (study site, age, sex, comorbidities, residence prior to admission, type of ICU admitted to, SOFA score, VAP infection with CRE, maximum heart rate and minimum mean arterial blood pressure on randomisation day, duration of intubation prior to developing VAP, reason for intubation, number of days from first respiratory symptom onset to first day of appropriate antibiotics) as independent variables.

7.1. Evaluation/Definition of Primary Outcome

The primary endpoint of the REGARD-VAP trial will be analysed as a binary outcome (i.e. not as a time to event outcome) because dates may reflect timing of observations rather than actual failure. In this analysis, any participant with incomplete follow-up and no event observed to date will be classed as not having experienced an endpoint.

7.2. Statistical Methods Used for Analysis of Primary Outcome

Based on both the ITT and PP population, the proportions of participants experiencing the primary endpoint will be tabulated by treatment group. If the absolute, upper two-sided 90% CIs around the absolute unadjusted difference is less than 12%, then the criteria of non-inferiority will be met.

A multivariable logistic regression model, will be applied on the ITT population to calculate the estimates of the treatment differences adjusted for baseline patient characteristics described above. Categories with low counts may be combined. For example, in the event of comorbidities with very low counts, these comorbidities may be combined to avoid difficulties with the maximum likelihood estimation of the logistic model. Where no

information has been entered on the comorbidities, the participants will be considered not to suffer from these comorbidities.

For the PP population, inverse probability weighting[9] will be applied to calculate the estimates of the treatment differences using the same baseline patient characteristics as above.

For these multivariate logistic regression models, residual and predicted values produced form the model will be examined to assess the assumptions of the model. Specifically, the assumption of linearity between the predicted log odds and the covariates is assessed by plotting lowess graphs. The independence of the error terms will be considered. Influential cases are investigated by plotting the standardised Pearson's residuals against the predicted probabilities and the leverage of the individual observations.

7.3. Adjustment of P values for Multiple Testing

There is no multiple testing as only a single primary outcome is considered. All additional analyses are undertaken with an intention to further inform the results from the primary analysis. Therefore significance levels used will be 0.05 and 95% confidence intervals will be reported.

7.4. Missing Data

Minimal amounts of missing data are expected in the REGARD-VAP study as most information could be retrieved from the medical notes and frequent follow-ups from the research teams. Loss to follow-up may arise primarily in cases where participants formally withdraw from all further follow-up or relocate or their medical records can no longer be accessed. Low percentages of missing data are not expected to have a significant impact on the trial results, and therefore no sensitivity analysis to examine the impact of missing data is planned if complete outcome data is available for more than 95% of participants (i.e. less than 5% of participants are lost or withdraw from follow-up before 60 days).

However, in the event that the extent of missing data will exceed this level, multiple imputation models will be used to adjust for missing data. Data will be imputed based on the covariates used in the multivariable models (allowing for a possible non-linear effect of age), the log(last follow-up time) and a treatment failure indicator which will be set to missing for those with incomplete follow-up due to loss / withdrawal. Twenty imputed datasets will be created, and results will be pooled using the standard methodology (Rubin's rule) and 'mice' package in R[10]. Visual checks to compare the distribution of observed and imputed values will be performed to ensure the imputation model includes non-linear effects of continuous variables where appropriate. Multiple imputation assumes that the missing data follows a missing at random mechanism, i.e. the probability of an observation being missing can be explained by the observed data. Under this assumption, an imputation model that includes the appropriate variables will produce unbiased results. A secondary sensitivity analysis to assess the potential impact of missing data will also include a data missing not at random scenario, assuming that participants with missing data have worse outcomes than those with complete follow-up data, following the approach by White *et al*[11].

7.5. Pre-specified Subgroup Analysis

All subgroup analyses will be based on the ITT population, and presented as forest plots.

Subgroup analysis will be performed using the primary outcome as the dependent variable amongst patients with VAP caused by Gram-negative non-fermenters and carbapenem-resistant bacilli. Gram-negative non-fermenting bacilli include *Pseudomonas* spp., *Acinetobacter* spp., and *Stenotrophomonas* spp.. Carbapenem-resistant bacilli are Gram-negative bacteria which are resistant to at least one agent from the carbapenem category of antibiotics. VAPs caused by Gram-negative non-fermenters have been previously shown to be associated with increased recurrence[12,13]. VAPs caused by carbapenem-resistant bacilli have no standardised treatment. This will be exploratory as the sample sizes for each of these groups will not reach adequate power.

Subgroup analysis will be performed with tests for interaction, where multivariable logistic regression models will be constructed with the occurrence of the primary endpoint as the outcome, and the randomised treatment as well as VAP caused by Gram-negative non-fermenters and carbapenem-resistant bacilli and the interaction between randomised treatment and pathogen type as explanatory variables. Odds ratio from the interaction tests will be presented with 95% CI and corresponding p-values.

For all pre-specified subgroup analyses, diagnostic checks will be performed as described in section 7.2.

7.6. Treatment by Centre Interaction

Consistency of potential effects will be assessed across all centres by informal examination of the within-centre effects. There will be limited capacity to investigate these formally due to small sample sizes. Treatment allocation by centre interaction will be explored and treatment effects will be presented as forest plots without the performance of statistical tests. This summary will only include centres where patients were randomised in both arms.

7.7. Sensitivity Analysis

No sensitivity analysis in addition to that discussed in the above sections is planned in the context of the primary analysis. The above-described analyses (including both the ITT and PP analysis, which are parts of the primary analysis described above) are sufficient to assess the robustness of the trial results.

8. ANALYSES TO ADDRESS SECONDARY AIMS

Secondary outcomes are defined in section 2.6.

8.1. Statistical Methods Used for Analysis of Secondary Outcomes

The primary analyses described in section 7.2 will be repeated for secondary outcomes, based on the ITT population. Similar analysis on the PP population will be included in the supplementary materials.

8.2. Ventilator-Associated Events

Episodes of ventilator-associated events will be summarised overall and by treatment arm (frequency and percentages). Participants will be categorised as either having or not having experienced ventilator-associated events. Using this as a binary outcome variable, the analysis described in section 7.2 will be repeated.

8.3. Adverse Events and Complications

Episodes of antibiotic-related diarrhoea, allergy reactions and other side effects, readmissions will be summarised overall and by treatment arm (frequency and percentages). Participants will be categorised as either having or not having experienced adverse events. Using this as a binary outcome variable, the analysis described in section 7.2 will be repeated.

A breakdown of serious adverse events (SAE) will also be presented in this section. SAEs in REGARD-VAP trial include deaths and pneumonia recurrences, which already contribute to the primary outcome. In a detailed breakdown of these SAEs, the number of participants with at least one recorded SAE, as well as the number of SAE reported per participant will be included. In addition, summaries will include the timing of the report from randomisation and whether complications were expected and / or thought to be related to the randomisation, and the outcome of any SAEs will be summarised.

8.4. Duration of Hospital Stay

Time from randomisation to discharge, and time from original admission to discharge, will be summarised overall and by treatment group using median (interquartile range) and compared using Wilcoxon rank sum test. Using this as a continuous outcome variable, the analysis described in section 7.2 will be repeated.

8.5. Duration of Mechanical Ventilation

Time from randomisation to extubation, and time from original intubation to extubation, will be summarised overall and by treatment group using median (IQR) and compared using Wilcoxon rank sum test. Using this as a continuous outcome variable, the analysis described in section 7.2 will be repeated.

8.6. Total Duration of Antibiotics

Total duration of antibiotics from randomisation to 60 days of follow-up will be summarised overall and by treatment group using median (interquartile range) and compared using Wilcoxon rank sum test. Using this as a continuous outcome variable, the analysis described in section 7.2 will be repeated.

8.7. Acquisition of Multidrug Resistant Infection or Colonisation

Acquisition of multidrug resistant infection or colonisation will be summarised overall and by treatment arm (frequency and percentages). Participants will be categorised as either having or not having acquired multidrug resistant infection or colonisation. Using this as a binary outcome variable, the analysis described in section 7.2 will be repeated.

8.8. Number and types of extrapulmonary infections during hospitalisation

Extrapulmonary infection events in sterile sites are most likely to occur in bloodstream infections. Unless infections from other sterile sites exceed >5 episodes in either arm, only bloodstream infections will be summarised overall and by treatment arm (frequency and percentages). Participants will be categorised as either having or not having experienced bloodstream infections post randomisation. Using this as a binary outcome variable, the analysis described in section 7.2 will be repeated.

8.9. Resource Use and Cost Data

A separate analysis plan for the health economics analysis will be written by the trial health economist. Resource use and cost data will only be assessed for the final analysis, but not for the interim analysis.

9. ADDITIONAL ANALYSES

9.1. Exploratory Analyses

No additional exploratory analysis is currently planned. If the trial team, in discussion with the DSMB or TSC intends to perform any additional analyses, the statistical analysis plan will be updated accordingly. Any exploratory analysis that has not been pre-specified will be clearly marked as such in the final statistical report.

9.2. Blinded Analysis

The trial statistician will not be blinded to treatment allocations while preparing and performing the statistical analysis for this trial.

9.3. Meta-Analyses

Results of this trial will be considered in a meta-analysis of antibiotic treatment duration for VAPs, specifically focusing on Gram-negative non-fermenting bacilli. A duration-response curve will be plotted using data from the meta-analysis, if the overall data allows. This meta-analysis will be published as a separate manuscript.

9.4. Safety Analysis

SAE and AE are collected as part of the primary and secondary endpoints and all relevant analysis is details in sections 7 and 8.

10. GLOSSARY OF ABBREVIATIONS

REGARD-VAP	Reducing antibiotics treatment duration for ventilator-associated pneumonia trial
MORU	Mahidol-Oxford Research Unit
PI	Principal investigator
DSMB	Data safety and monitoring board
TSC	Trial steering committee
CI	Confidence interval
ITT	Intention-to-treat
PP	Per-protocol
SAP	Study analysis plan
AE	Adverse event
SAE	Serious adverse event

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