NeoVT-AMR

Strategies to reduce vertical transmission of multi-drug resistant pathogens to neonates (NeoVT-AMR)

NeoVT-AMR Statistical Analysis Plan			
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Author	**Position**	**Signature**	**Date**
Michelle Clements	Trial statistician	DocuSigned by: Michelle Clements AD78954D2C79466...	2023-May-10
Approved by			
Sarah Walker	Co-investigator & statistical reviewer	DocuSigned by: Ann Sarah Walker A21A8226533E49A...	2023-May-10
James Carpenter	Independent reviewer	DocuSigned by: James Carpenter D39C318C5788411...	2023-May-10

Revision History

Version	Author	Date	Reason for Revision
Draft 0.1	MC	02/02/2022	First draft – copy of protocol version 3.0
Draft 0.2	MC	12/08/2022	First substantial draft
Draft 0.3	MC	12/09/2022	Updated after comments from SW
Draft 0.4	MC	21/09/2022	Updated with subgroups following discussion in TMG
Draft 0.5	MC	04/10/2022	Updated following comments from James Carpenter
Draft 0.6	MC	22/02/2023	Updated following comments from James Carpenter and Andrew Atkinson and following interim analyses of safety data (see interim analysis section below)
Draft 0.7	MC	13/04/2023	Updated with prior information for Bayesian analysis
Draft 0.8	MC	24/04/2023	Updated after comments by EB
V1.0	MC	10/05/2023	Updated after final comments by JC and upversioned

Contents

1. Trial design

1.1. Design & outline

NeoVT-AMR is a randomised controlled factorial trial with 3x2 + standard of care (SOC) design with two independent strata – labouring mothers and hospitalised neonates. Within each stratum, 147 individuals will be recruited – 21 per treatment combination or SOC. Additionally, 147 babies from mothers in the maternal stratum will also be swabbed at birth.

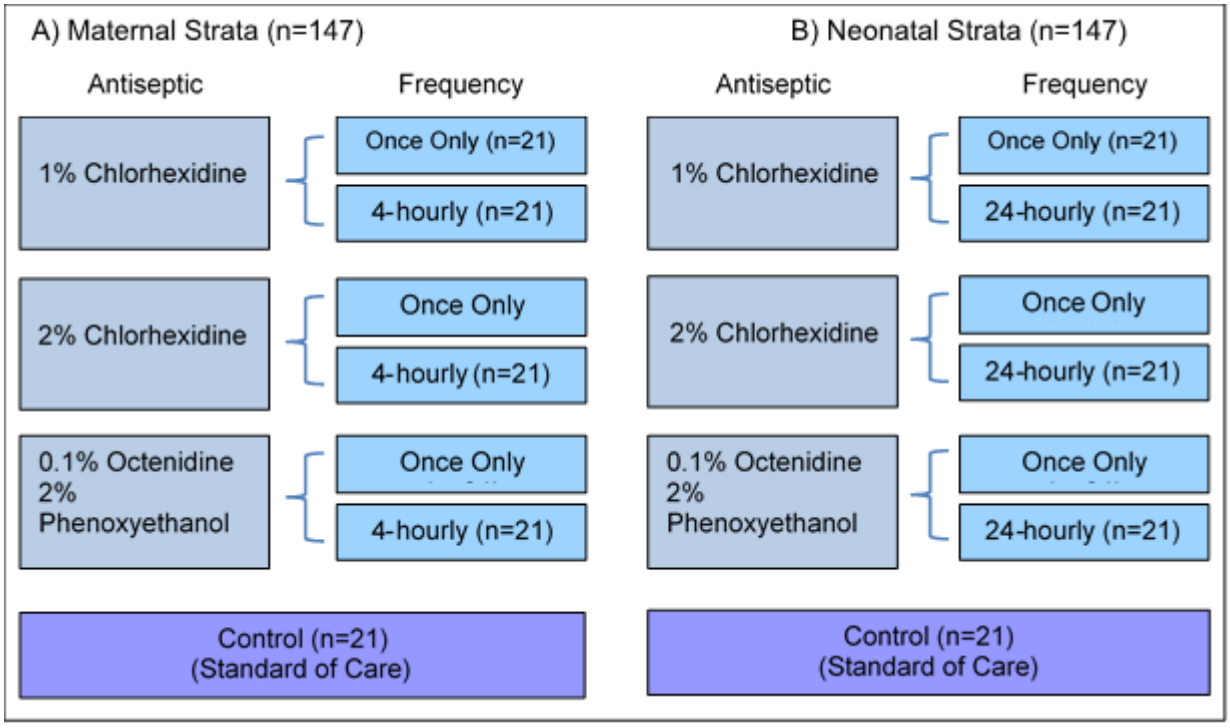
The two factors are:

- Antiseptic: 1% chlorhexidine (CHG), 2% CHG and octenisept (OCT)
- Frequency of application: once only and multiple applications

Multiple applications timings are 0/4/8/24/28/32h in maternal strata and 0/1/2/3d in neonatal strata.

The SOC group will receive standard of care. For analyses, the reference arm will be 1% CHG once only as there are more effective replicates of each of the factorial arms and therefore greater power for comparison than with SOC. 1% CHG once only was selected as it would be the most straightforward to implement and CHG is more widely available than OCT so evidence of superiority of OCT over CHG would have to be shown for implementation to be advised.

The trial design is summarised in the trial scheme below. Note that the 'once only' frequency boxes are (n=21) also.



1.2. Population

Eligibility to the trial is based on the participant meeting all of the inclusion criteria and none of the exclusion criteria.

Inclusion criteria

Maternal strata

1. Presenting in labour with or without rupture of membranes

Neonatal strata

1. Born in a healthcare facility
2. Postnatal age at randomisation <24 hours
3. Birth weight >1000g

Exclusion criteria

Maternal strata

1. Under the age of 18 (minor in Malawi)
2. Any contra-indication to digital vaginal examination
3. In active labour (regular contractions and/or cervical dilatation of more than 4cm)
4. Poor perineal and vaginal skin condition as judged by clinician
5. Planned elective caesarean-section delivery
6. Known or suspected allergy to chlorhexidine or octenidine or phenoxyethanol
7. Intrauterine death confirmed or expected before randomisation
8. Antiseptic application or enrolment in the trial determined inappropriate in the opinion of the enrolling clinician
9. Any recent or planned (within 4 hours) iodine application to the perineum or vagina
10. Unable to obtain consent

Neonatal strata

1. Born by planned elective caesarean section
2. Born to mothers recruited in the trial
3. Poor skin condition (skin score of 2 or more in any of three domains (see Appendix I)) at the time of enrolment
4. Known congenital or acquired skin disorder or defect at time of enrolment
5. Antiseptic application or enrolment in the trial determined inappropriate in the opinion of the enrolling clinician
6. Any recent or planned (within 4 hours) iodine application to body
7. Any planned or previous lumbar puncture
8. Unable to obtain parental or guardian consent

The primary analysis population is intention-to-treat, including all randomised babies, regardless of treatment received. This corresponds to estimating the impact of the effectiveness of the treatments. However, in secondary analyses we will also use inverse-probability weighting methods to adjust for deviation from randomised strategy if non-compliance rates are >15%, which is a more efficient approach than defining a per-protocol population (1).

Loss-to-follow rates are unknown but may be high as participants may give birth (in the maternal strata) or be discharged (in the neonatal strata) before all planned swabs are taken. Missing swabs

will be monitored and participants without baseline and two post-baseline swabs may be replaced to maintain power. However, all baseline and post-baseline results will be included in analyses.

2. Outcome Measures

2.1. Co-Primary Outcome Measures

Maternal strata

- Change in vaginal and perineal bacterial load (in colony forming units)

Neonatal strata

- Change in neonatal skin bacterial load (in colony forming units)

Bacterial load will be assessed from vaginal and perineal swabs taken separately in the maternal strata and from neck and perirectal swabs taken separately in the neonatal strata. The log CFU for each swab will be summed to get the total log CFU. Timings of the swabs are the same as the timings for applications in the multiple application arm. Swabs are taken before antiseptic is applied.

2.2. Secondary Outcome Measures

Maternal strata

- Maternal toxicity – score and grade (tolerability and safety)
- Bacterial load in neonates exposed to maternal antiseptic, compared to control
- Serious adverse events

Neonatal strata

- Adapted neonatal skin condition score (safety) (absolute score and grade)
- Temperature (change in absolute temperature and grade (hypothermia))
- Serious adverse events

2.3. Additional secondary efficacy outcomes not in protocol

Maternal strata

- Change in vaginal and perineal bacterial load (in log CFU) of specific species:
  - All gram positive
  - All gram negative
  - Yeast
  - All enterobactereales combined (E. coli, Enterobacter, klebsiella, Proteus-Providencia-Morganella, Serratia, other enterobactereales)
  - Klebsiella spp.
  - E. coli
  - Serratia spp.
  - Proteus-Providencia-Morganella
  - Enterobacter spp.
  - Acinetobacter spp.
  - Staphylococcus aureus
  - Group A streptococcus (GAS)
  - Group B streptococcus (GBS)
  - GAS and GBS combined
  - Enterococcus spp.
  - Candida spp.
  - Pseudomonas spp.
  - Salmonella spp.

  - Other Enterobacterales
- Acquisition and loss of specific species above
- Components of total CFU considered separately
- Components of skin scores considered separately

Neonatal strata

- Change in neonatal skin bacterial load (in log CFU) of specific species as per maternal strata
- Acquisition and loss of specific species as per maternal strata
- Components of total CFU considered separately
- Components of skin scores considered separately

2.4. Sample Size Calculation

There is very little data on bacterial load in neonates in LMICs. Additionally, bacterial loads are expected to differ across locations complicating interpretation of previous studies. For example, previous studies in neonates have found varying effects of CHG on log colony counts: a decrease from baseline to 24h of 0.2 SD with 1% CHG in Nepal (2) and 2SDs with 2% CHG in the USA (3). The primary endpoints of log colony forming units (CFUs) are widely accepted to be normally distributed. Sample size calculations used standard formula with Bonferroni adjustment for multiple testing within arms of frequency and antiseptic. Within each stratum, 147 individuals randomized to 7 treatment groups, 21 participants per group, provides 90% power to detect a difference of 0.82 standard deviations (SDs) between antiseptics (two-sided α=0.012) and 0.58 SDs between frequencies (α=0.05) (80% power for 0.72 and 0.50 SDs, respectively). This also provides 90% power to detect a difference of 1.01 SD between each antiseptic and SOC (α=0.012), and 0.90 SD between frequency and SOC (α=0.025) (0.88 and 0.79 SDs at 80% power). These expected effects were judged by clinical and statistical members of the team to be sufficient for selecting a treatment regimen for a future trial.

2.5. Method of Randomisation

Randomisation is be performed using a random number generated as implemented in the statistical computing environment R. Randomisation is by permuted blocks (random block sizes of 7 and 14 randomly selected) to guard against bias introduced over time, such as outbreaks of pathogenic bacteria in the hospital. Participants are randomised 1:1:1:1:1:1:1 to the 6 treatment and 1 SOC arms.

Randomisation is built into electronic data systems using algorithms developed by the data management team at the Malawi Liverpool Wellcome Trust (MLW). The participant's trial number, treatment allocation and the date of randomisation is entered into the trial register at the site.

2.6. Estimands

The intervention is the randomised antiseptic and frequency of application.

The patient population is labouring mothers and new-born babies, with birth weight over 1kg admitted to hospital, as defined by the inclusion and exclusion criteria.

The co-primary endpoints are change in skin bacterial colony load, taken from vaginal and perineal swabs in the maternal strata and neck and perirectal swabs in the neonatal strata.

The population-level summary which provides a basis for comparison between treatment conditions is the difference in mean log colony forming units adjusted for baseline value.

Anticipated intercurrent events and associated strategy are:

- Loss to follow-up: rates are unknown but may be high as not many women are anticipated to labour for 32h. The mixed model gives valid estimates assuming outcomes are missing at random.
- Deviation from randomised strategy (antiseptic application and frequency as per protocol) will be assessed using multiple sensitivity analyses:
    - sensitivity analysis will censor at deviation from randomised strategy. Estimated treatment effects will then correspond to the (hypothetical) setting of no deviation.
    - sensitivity analysis using (a) post-intercurrent event data where appropriate and (b) MI will be used to assess robustness of treatment effect inferences where rates of deviation from protocol are above 15%.
    - sensitivity analysis using Inverse-probability weighting methods to adjust for deviation from randomised strategy if deviation rates are above 15%.

2.7. COVID-19

Eligible patients will be mothers and neonates within the hospital so risk of COVID-19 will be low. NeoVT-AMR was designed before the COVID-19 pandemic and so there are no explicit statistical mitigation strategies. Rate of positive COVID tests in trial participants and any resulting changes to treatment will be monitored and addressed in the analysis if necessary.

2.8. Interim analyses

The DMC met around the time the trial started (without reviewing any data) to ensure they were familiar with its design and conduct. They formally reviewed data halfway through planned recruitment. They may call additional meetings at any time at their discretion. In addition, SARs are sent in real time during the trial to the DMC. The DMC can recommend premature closure or reporting of the trial, or that recruitment to any randomised group be discontinued or modified. Such recommendations would be made if, in the view of the DMC, there is an unacceptable rate of adverse reactions. There will be no early stopping for clinical efficacy because this pilot trial is powered only to detect differences in skin bacterial load, not clinical efficacy outcomes.

The review halfway thorough recruitment was held on 8th November 2022 and reviewed safety data but not efficacy, as per the protocol. The SAP was not signed off at this point as there were still some unresolved questions from the statistical and DMC reviewer around the analyses of the efficacy endpoints. It was agreed with the DMC that it was preferable to go ahead with an incomplete SAP than delay safety analyses. Note that there were no issues around analyses of the safety endpoints considered during the DMC. V0.5 was the version in use during the DMC review. A SOP deviation was completed for this.

There will be no adjustment to the significance level due to interim analysis.

2.9. Final analyses

Final analysis will be after database lock following last-patient last-visit.

2.10. Software

Primary and secondary outcome analyses will be done in Stata v17 or later. Data preparation and analyses of other data will be done in R v4.2.1 or later.

3. Derivation of data to be analysed

3.1. Definition of baseline

Baseline values for all measurements will be those recorded at screening either on the screening and enrolment form, or zero hour treatment and microbiology data from the treatment and microbiology forms.

3.2. Follow-up timings

Timings of swabs will be:

Maternal strata

- 0h (baseline)
- 4h
- 8h
- 24h
- 28h
- 32h

Neonatal strata

- 0 day (baseline)
- 1 day
- 2 day
- 3 day

3.3. Loss to follow up

Loss-to-follow up rates are unknown but may be high. Missing swabs will be monitored and participants may be replaced to maintain power. However, all baseline and post-baseline results will be included in analyses.

3.4. Free text

Free text fields in CRFs may be corrected for spelling and further categorised.

4. Statistical Analyses

Information will be presented in tables and may also be presented graphically to aid interpretation.

Recruitment data will be presented as per standard CONSORT diagrams (4). Baseline data tables will be presented overall. Unless stated, post-baseline data tables will be presented aggregated by antiseptic and by frequency (working days, alternate working days), with SOC presented separately. Variables will also be presented by factorial randomisation or arm if there is difference between randomised groups of $p<0.05$, used as a flagging device for imbalance and expected for 1 in 20 characteristics by chance, with p-values from t-tests of differences between means for numeric variables and chi-squared tests or Fisher's exact test if cell values are small for categorical variables.

Statistical tests will use 95% confidence intervals unless otherwise stated. Associated two-sided p-values will be produced but binary conclusions of significant/not significant will not be drawn. As this is a pilot trial there will be no adjustment for multiple testing, although interpretation of results will take this into consideration.

All analyses will be included in the interim and final reports unless stated. All analyses will be presented by strata separately and not combined.

IQR: interquartile range

4.1. Recruitment & Randomisation

The following metrics will be presented by strata:

- Total randomisation, n(%)
- Randomisation to each antiseptic concentration: n(%)
- Randomisation to each application frequency: n(%)
- Randomisation to each antiseptic/freq combination: n(%)
- Eligibility: number and reasons for any participants randomised in error and excluded or ineligible children included in the analysis

4.2. Baseline

Maternal

- Age at randomisation (years): median (IQR)
- Estimated gestational age at labour: median (IQR)
- Gravidity: median (IQR)
- Parity: median (IQR)
- Antenatal care during pregnancy: n(%)
- Tuberculosis: n(%)
- HIV: n(%) positive, negative, unknown
- Gestational diabetes: n(%)
- Antibiotics during pregnancy: n(%)
- Antibiotics since admission: n(%)
- Cervical dilation at enrolment: median (IQR)
- Rupture of membranes at enrolment: n(%) yes, no

- If ruptured, how ruptured: n(%) spontaneous, assisted
- Prolonged rupture of membranes (>18h): n(%) yes, no, unknown
- Offensive liquor: n(%) yes, no, unknown

Neonatal

- Prolonged rupture of membranes (>18h): n(%) yes, no, unknown
- Offensive liquor: n(%) yes, no, unknown
- Mother received antibiotics in labour: n(%) yes, no, unknown
- Mode of delivery: n(%) vaginal spontaneous, vaginal assisted, emergency caesarean section
- Maternal HIV status: n(%) positive, negative, unknown
- Age at enrolment (hours): median (IQR)
- Estimated gestational age at labour: median (IQR)
- Sex: n(%) male, female
- AGPAR Score at 1 minute: median (IQR)
- AGPAR Score at 5 minutes: median (IQR)
- AGPAR Score at 10 minutes: median (IQR)
- Where admitted: n(%) postnatal ward with mother, nursery HDU, nursery
- Reason for admission: n(%) routine for mum and baby, maternal complication – baby admitted with mum, birth asphyxia, other
- Any antibiotics prescribed before enrolment since birth: n(%)
- Chlorhexidine cord care given: n(%)

4.3. Non-Trial Treatment

Maternal and neonatal strata

- Antibiotics received post-baseline: n(%)
- Other conmeds received post-baseline: n(%)
- Length of hospital stay: median (IQR)

4.4. Trial Treatment

The following metrics will be presented by factor and strata. Note that compliance to antiseptic will not be assessed as patients have individual dose bottles for application.

Note that swab timings are 0/4/8/24/28/32h in maternal strata and 0/1/2/3d in neonatal strata.

- Total number of antiseptic applications: median (IQR) and 1, 2… n(%)
- Timing of last antiseptic application: n(%) 0/4/8/24/28/32h in maternal strata and 0/1/2/3d in neonatal strata
- Number of antiseptic applications before each swab: median (IQR)
- Temperature taken before antiseptic application: n(%) in neonatal strata only
- Skin score assessed before antiseptic application: n(%)
- Temperature taken after antiseptic application: n(%) in neonatal strata only
- Skin score assessed after antiseptic application: n(%)

4.5. Follow-up

The following metrics will be presented by strata:

- Baseline swab: n(%) yes
- Number of swabs: median (IQR)
- Time in hours from baseline when each swab taken: median (IQR)
- Number of skin score assessments: median (IQR)
- Last time point of skin score assessment: : n(%) 0/4/8/24/28/32h in maternal strata and 0/1/2/3d in neonatal strata
- Day 28 follow-up: n(%)

4.6. Primary outcome analyses

Note that no efficacy outcome data will be presented at the interim analysis. Analyses beyond the primary analysis may not be presented at final analysis, depending on results from primary analysis.

A summary table will be presented:

- Total log10 CFU at baseline: mean (SD) [N]
- Total log10 CFU at each assessment time point: mean (SD) [N]
- Change in total log10 CFU from baseline to each assessment time point: mean (SD) [N]

Primary analysis

- The primary analysis population is intention-to-treat, including all randomised participants, regardless of treatment received.
- The outcome variable will be change in Total log10 CFU from baseline to assessment time point.
- The primary analysis will include all randomised participants with baseline and at least one post-baseline measure.
- The dependent variable will be total log CFU.
- The model will be a mixed effects model
- Fixed effects in the model will be:
    - Antiseptic (1% CHG, 2% CHG, OCT)
    - Frequency of application (once, multiple)
    - Time point of assessment: hour of swab (continuous; mfp will be used to assess linearity)
    - Baseline total CFU (continuous; mfp will be used to assess linearity)
- Individual will be fitted as a random effect to account for repeated measures within individuals. Goodness of fit of an unstructured covariance matrix will be assessed and used if it is clearly a better fit to the data than random intercepts only.
- Goodness of fit of an interaction between baseline total CFU and timepoint of assessment will be assessed and included in the model if clearly a better fit.
- Normally distributed errors will be fitted.
- The intercept (reference treatment category) will be once only 1% CHG as this is the lowest concentration of CHG, which is believed to be more readily available that OCT, and so would be the most straightforward to implement. The standard of care control will not be used as the reference as the sample size for this arm is lower than for the factorial arms and so power for comparisons is correspondingly lower.
- Comparison of effects between factorial arms will be assessed by testing whether the null hypothesis of no treatment effect can be rejected (equivalent to using the estimate and 95% confidence interval of the estimate of the difference to the intercept).
- Comparisons between other arms and between arms and the SOC arm may also be performed. There will be no formal correction for multiple testing but interpretation will take multiple testing into consideration.
- Model fitting will use restricted maximum likelihood (REML) and p-values are derived using the Kenward-Roger approximation.

Interaction analysis

- Interactions between antiseptic and frequency, antiseptic and timepoint of swab, and frequency and timepoint of swabs will also be fitted in separate models, to assess evidence for the presence of interactions.
- Models and reporting of results will be as per the primary analysis.

Repeated samples efficacy sensitivity analysis

- Change from baseline to the:
    - first outcome measure after baseline
    - final outcome measure

  will also be modelled
- Model and reporting of results will be as the primary analysis except individual will not be fitted as a random effect (as there will be no repeated measures within individuals)

Time since antiseptic efficacy sensitivity analysis

- The primary analysis described above will be repeated with time since last antiseptic application fitted as a fixed effect (continuous; mfp will be used to assess linearity)

IV antibiotics efficacy sensitivity analysis

- The primary efficacy analysis described above will be repeated with received IV antibiotics in last 24h/not fitted as a fixed effect (factor)

Deviation from randomised strategy sensitivity censoring analysis

- The primary efficacy analysis described above will be repeated with individuals censored at the point of deviation from randomised strategy if deviation rates are above 15%.

Deviation from randomised strategy multiple imputation analysis

- The primary efficacy analysis described above will be repeated using multiple imputation on individuals censored at the point of deviation from randomised strategy if deviation rates are above 15%.

Deviation from randomised strategy multiple IPW analysis

- The primary efficacy analysis described above will be repeated using IPW analysis on individuals censored at the point of deviation from randomised strategy if deviation rates are above 15%.

Bayesian analysis

- Primary outcome main analysis will also be conducted in a Bayesian framework using ACCEPT analyses (5) reporting the probability that one treatment arm is more effective than another treatment arm based on a continuous range of efficacy thresholds.
- Models will be fitted as the primary analyses above.
- Sensitivity analysis to prior assumptions will be performed using non-informative, optimistic and sceptical priors (see appendix for details). The analysis will focus on non-informative

priors, with informative priors used for sensitivity analysis, unless there are model fitting issues with non-informative priors.

- Posterior probability curves will be created for each factor compared to SOC and between arms within a factor.
- The posterior probability of each arm truly being better that the comparator will be calculated for each factor arm compared to SOC and between arms within a factor.

Subgroup analyses

Subgroup analyses will be:

Maternal

- HIV
- Antibiotics since admission
- Rupture of membranes at enrolment
- Prolonged rupture of membranes (>18h)
- Offensive liquor

Neonatal

- Maternal HIV status
- Mother received antibiotics in labour
- Prolonged rupture of membranes (>18h)
- Offensive liquor
- Age at enrolment
- Estimated gestational age at labour
- Where admitted
- Any antibiotics prescribed before enrolment since birth
- Chlorhexidine cord care given

The main effect will be fitted as above for the primary analysis with additional main effect of subgroup. The interaction between subgroup and each factor (antiseptic and frequency) will also be fitted one at a time, in separate models. Interpretation of results will take multiple testing and statistical power into consideration.

4.7. Secondary efficacy outcome analyses

Note that no efficacy outcome data will be presented at the interim analysis.

Analysis of bacterial load in neonates exposed to maternal antiseptic will be as per primary outcome analyses described above.

4.8. Secondary safety outcome analyses: skin score

A summary table will be presented:

- Skin score at baseline: mean (SD) [N]
- Skin score at each assessment time point: mean (SD) [N]
- Change in total log10 CFU from baseline to each assessment time point: mean (SD) [N]

- Absolute skin score at baseline: n(%) 0, 1, 2…
- Absolute skin score at each assessment time point: n(%) 0, 1, 2…

Skin score analysis

- Analyses will be as per the primary outcome above, except baseline skin score will be used in place of baseline log CFU.
- Transformations may be performed if there is clear evidence of deviations from normally distributed errors in the safety outcome. If more than 70% of the safety endpoint are in one category, goodness of fit of alternative models (e.g. ordinal) will be considered

4.9. Secondary safety outcome analyses: temperature

Temperature analysis will be performed in the neonatal strata only.

A summary table will be presented for all babies randomised to treatment groups:

- Temperature at baseline: mean (SD) [N]
- Temperature pre-application at all time points: mean (SD) [N]
- Temperature at post-application at all time points: mean (SD) [N]

Temperature analysis

- The analysis will include all babies randomised to treatment with at least one antiseptic application. Babies in the SOC group will be excluded from the analysis.
- The outcome variable will be:
    - Change in temperature from pre to post application.
- Fixed effects in the model will be:
    - Antiseptic (1% CHG, 2% CHG, OCT)
    - Frequency of application (once, multiple)
    - Day of trial (continuous; mfp will be used to assess linearity)
    - Temperature pre application (continuous; mfp will be used to assess linearity)
- Individual will be fitted as a random effect to account for repeated measures within individuals.
- Normally distributed errors will be fitted.
- Comparison of main effects within arms will be assessed using the estimate and 95% confidence interval of the comparison of differences between factors and in comparison, to SOC.
- The reference group will be 1% CHG once only as it is the most straightforward and conservative to implement. The SOC will not be used as the reference as the sample size for this arm is lower than for the factorial arms.

Additional analyses, as per the primary outcomes, may also be performed.

4.10. Secondary safety outcome analyses: AEs and SAEs

SAEs and AEs will be presented overall and split by MedDRA System Order Class (SOC) and Preferred Term (PT). AEs and SAEs will be displayed as n(%)M where M is the number of events experienced for all children experiencing at least one event (M>n).

- SAEs: n(%)M
- Antiseptic related SAEs: n(%)M
- Grade 3 or 4 AEs: n(%)M
- Grade 3 or 4 skins scores (appendix I and II of protocol) : n(%)M
- Grade 3 or 4 hypothermia in neonates only (section 4.7.3 of protocol) : n(%)M

AE and SAE analysis

- Frequency of SAEs will be compared using exact logistic models.
- Time-to-event models will be used if AEs/SAEs occur in >10% of the trial population overall.

4.11. Additional secondary efficacy outcomes analysis

Log CFU of specific species

- Will be analysed as per the primary outcome.
- Analyses beyond the primary analysis will be performed if considered necessary.

Acquisition and loss of specific species

- Will be analysed as per the primary outcome except Binomial generalised linear mixed models will be fitted with logit link will be used.
- Analyses beyond the primary analysis will be performed if considered necessary.
- Margins will be taken for presentation of results.

Components of total CFU considered separately

- Will be analysed as per the primary outcome.
- Analyses beyond the primary analysis will be performed if considered necessary.

Components of skin scores considered separately

- Will be analysed as per total skin score above.
- Analyses beyond the primary analysis will be performed if considered necessary.

5. References

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6. Appendix: Bayesian priors

6.1. Non-informative priors

Non informative priors were selected to capture a wide range of possible values. The same priors will be used for the maternal and the neonatal strata.

- Intercept mean =4, sd = 50
- Difference mean =0, sd = 20

6.2. Informative priors (optimistic and sceptical)

Informative priors were determined based on NeoCHG data (unpublished) and expert clinician opinion (Nicholas Feasey, David Lissauer, Emily Beales, Louise Hill, Neal Russell) during an online meeting on 21/03/2023. There is a lack of robust data to support the use of strong priors, especially in the maternal strata.

Intercept specification

The intercept (Control) was selected as data from NeoCHG could be used. The maternal strata will use data from d8 in NeoCHG, reflecting bacterial load with time to accumulate, whereas the neonatal strata will use data from baseline reflecting the recent birth of the neonates. Using mean values from the study and approximations of sd with a multiplier on the sd to capture extra uncertainty, the intercepts will be specified as following a normal distribution with parameters:

- Log CFU in maternal strata: mean 8.4; standard deviation 2.6; multiplier 3
- Log CFU in neonatal strata: mean 4.5; standard deviation 3; multiplier 2

A higher multiplier was chosen for the maternal strata as there is greater uncertainty in the expected values. Priors may be reformulated to a reference of 1%CHG/once before analysis to match reference specification in focal analysis.

Difference specification

Clinicians specified the mean difference that optimistic and sceptical persons knowledgeable on the topic might assume, using a reference of no antiseptic application. The approach taken was that both optimistic and sceptical persons were assumed to have some belief that the opposing view could be correct, given the assumption of equipoise for the trial to be able to go ahead. The mean values selected were:

Maternal strata

Parameter	Optimistic mean	Sceptical mean
0.5% CHG	Lower by 0.5	No effect
2% CHG	Lower by 1	No effect
OCT	Lower by 1	No effect
Single application	Lower by 0.5	No effect

Multiple application	Lower by 1	No effect

Neonatal strata

Parameter	Optimistic mean	Sceptical mean
0.5% CHG	Lower by 0.5	No effect
2% CHG	Lower by 1	No effect
Emollient	Lower by 1	No effect
Alternate day application	Lower by 0.5	No effect
Control	Lower by 1	No effect

Larger differences were selected than those found in NeoCHG due to the differing site conditions in NeoVT-AMR.

Differences were specified to follow a Normal distribution with mean as above and a certain percentage of the distribution more extreme than the opposing value (sceptical for optimistic priors and optimistic for sceptical priors) reflecting probability of alternate scenario being correct. This equates to a Normal distribution with:

- Mean = optimistic or sceptical mean
- sd = abs(sceptical mean – optimistic mean)/critical_value

Where the critical value is from the t-distribution with infinite degrees of freedom and probability equal to the alternate scenario being correct.

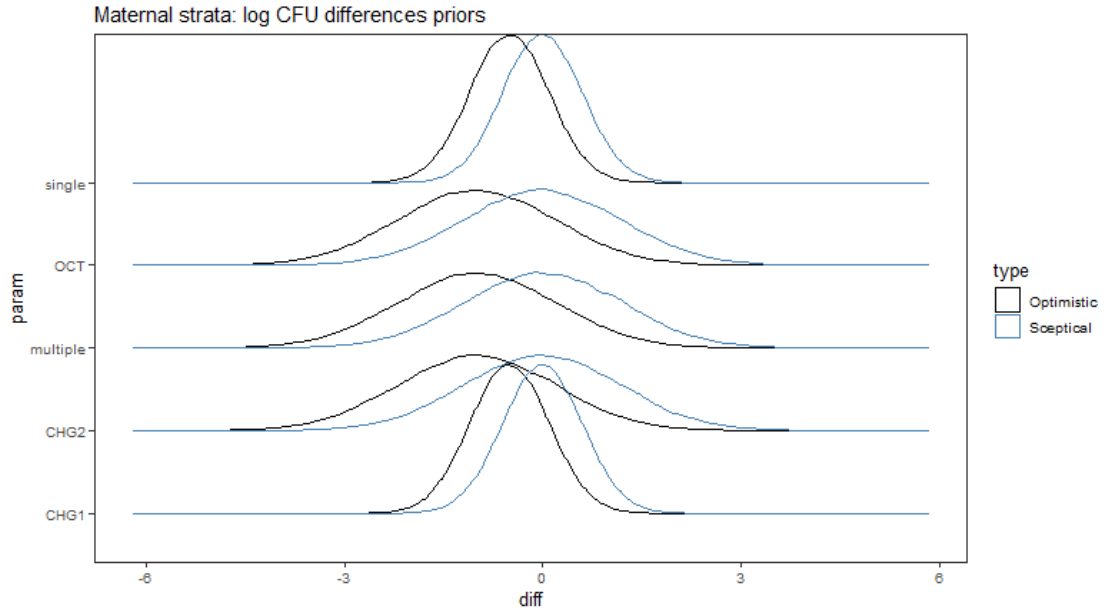
The probability of the alternate scenario being correct was

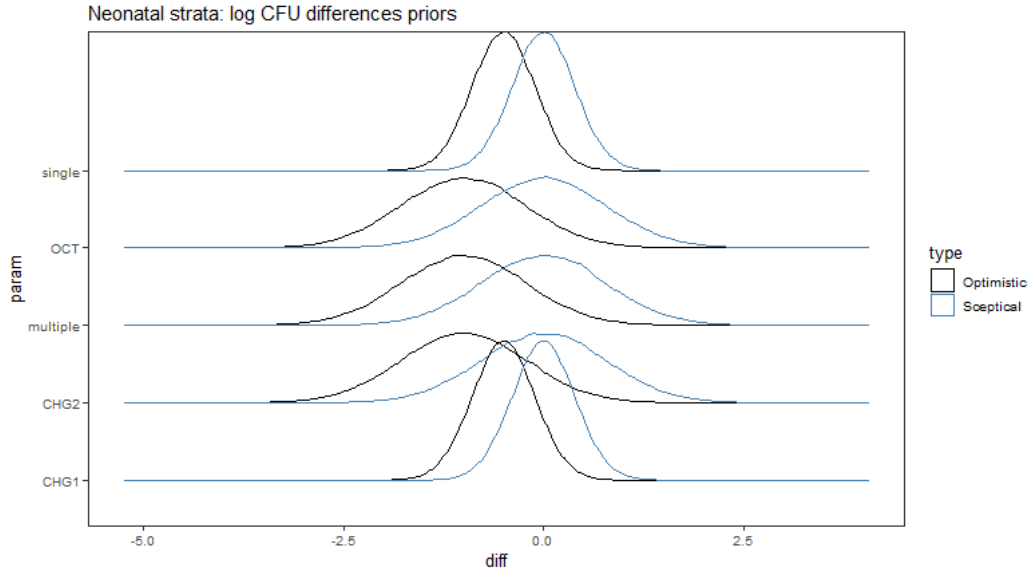
- Maternal strata: 0.2
- Neonatal strata: 0.1

A larger probability was chosen for the maternal strata to reflect the greater uncertainty in the this strata.

Plots of the differences are:

Maternal strata





Note the difference scales in each graph.