Study Protocol

Full title	A non-interventional, open observational non-inferiority study in two cluster-assigned cohorts of children aged 14 months into the safety of NeisVac-C® vaccines manufactured at two different production sites and given simultaneously with measles-mumps-rubella vaccine, assessed by web-based intensive monitoring
Short title	Web-based intensive safety monitoring of NeisVac-C® vaccination in toddlers
Study name	Peuterprik
Version	Version 1.2, 15 January 2014
EU PASS register	Not registered
Active substances	ATC code for study product NeisVac-C® J07AH07, for concomitant product M-M-R-VaxPro® J07BD52
Product reference	Netherlands Marketing Authorisation Number of NeisVac-C®: RVG 26343
Procedure number	UK/H/0435/001
Marketing authorisation holders	NeisVac-C®: Baxter Ltd M-M-R-VaxPro®: Sanofi Pasteur MSD
Joint PASS	No
Research question and objectives	To compare the safety of the NeisVac-C® bulk material produced at Baxter in Orth/Donau (new) (group A) with the material produced in Beltsville (old) (group B) in toddlers aged 14 months, administered simultaneously with MMR vaccine.
Country of study	The Netherlands
Protocol authors	Linda Härmark PhD, Hans Rümke MD PhD, Agnes Kant MPH PhD
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PROTOCOL SIGNATURE PAGE

Protocol details	
Study title:	A non-interventional, open observational non-inferiority study in two cluster-assigned cohorts of children aged 14 months into the safety of NeisVac-C® vaccines manufactured at two different production sites and given simultaneously with measles-mumps-rubella vaccine, assessed by web-based intensive monitoring
Study name:	Peuterprik

Name and role / function	**Signature**	**Date**
Agnes Kant Sponsor Director of Lareb		
Linda Härmark Principal Investigator Lareb		
Hans Rümke Coordinating Investigator Lareb		

PROTOCOL SYNOPSIS

STUDY TITLE	A non-interventional, open observational non-inferiority study in two cluster- assigned cohorts of children aged 14 months into the safety of NeisVac-C® vaccines manufactured at two different production sites and given simultaneously with measles-mumps-rubella vaccine, assessed by web-based intensive monitoring
STUDY NAME	Peuterprik
STUDY PRODUCT(S)	NeisVac-C®, Meningococcal Group C Polysaccharide Conjugate Vaccine Adsorbed, 'old' and 'new'
STUDY PHASE	Post marketing surveillance
STUDY OBJECTIVES Primary Secondary	To compare the safety of the NeisVac-C® bulk material produced at Baxter in Orth/Donau (new) (group A) with the material produced in Beltsville (old) (group B) in toddlers aged 14 months, administered simultaneously with MMR vaccine. The primary endpoint is the proportions of subjects with fever reactions within 4 days after administration. The secondary endpoints are proportions of subjects with - solicited other systemic and local reactions within 4 days after administration - non-solicited other systemic and local reactions within 4 days after administration - non-solicited other systemic and local reactions from 5 to 28 days after administration
STUDY DESIGN	non-interventional, open observational non-inferiority study with two cluster- assigned cohorts
SUBJECTS	At least 2430 healthy toddlers aged 13-18 months old, eligible to receive MenC and MMR vaccinations according to the NIP
INCLUSION CRITERIA	- age 13-18 months old (boundaries included) - eligible to receive MenC and MMR vaccinations according to the NIP
EXCLUSION CRITERIA	- children receiving either MenC or MMR vaccine and not both at same time - children with parents that are not able or willing to understand or fill the questionnaires
STUDY PARAMETERS	Primary: reactogenicity defined as % of children with rectal temperature 38,0 Celsius and above during the first 4 days after vaccination Secondary: other solicited and unsolicited complaints and symptoms at injection site as well as systemic reactogenicity
STUDY GROUPS	Two study groups of similar size will be formed according to the MenC vaccination that has been given at the baby clinics, i.e. NeisVac-C® from either Orth/Donau (group A) of Beltsville (group B).
STUDY PRODUCT REGIME	Children receive one injection of either MenC vaccine while MMR vaccine is given at a different injection site simultaneously
STUDY PERIOD	one year
STUDY DESCRIPTION	non-interventional, open observational non-inferiority study with two cluster- assigned cohorts
STATISTICAL ANALYSIS	The primary endpoint of the study, fever cases observed within 4 days after vaccination will be analyzed using logistic regression with vaccination groups ("old" / "new" product) and potential confounders as explanatory factors, applying a log link in order to obtain relative risk estimates at the end. Relative risk and its 95% CI of occurrence of fever cases with the "new" and "old" NeisVac-C® product will be calculated from the regression model assessing a potential increase of fever reactions with the "new" product. If the upper limit of the 95% CI is below 1.5 then the "new" product is considered to be non-inferior to the old one as far as fever reaction is concerned. The secondary endpoints will be analyzed similarly and descriptive without the non-inferiority considerations.
SPONSOR	Lareb

ABBREVIATIONS

Term	Explanation
AE	Adverse events
CI	Confidence Interval
CRF	Case Report Form
DVP	Dienst Vaccinvoorziening en Preventieprogramma's (till 2013 termed Regionale Coördinatie Programmas RCP), i.e. regional vaccine distribution and registry (part of RIVM)
ENCEPP	European Network of Centres for Pharmacoepidemiology and Pharmacovigilance
GCP	Good Clinical Practice
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
ITT	Intention to treat
MenC	NeisVac-C® vaccine to prevent infections by meningococci group C
MHRA	Medicines and Healthcare products Regulatory Agency
MMR	M-M-R-VaxPro® vaccine to prevent measles-mumps-rubella
NIP	Netherlands/National Immunisation Programme
PP	Per protocol
RIVM	Rijksinstituut voor Volksgezondheid en Milieu (National Institute of Public Health and Environment)
RMP	Risk Management Plan
RMS	Reference Member State
RVP	Rijksvaccinatieprogramma (National Immunisation Programme)
SAE	Serious adverse events
s.d.	Standard deviation
SmPC	Summary of Product Characteristics
SSAR	Suspected Serious Adverse Reaction
SUSAR	Suspected Unexpected Serious Adverse Reaction

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CHAPTER 1. INTRODUCTION

1.1 Rationale for the study

NeisVac-C® is a meningococcal group C polysaccharide conjugate vaccine (tetanus toxoid protein conjugate). The vaccine stimulates the immune system to produce antibodies against group C meningococcal bacteria, thereby preventing the development of meningitis/meningococcal disease. It is indicated for active immunisation of children aged 2 months and older, adolescents and adults, for the prevention of invasive disease caused by *Neisseria meningitidis* serogroup C.

NeisVac-C® is a product of Baxter Ltd. Due to a transfer of the manufacturing site of the drug substance from Baxter's facility at Beltsville, Maryland, USA to Baxter's facility at Orth/Donau, Austria, Baxter has applied for a Type II variation application (UK/H/0435/001/II/047/G) to allow release of Orth/Donau batches based on comparability to clinically qualified Beltsville batches to the United Kingdom's Medicines and Healthcare Products Regulatory Agency (MHRA) as the authority of the Reference Member State (RMS). Baxter was also requested by the MHRA to submit a Risk Management Plan (RMP) in which it should be considered how the safety and efficacy of the vaccine manufactured at the new Orth/Donau site would be actively monitored in comparison with the Beltsville vaccine. In particular it was asked to consider the extent to which there would be co-marketing of the vaccines and to specifically address batch traceability, the limitations of passive surveillance to evaluate relative reactogenicity and to formulate proposals to either overcome this or to conduct active surveillance to demonstrate comparable reactogenicity.

In The Netherlands Immunisation Programme NeisVac-C® is given to children of an age of about 14 months simultaneously with MMR vaccine. MMR is a live, attenuated virus vaccine, with a latency window of about 5 days for side effects that are associated with the replication of vaccine viruses. Any febrile reaction occurring during the first 4 days after administration of both vaccines, is more likely to be associated with NeisVac-C® than with MMR vaccine. Because of this co-administration, The Netherlands is a suitable country where reactogenicitiy of NeisVac-C® can be evaluated, compared to countries where NeisVac-C® is co-administered with non-live vaccines (as is the case with most other European countries).

We therefore will investigate systemic and local adverse events following NeisVac-C® and MMR administered simultaneously, with emphasis on the first 4 days after administration, and with subsequent follow-up during the remaining month.

CHAPTER 2. STUDY OBJECTIVES

2.1 Primary study objective

The primary objective of this study in children aged 14 months is:
- to compare the proportions of vaccinees with fever of ≥38ºC within 4 days after injections of the Baxter NeisVac-C® vaccine bulk material produced in Orth/Donau (new, group A) or NeisVac-C® vaccine of which the bulk material was produced in Beltsville (old, group B), and simultaneous MMR vaccine for both groups of NeisVac-C® recipients.

2.2 Secondary study objective(s)

Secondary objective of this study is to investigate
- the safety profile with other parameters (solicited and non-solicited) within 4 days after simultaneous injections of either MenC, and MMR vaccines.
- the safety profile of these vaccines with non-solicited parameters from 5 to 28 days after injection.

CHAPTER 3. STUDY DESIGN

3.1 Study design

This is a non-interventional, open observational non-inferiority study with two cluster- assigned cohorts .

3.2 Study description

Toddlers (14 months old) will receive at vaccination centers NeisVac-C® vaccination with either

- "old" lots produced in Beltsville (group B), or
- "new" lots from Orth/Donau (group A), simultaneously with MMR vaccine.

From 4 full days after the vaccines were administered, parents will receive web-based questionnaires with questions about any ADRs that occurred after vaccination.
The 1st questionnaire is sent at day 5 and covers days 1, 2, 3 and 4.
The 2nd questionnaire will be sent after two weeks and covers days 5 to 14.
The 3rd questionnaire will be sent after four weeks and covers days 15 to 28.

Day numbers are defined as:

- Day 1: day of vaccination, e.g. Monday
- Day 2: 1st day after vaccination, e.g. Tuesday
- Day 3: 2nd day after vaccination , e.g. Wednesday
- Day 4: 3rd day after vaccination, e.g. Thursday = last day of fever counting for primary endpoint.
- Day 5: 4th day after vaccination, e.g. Friday

3.3 Duration of the study and milestones

The study will continue for a whole year, to cover all seasons, and multiple vaccine batches as supplied by Baxter Ltd.
The duration of the study for each participant is approximately one month from vaccination.

Proposed timing of events (dependent on availability of study products and schedules of their delivery by RIVM). In case of vaccine batches to be studied run out of their period of approved use or pass their expiry date, the study may stop earlier.

year	month	activity / step
2014	APR	delivery of the appropriate batches in the study area
2014	APR	sending letters to parents of eligible children, from start weekly letters
2014	MAY	first inclusions
2014	JUN	first fully completed questionnaires
2015	APR	last letters sent
2015	MAY	last inclusions
2015	JUN	last completed questionnaires
2015	SEP	final draft study report
2015	OCT	study report

CHAPTER 4. SUBJECTS

4.1 Study population
Participants will be recruited from a regionally defined population of healthy children who are eligible to receive regular vaccinations with MenC and MMR vaccines according to the Netherlands Immunisation Programme.

In this study at least 2430 fully evaluable toddlers, aged 14 months, equally distributed between the 'old' and the 'new' MenC vaccine who are vaccinated with simultaneous MMR vaccine according to the Dutch Immunization Programme in clinics are required for Per Protocol analysis.

4.2 Inclusion criteria
- children 14 months old (age range 13-18 months; boundaries included) who receive the NeisVac-C® and MMR vaccination in one of the participating clinics according to the NIP

4.3 Exclusion criteria
- children receiving either MenC or MMR vaccine and not both at the same time
- children that have received simultaneously any other vaccine than MenC and MMR
- children that are vaccinated at baby clinics that do not participate in the study
- children whose parents/caretakers register for the study later than day 4 after vaccination.
- children with parents that are not able or willing to understand/fill the questionnaires in Dutch.

4. 4 Subject recruitment
In the target region designated vaccination centers/baby clinics will be supplied with NeisVac-C® vaccines with either "old" lots produced in Beltsville or "new" lots from Orth/Donau.
Approximately one month before the scheduled vaccination (usually at age 14 months) the parents/caretakers of children in the target region, will receive a letter with information about the study and the possibility to already sign up for the study, both online and through a paper form.

At the baby clinic, following NeisVac-C® and MMR vaccination, parents will be asked to participate, and the information required for participation will be written down on the study flyer (specification of injection sites and batch number per vaccine). Based on information in the letter and subsequent flyer, parents may decide to participate and register at the study web-page. Parental consent is considered a valid equivalent of a signature if they voluntary ticked a box stating that they have understood all information and confirm their participation.

4.5 Instructions and technical assistance during the study
During the study, the parents may contact the Netherlands Pharmacovigilance Centre Lareb at any time through mail or telephone if they have any questions concerning the study, especially if technical assistance in filling the questionnaires is required.

4.6 Subject discontinuation during the study
The participant can withdraw from the study at any moment without stating a reason. If a questionnaire has not been completed after a reminder has been sent, the patient is considered lost to follow up and will not receive any further questionnaires. Discontinuation has no medical consequences for the participant.

CHAPTER 5. STUDY PARAMETERS

5.1 Primary study parameter
Proportions of children with fever (rectally measured body temperature of ≥38.0°C) within 4 days after vaccination with NeisVac-C® and MMR.

5.2 Secondary study parameter(s)
Proportions of children with solicited other systemic and local reactions within 4 days after vaccination with NeisVac-C® and MMR.

The occurrence of the following reactions will be asked actively:
Local (injection site) reactions at the NeisVac-C® and/or MMR injection site
- redness, warmth, pain, swelling, induration, blue, itch

General (systemic) reactions
- rash
- decreased appetite
- less sleeping
- listlessness
- sleepiness/somnolence
- vomiting
- diarrhea
- (febrile) seizures

In addition a free text field is available so that symptoms not listed above can be reported.

5.3 Other parameters
The following base line data will be asked, to demonstrate comparability of both study groups. The influence of these potentially confounding characteristics will be addressed in the analysis:
- gender
- weight
- length
- medical history
- house hold size
- daycare and kindergarten

CHAPTER 6. STUDY PROCEDURES AND ASSESSMENTS

6.1 Schedule per eligible participant

time	actor	activity
- 4 wks	RIVM/DVP	sending letter to parents in region with predefined baby clinics. The letter is with headings of Lareb, and sent by actor.
-4 wks to-1 day	parents	pre-registration at Lareb LIM site, with contact details and planned date of vaccination
Day 1	baby clinic	- give MenC and MMR vaccinations - remind parents to the study described in the letter that parents received earlier. - record vaccinations on the study letter from the parents, and recording as usual for adequate registration by RIVM/DVP.
Day 4	parents	latest possibility to register for study, confirmation of date of vaccination
Day 5	Lareb	send questionnaire 1, reminder at day 7
Day 14	Lareb	send questionnaire 2, reminder at day 16
Day 28	Lareb	send questionnaire 3, reminder at day 32
Day 36	Lareb	closure

6.2 Procedures

Vaccinations take place at the baby clinic / health center of the toddler. Prior to vaccination, the child health physician or nurse reviews the health status of the child to exclude any contra-indication for MenC and MMR vaccinations according to the guidelines of the National Immunisation Programme. These vaccinations are given to all eligible children, regardless of later (non)participation.
Health personnel at the baby clinic will record the vaccinations (name clinic, name and batch no of vaccine, date of vaccination) in the child health dossier, as well as on the vaccination cards, which are sent to the RIVM vaccination registry (DVP), as prescribed in the NIP.

After vaccination, parents are reminded to the letter that they received earlier, and that they still may sign-up for the study with the use of a reporting code. If they intend to participate, baby clinic staff will record the vaccines given at that day, specifically batch numbers, and injection sites. The letter also contains a diary table, to be used by the parents as an aide-memoire, to record any health event that is to be filled in the electronic questionnaires.

Parents of children who have registered to participate will receive a web-based questionnaire at day 5 after the vaccines were administered with questions about any ADRs that occurred within 4 days of vaccination. A second questionnaire will be sent after two weeks, and a third after 4 weeks.

6.3 Assessments / collection of data

6.3.1 Preregistration and registration

Upon receipt of the first letter parents may pre-register for the study by reporting their email address and scheduled vaccination date. At this date, Lareb will send them an email with a link enabling the parents to register for the study.

After vaccination, parents are given a flyer with information about the study, and how to sign-up for the study (web address and study specific access code).

The following data are requested.

- general data
    1. are MenC and MMR vaccines administered both? If only one, or more than 2, vaccines were given, the child cannot further participate.
    2. reporting code
    3. date of vaccination
    4. batch numbers of administered vaccines (to be recorded at baby clinic)
    5. injection sites of administered vaccines (arm/leg/left/right) (to be recorded at baby clinic)
    6. registration of email address. Per email address, only one child can participate. For more children in a household/family (twins, triplets,..) separate email addresses are required.
    7. accept Lareb privacy rules yes/no. Only proceed if ticked: yes.

- data of reporting parent and child
    1. confirm email address
    2. child: date of birth
    3. child: sex
    4. child: height at time of vaccination
    5. child: weight at time of vaccination
    6. baby clinic: place and street address (choose from list).

- base line data
    1. medical history, did your child have: … (tick if applicable)
        - infections,
        - coughing/dyspnea,
        - skin rash,
        - hypersensitivity or allergy,
        - vomiting and/or diarrhea,
        - other: please specify …,
        - none of the above.

- concomitant medication
    1. Does your child use any medication: if yes: specify the medicine, dose, start date and stop date if applicable

6.3.2 Questionnaire 1 (4 days after vaccination)
Start question: did your child experience an adverse reaction/event: Y/N.
If N: have other medicines been used: click to add
If Y: which adverse events have occurred

- injection site reaction, if yes
    - where did the reaction occur: one or both sites: specify arm/leg/left/right
    - further questions per injection site
        - tick symptoms: red, warm, pain, swelling, indurated patch, blue, itch, other: please specify
        - size: largest possible diameter in centimeters (rounded upwards)
        - onset time after injection
        - treated, if yes specify
        - recovery status, if recovered: when
        - size will be categorised by diameter <2 cm: mild, 2-5 cm: moderate, >5 cm: severe

- fever, if yes
    - what was temperature (max value, rectally measured): __,_ºC
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- skin rash
    - please describe …,
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- less appetite
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- sleeping less
    - please describe …,
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- sleeping more/somnolence
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- listlessness
    - onset time after injection
    - treated, if yes specify
    - recovery status, if recovered: when
    - impact as experienced by parents, graded as: not-mildly-moderately-very

- vomiting
  - onset time after injection
  - how often
  - treated, if yes specify
  - recovery status, if recovered: when
  - impact as experienced by parents, graded as: not-mildly-moderately-very

- diarrhea
  - onset time after injection
  - how often
  - treated, if yes specify
  - recovery status, if recovered: when
  - impact as experienced by parents, graded as: not-mildly-moderately-very

- (febrile) seizures
  - please describe: ……
  - what was temperature at the time of the seizure: ….
  - onset time after injection
  - treated, if yes specify
  - recovery status, if recovered: when
  - impact as experienced by parents, graded as: not-mildly-moderately-very

- other reactions (more than 1 additional unsolicited reaction can be added).
  - please describe: ……
  - onset time after injection
  - treated, if yes specify
  - recovery status, if recovered: when
  - impact as experienced by parents, graded as: not-mildly-moderately-very

For all events together it will be asked :
- was the child seen by a doctor for this complaint.
- did any of the reported events lead to one of the following:
  - No, if Yes, tick if applicable:
    - hospitalisation
    - immediately life threatening
    - death

Any other data to label the reported AE as serious according to CIOMS criteria will be obtained from free text entries. They will be individually assessed by the Lareb investigator, and further regarded as SAE if applicable.

Co-medication:
- did your child have changes in medication.
  Medication listed at Start are shown again and open for editing (start date, stop date, dose, add new)

Characteristics of the household
- how many children are living in your household (including the vaccinee):
  choose from list: 1, 2, 3, 4, 5, 6 or more.
- does your child go to guest parents, daycare centre, kindergarten

Any reported complaints that are not resolved will be open to edit in next questionnaires.
In addition, parents are asked to report new events if applicable. All new events are to be entered in a free text field, and will be manually coded.

6.3.3 Questionnaire 2 (14 days after vaccination)
- questions about any events that are open since questionnaire 1. Treatment, recovery and severity data: earlier entries can be changed.
- questions about any new unsolicited events that have started after completion of questionnaire 1, including any occurrence of fever.
- did your child have changes in medication

6.3.4 Questionnaire 3 (28 days after vaccination)
- questions about any events that are open since questionnaires 1 or 2
- questions about any new unsolicited events that have started after completion of questionnaire 2, including any occurrence of fever.
- did your child have changes in medication
- is your child healthy at present
- do you have any other remarks (contact form to be sent to email server; not part of the data collection of the study)
- thank you!

6.4 Reminders

Parents are sent reminders if the filled questionnaire is not received by Lareb.

- after Q1 (day 5), reminder at day 7, valid time of return: day 8
- after Q2 (day 14), reminder at day 16, valid time of return: day 18
- after Q3 (day 28), reminder at day 32, valid time of return: day 36.

Questionnaires received after the valid time of return will not be included in the Per Protocol analysis.

6.5 Measurements

If parents suspect their child to have fever, they are asked to measure body temperature rectally with their own thermometer. It is therefore assumed that all reported temperature values are from rectal measurements. In the current setup of the study there is no possibility to verify if this is actually done. However, we assume that temperature values obtained otherwise, will be similarly distributed over both groups of ‘old’ and ‘new’ vaccine recipients.

CHAPTER 7. STUDY PRODUCTS

7.1 Description of study products

All vaccines used in this investigation are regularly licensed products, currently used in the NIP. Their good record of safety and effectiveness is well established. For details of both products, see the SmPC.

NeisVac-C® vaccine is a suspension for injection in a pre-filled syringe of 0,5 ml. It is a conjugated meningococcal group C-polysaccharide vaccine (adsorbed). RVG number 26343. One dose (0,5 ml) contains *Neisseria-meningitidis* polysaccharide (O-deacetylated) of group C (C11-strain) 10 microgram conjugated to tetanus toxoïd 10 – 20 microgram, adsorbed to aluminumhydroxide, hydrated 0,5 mg Al3+. The product further contains sodium chloride and water for injections.

Originally, NeisVac-C® was manufactured in USA. Baxter Ltd has decided to continue production in Austria. The change of production site was approved by EMA, as all in process tests and preclinical tests showed no deviations. License was continued, with the advice that a Post-Authorization Safety Study should be performed, which is the subject of the study described in this protocol.
Baby clinics are randomised and supplied with either product (new vs old production facility, product A vs product B).

Simultaneously, all children are given a MMR (measles-mumps-rubella vaccine, live attenuated).
M-M-R-VAXPRO® powder and solvent for suspension for injection. Measles-Mumps-Rubella vaccine (live). EMA number EU/1/06/337/001.
After reconstitution one dose (0,5 ml) contains:
- Measles virus[1] Enders' Edmonston strain (live, attenuated), no less than $1x10^3$ CCID50*
- Mumps virus[1] Jeryl Lynn™ [Level B] strain (live, attenuated), no less than $12,5x10^3$ CCID50*

Rubellavirus[2] Wistar RA 27/3 strain (live, attenuated), no less than $1x10^3$ CCID50*
* 50% cell culture infectious dose.
[1] produced in chicken embryo cells. [2] produced in WI-38 human diploïd lung fibroblasts.
The vaccine may contain traces of recombinant human albumine (rHA). The vaccine contains traces of neomycine. The vaccine contains 14,5 mg sorbitol, and further sodium phosphate, potassium phosphate, sucrose, hydrolysee gelatin, Medium 199 with Hanks salts, minimum essential medium Eagle (MEM), monosodium L-glutamate, phenol red , sodium bicarbonate, sodium chloride and hydrochloric acid to obtain desired acidity.

At present M-M-R-VaxPro® (Sanofi Pasteur MSD) is used in the NIP. Throughout the present study, it is foreseen that this vaccine is the only brand used. Children may receive any batch of the MMR vaccine, and are not vaccinated with specified batches.

7.2 Description of route of administration and dosage

NeisVac-C® will be injected intramuscularly (one dose is 0,5 ml). MMR will be injected subcutaneously (one dose is 0,5 ml, after reconstitution).
Injection sites are not prescribed in this protocol. The baby clinic personnel is asked to follow their usual practice. Neither the various organisations of child health care, nor the National Immunisation Programme prescribe the injection sites.
After injection, baby clinic staff is asked to write down which vaccine is injected where, and which batch numbers were used. Parents are asked to enter these data into the questionnaires.

7.3 Preparation, packaging, labeling and storage

All study products are packed and labeled according to their license. They are further distinguished by their batch numbers.
Transport and storage are done according to the standards of the NIP.

7.4 Product shipment, accountability

All baby clinics in the study region will receive only one batch of NeisVac-C® during the course of the study (one year). Any next shipment to a baby clinic should ensure that the right batch is delivered. From these shipments, also children who do not participate, will receive their NeisVac-C® vaccinations.
For MMR vaccine, there is no preference for particular batches to be used per clinic.
Throughout the study period, the baby clinics will be (re)supplied regularly (every 2-3 months).
Accountability is not a requirement for this study. However, the number of doses per baby clinic is recorded as part of the usual administration and surveillance of the NIP.
It may occur rarely that vaccines from one baby clinic are transferred to another. Therefore we verify the batch numbers by asking the parents.

CHAPTER 8. STUDY GROUPS, RANDOMISATION AND (UN-) BLINDING

8.1 Study groups
The study comprises of two groups, according to the NeisVac-C® vaccine that has been given. The parents will not be informed which product (old vs new) their child has received.

8.2 Randomization and stratification
Central randomisation: baby clinics are randomly assigned to use either the old or the new NeisVac-C®. Shipments are done according to a distribution list, generated to randomly assign either batch to any participating baby clinic, to ensure that both groups are similar in size, and that local differences in confounding factors (e.g. concurrent infections, habits and attitudes towards vaccinations, …etc) are balanced out.
RIVM/DVP will generate the distribution list, and will provide the baby clinics accordingly (Appendix 2).

8.3 Study size and recruitment population.
In order to evaluate 1215 children per group according to protocol (see paragraph 9.2) we need to recruit higher numbers of children. The following estimations are done to determine the population size to target for participation. With 1215 per group we need 2430 fully evaluable and complete datasets. We assume that 90% of data from an estimated 2700 respondents that have completed all questionnaires are evaluable. We assume that after starting the questionnaires 75% of responders will also complete the last questionnaire, thus giving an estimation of 3600 responding parents that start and agree to participation. Experience of RIVM indicates that about 40% of parents participate to a similar questionnaire proposal. However, to be sure to include the desired number of respondents we assume a lower percentage: 15-20%. Thus we assume that 18.000 to 24.000 parents need to be addressed for taking part in the investigation. In addition, to compensate for other interferences or loss, we intend to address 25.000 parents.

8.4 (Un) blinding procedure
Not applicable

CHAPTER 9. STATISTICS

9.1 Statistical formulation of the study hypothesis

The primary endpoint in this study is defined as the number of children with fever reactions within 4 days after vaccination with NeisVac-C®. Fever defined as rectally measured 38º Celsius or higher.

Kroesbergen et al found in a study following 863 Dutch children that fever occurred in 46 (5,3%; 95% CI 3,83-8,83) of children at day 0, 1 and/or 2. Fever at days 3 and 4 is not mentioned. A graph shows that the daily prevalence of fever is about max 3,5%. From day 5 the % of children with fever is rising to 19% at day 9 (ascribed to simultaneously given MMR vaccine).
Our primary endpoint is based on pooled daily presence of fever from days 0 to 4 after vaccination. Based on Kroesbergen et al we expect this to occur in about 6% of the children (95% CI 4,44-7,71, when 52/863 cases).

Baby clinics will be randomized to vaccinate with either product
- investigational new product Orth/Donau (Austria) = Active = Group A
- comparator old product Beltsville (US) = Reference = Group B

Randomisation procedure: to be described

9.2 Sample size calculation

For a non-inferiority approach the following assumptions and choices were made:
- null hypothesis: group A receiving the new product, has a higher prevalence of fever than children in group B. The risk ratio between group A and B is 1.5 or more.
- alternative hypothesis (one-sided): the risk ratio between group B and group A is less than 1.5.
- study groups equally sized (approximately)
- type 1 error alpha: rejection null hypothesis when it is actually true, is set at 0,025, one-sided (ICH E9)
- type 2 error beta: rejection null hypothesis, is set at 0,80
- clinically acceptable or meaningful difference between endpoints in group A and group B: risk ratio of 1.5 indicating a higher risk of fever in group A (corresponding with a fever prevalence of 9% in group A).
- sample size is calculated to obtain numbers needed for the Per Protocol Analysis set.

Prior data indicate that the prevalence of fever among the reference group B is about 6%. Sample size was calculated using a web-based sample size calculator Sealed Envelope, based on Blackwelder. We will need to study 1215 children in each group to be able to reject the null hypothesis that the fever rates between in group A are higher than in group B, with probability (power) of at least 0,8. The type I error probability associated with this test of this null hypothesis is 0,05 (two-sided, 0.025% one-sided). If there is truly no difference in fever prevalence between the two groups, then 2430 children are required to be 80% sure that the upper limit of a one-sided 97.5% confidence interval (or equivalently a 95% two-sided confidence interval) of the risk ratio will exclude a difference in favour of the reference group of more than 3%.

9.3 Analysis sets

Participants that violate the protocol are excluded from the Per-Protocol Analysis Set. Protocol violations involve:
- not fulfilling criteria for inclusion and exclusion
- major deficiencies / discrepancies in the completion of questionnaire 1.
- non-adherence to pre-specified timelines.
- any concomitant vaccinations and other potential confounder that may interfere with the reactogenicity under study, or with the interpretation of the data.

As the electronic questionnaires enforce most criteria above, in the majority of cases decisions to exclude from the PPA data set are done automatically.

9.4 Descriptive statistics

Descriptive characteristics will be presented per study group, as numbers and/or proportions of children with an applicable parameter per group. The results will be displayed per group. For continuous variables, data will be presented as mean ± SD (confidence interval) and proportions, and median (range). For binary variables data will be presented as n/N (%), and 95% CI. The secondary endpoints will only be descriptively analysed.

9.5 Comparability of study groups

Both study groups should be comparable for baseline characteristics and potential confounders, as defined in Chapter 5.

9.6 Statistical methods for data analysis

The primary endpoint of the study, fever cases observed within 4 days after vaccination will be analyzed using logistic regression with vaccination groups (“old” / “new” product) and potential confounders as listed in 5.3 as explanatory factors, applying a log link in order to obtain relative risk estimates at the end. Relative risk and its 95% CI of occurrence of fever cases with the “new” and “old” NeisVac-C® product will be calculated from the regression model assessing a potential increase of fever reactions with the “new” product. If the upper limit of the 95% CI is below 1.5 then the “new” product is considered to be non-inferior to the old one as far as fever reaction is concerned.
The secondary endpoints will be analyzed similarly and descriptive without the non-inferiority considerations.

CHAPTER 10. SAFETY REPORTING

10.1 Definitions
All symptoms, reported by the parents occurred during the study period will be considered as adverse event.

A Serious Adverse Event (SAE) is any untoward medical occurrence or effect that at any dose:
- results in death
- is life-threatening (at the time of the event)
- requires hospitalisation or prolongation of existing inpatients' hospitalisation
- results in persistent or significant disability or incapacity
- is otherwise medically important.

A SAE is defined as a *Suspected Serious Adverse Reaction (SSAR)* if it is:
- judged to be at least possibly related to the study product by investigator.
- expected (listed in the Summary of Product Characteristics.

A SAE is defined as a *Suspected Unexpected Serious Adverse Reactions (SUSARs)* if it is:
- judged to be at least possibly related to the study product by either investigator or sponsor, and
- unexpected (not listed in the SmPC).

The following events are not regarded as SAE:
- Hospitalisation that was already planned or anticipated before participation to the study.
- Any SAE occurred before vaccination.

However, prolongation of these hospitalisations due to complications or other aspects mentioned in the definition of an SAE must be handled as an SAE.

10.2 (Serious) Adverse Events handling

10.2.1 Recording of (S)AEs
In case of an SAE, the event will be transferred to the current Spontaneous Reporting System: the case will be entered in the Lareb database, and will be reviewed and processed as a regular report with the exception that it will not be forwarded to Eudravigilance.

10.2.2 SAE reporting
The content of filled questionnaires, including any reported non-serious and serious events, will be transferred electronically to Baxter Ltd on a weekly basis, after coding by a Lareb investigator.
Serious AEs are reported to Baxter within 2 business days or 4 calendar days if a weekend is included. Potential signals will be reported within 3 business days or within 5 calendar days if a weekend is included.

10.2.3 Follow-up of (S)AEs
All SAEs are followed by Lareb until they have abated, or until a stable situation has been reached. This will be done according the usual Lareb standards.

10.3 New relevant safety information
The investigator will inform the parents of the toddlers, the authorities including the National Public Health Institute responsible for the National Immunisation Programme, and the applicable manufacturers if anything occurs that may negatively affect the burden or risks of participation as foreseen in the research proposal. The study may be suspended pending further review by the Lareb Scientific Advisory Board, provided that suspension does not jeopardise the toddlers' health. Lareb will take care that parents of all participating toddlers are kept informed.

CHAPTER 11. ETHICAL CONSIDERATIONS

11.1 Basic principles and regulations

The study team must ensure that this study is conducted in full conformance with the principles of the 'World Medical Association Declaration of Helsinki' (18th WMA General Assembly, Helsinki, Finland, June 1964, with last amendments by the 64th WMA General Assembly, Fortaleza, Brazil, October 2013), ICH guidelines for Good Clinical Practice as appropriate to pharmaceutical products, and local legislation of the country in which the research is conducted, whichever affords the greater protection to the individual.

11.2 Ethics Committee

This protocol and any accompanying informational material provided to the parents of the toddlers, will be submitted to the applicable Ethics Committee by the investigator according to local legislation. The Ethics Committee will firstly decide whether the proposed study fullfills criteria that necessitates a full review according to the Netherlands law on Investigations in humans (WMO). If so, approval from the Ethics Committee must be obtained before starting the study. Approval should be documented in a letter to the investigator specifying the date on which the Ethics Committee met and granted the approval, the composition of the Ethics Committee, and version and date of all submitted documents. If a formal review is not required for this non-invasive, non-interventional questionnaire based study, the Ethics Committee is asked for a Waiver letter.

11.3 Informed Consent

Prior to study enrolment, no written informed consent is obtained from the parents of each toddler. Instead, based on the information in the letter and/or flyer, they report themselves to participate by giving their email adress, and by agreeing to the Lareb privacy policy.

11.4 Confidentiality of study data

The investigator is responsible for treating subject and study information as confidential. The investigator should ensure that the subject identity will not be made publicly available. All subject study records are identified only by the subject identification number to maintain subjects' confidentiality. Identification codes lists that link the subjects' names to the subjects' identification number must be stored in the Investigator Site File.

11.5 Benefits and risks assessment

No known or potential risks are expected from the use of the study products, other than those applicable for the national Immunisation Programme.

11.6 Incentives/Compensation for subjects

Neither parents nor toddlers will receive a financial or other compensation for their participation in the study.

11.7 Insurance

Not applicable.

CHAPTER 12. ADMINISTRATIVE ASPECTS AND PUBLICATION

12.1 Roles and responsibilities
During the study a study team will be available on telephone to assist parents in filling the questionnaires.

12.2 Source data
All information obtained during the study will be directly recorded in an electronic database. Manually coded entries from free text field can be verified.

12.3 Data handling
All study data are kept on the central Lareb server. Backups are made daily.
Investigators are mandated to
- edit data if they are redundant,
- to code reactions according to MedDRA terminology from free text field
A data trail is formed by logging any changes.

12.4 Criteria for premature termination of the study
The study may be terminated as signals arise that would make continuation unsafe. As the vaccines under study are in general use, such situation may also have consequences beyond the scope of the study and its participants.

12.5 Publication policy
Lareb is free to publish any data obtained from the study. Results of the study will be published in a scientific journal.

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