Statistical Analysis Plan

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CTC study number:	CTC14931
Study title:	An open, single center Phase I trial to assess the safety, tolerability and immunogenicity of two ascending doses of the candidate vaccine MVA-MERS-S
Test product:	MVA-MERS-S
Development phase:	Phase I
Sponsor / affiliate:	University Medical Center Hamburg-Eppendorf Martinistr. 52 20246 Hamburg, Germany
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1 Version Control

Version	Date	Section	Description
Final v01	28MAY2019	ALL	Initial release

2 Statistical Analysis Plan Approval

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Sponsor approval:

Sponsor representative University Medical Center Hamburg-Eppendorf, Hamburg, Germany	Prof. Marylyn M. Addo, MD, PhD, MSc, DTM&H	28 May 2019
	Name	Date, Signature

3 Table of Contents

4 Abbreviations and Definitions

AE	Adverse event
ANOVA	Analysis of variance
ATC	Anatomical therapeutic chemical
BMI	Body mass index
CI	Confidence interval
CRF	Case report form
CRO	Clinical research organization
CV	Coefficient of variation
DRM	Data review meeting
ECG	Electrocardiogram
ELISA	Enzyme-linked Immunosorbent Assay
GeoM	Geometric mean
GMT	Geometric mean titer
GSD	Geometric standard deviation
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
Max	Maximum
Mean	Arithmetic mean
Med	Median
MedDRA	Medical Dictionary for Regulatory Activities
Min	Minimum
PP	Per protocol set
PT	Preferred term

QC	Quality control
SAE	Serious Adverse Event
SAF	Safety set
SD	Standard deviation
SOC	System organ class

5 Introduction

This statistical analysis plan is in accordance with the statistical methods planned in the final version of the protocol for study UKE-DZIF1-MVA-MERS-S, dated 27Nov2018 (version 6.0). It follows the principles of the Guidelines ICH Topic E3 [1] and ICH Topic E9 [2]. It gives all details for the final statistical analysis of this study.

The parts written in italic font are original wording from the protocol.

Protocol: *This study is designed to assess the overall tolerability and safety of two ascending doses of the experimental MVA-MERS-S vaccine (10^7 pfu and 10^8 pfu) administered to healthy subjects and to evaluate the reactogenicity after administration of two dosage levels of MVA-MERS-S.*

The following persons will perform the statistical analysis (including SAS® programming), the biometrical interpretation, the biometrical reporting and the internal quality control:

Principal investigator:	Prof. Marylyn M. Addo
Project manager:	Laura Kaltenberg
Responsible project biometrician:	Angelika Böhm
Study data manager:	Noelia Carabelos

6 Study Objectives and Endpoints

6.1 Study Objectives

Protocol: *Primary objective:*

- *To evaluate the safety and tolerability of two dosage levels of the experimental vaccine MVA-MERS-S.*
- *To evaluate the reactogenicity after administration of two dosage levels of MVA-MERS-S.*

Secondary objectives:

- *To evaluate MERS-S-specific antibody responses induced by two dosage levels of MVA-MERS-S.*

Exploratory objectives:

- *To evaluate MERS-S-specific cellular immune responses after administration of MVA-MERS-S.*
- *To characterize MERS-S-induced B and T cell memory responses.*
- *To evaluate innate cell subset phenotypes and function induced by MVA-MERS-S.*
- *To evaluate early innate immunity gene expression signatures induced by MVA-MERS-S.*
- *To comprehensively investigate vaccine-induced humoral immune responses and antibody functions.*
- To analyse 3rd boost vaccination

6.2 Endpoints

Protocol: *Primary endpoints*

The nature frequency and severity of adverse events associated with MVA-MERS-S vaccine will be collected and measured as followed:

- *Occurrence of solicited local reactogenicity signs and symptoms for 14 days after vaccination.*
- *Occurrence of solicited systemic reactogenicity signs and symptoms for 14 days after vaccination.*
- *Occurrence of unsolicited adverse events (AE) for 28 days after vaccination.*
- *Change from baseline of safety laboratory measures.*
- *Occurrence of serious adverse events (SAE) throughout the study period.*

Secondary endpoints

Immunogenicity

- *Humoral immunity: Magnitude of MVA-MERS-S antibody responses.*

7 Study Methods

7.1 General Study Design and Plan

Protocol: *This is an open, single-center phase I dose escalation trial in 24 healthy subjects of both sexes. Subjects will be allocated to two different dose cohorts each receiving two vaccine injections, one at day 0 and another on day 28. For safety reasons, subjects will be vaccinated in both cohorts in a staggered manner. Dosing of subjects will commence in dosing cohort 1 (1x10^7 pfu) followed by dosing cohort 2 (1x10^8 pfu). Dosing of cohort 2 will not start before both vaccinations and safety assessments of the first dose cohort are completed. […] In addition, irrespective of cohort, eligible subjects that consent to participate in and are eligible for a 3rd boost vaccination will receive a 3rd single dose of 1 x 10^8 pfu MVA-MERS-S in 0.5mL (the total injected volume will then be 1.5mL).*

The total study duration will cover a period of approximately 13 months. For subjects who gave consent for a 3rd vaccination the total study duration will cover a period of approximately 18 months.

Activities during the study are outlined in Table 1a in Section 2 in the protocol.

7.2 Inclusion-Exclusion Criteria

24 healthy male and female subjects between 18 and 55 of age with a body mass index (BMI) between 18.5 – 30.0 kg/m^2 (inclusive) will be screened and included into the trial.

The detailed inclusion and exclusion criteria are presented in Section 9.3 of the protocol.

7.3 Randomisation and Blinding

This is an open-label study and therefore blinding is not necessary. Furthermore, no randomization is planned for this study.

7.4 Sample Size

The planned sample size of 24 subjects will be separated into 2 cohorts with 12 subjects per cohort.

Detailed information about the determination of sample size are presented in Section 9.8.2 in the protocol.

7.5 Study Variables

For the detailed study variables see Table 1 in Section 2 in the protocol. The detailed times and events are presented in Table 2a in the protocol.

7.6 Interim Analysis

Protocol: *Interim analyses may be performed upon request of the sponsor.*

8 General Considerations

The final analysis will be performed after the Data Review Meeting (DRM) and after the data base lock.

8.1 Analysis Sets

The following analysis sets are defined for this trial (see Section 9.8.1 in the protocol).

- Safety Set (SAF):
  Protocol: *All subjects receiving at least once the study medication will be included into the safety evaluation (safety collective).*
- Per protocol Set (PP):
  Protocol: *All subjects who have received both injections and who have not withdrawn within the first 7 days after the second injection will be included for the per protocol analysis.*

Protocol: *All safety endpoints will be investigated in the safety population. As a sensitivity analysis, the analysis of the safety endpoints will be repeated in the per protocol population. All other endpoints will be investigated in the per protocol population only.*

8.2 Data Review Meeting

The decision on the allocation of subjects to populations will be made at the DRM.

8.3 Covariates and Subgroups

Not applicable.

8.4 Missing Data

In general, missing data will not be imputed. All data recorded in the CRF will be analysed.

8.5 Baseline Definition

Protocol: *Unless otherwise specified, baseline is defined as the time-point closest to but prior to the first administration of the vaccine.*

9 Statistical Analysis

9.1 Specification Related to the Whole Analysis

Statistical analyses are done on the data from the first and second vaccination. The third boost vaccination is one of the exploratory objectives and not part of this SAP.

9.1.1 Tables

Categorical data will be summarised by dose level with number of missing values, frequencies, and percentages. For metric data, the basic statistics containing number of subjects, number of missing values, the arithmetic mean (Mean), standard deviation (SD), absolute minimum (Min) and maximum (Max), coefficient of variation (CV) and median (Med) will be reported per dose level. In addition, the geometric mean (GeoM) and the coefficient of variation (CV) will be computed, if parameters appear to be log-normally distributed. Additional 95% confidence intervals may be calculated for means (metric data) and percentages (categorical data), if deemed appropriate.

9.1.2 Listings

CRF data will be listed and documented as well as all relevant generated and transformed variables. In all listings, the dose level and analysis set for each subject will be included. The listings will be sorted by dose level, subject identifier, visit, and time, if applicable.

9.1.3 Calculation of Non-Pharmacokinetic Variables

Age will be calculated as time between date of birth and date of written informed consent. BMI will be calculated as weight (kg)/[height (m)]².

9.2 Subject Disposition

The number of enrolled subjects, the number of subjects in each analysis set and the number who withdrew prior to completing the study as well as the reasons for exclusion from each analysis set will be summarised in total and by dose level.

9.3 Demographics and Baseline Characteristics

Demographics and other baseline data will be presented and summarised by dose level for the safety set.

9.4 Medical history

Medical history will be listed by subject and grouped by body system and medical event for the SAF.

9.5 Prior and Concomitant Medications

Prior medications are defined as medications with a start date prior to the date of the first vaccination. If the start date is missing, medications are defined as prior only when the stop date is earlier than the first vaccination. Concomitant medications are defined as medications with an end date on or after the date of the vaccination or that are ongoing.

There is no Drug coding planned.

All medication (i.e., prior and concomitant) will be summarised for each dose level by indication and substance. Furthermore they will be listed by subjects for the safety set.

9.6 Treatment Compliance

The assessments whether or not the vaccination maneuvers have been correctly performed will be listed for the PP.

9.7 Immunogenicity Analyses

The main immunogenicity endpoint is the geometric mean MVA-MERS-ELISA antibody endpoint titers (GMT) on study Days 0, 7, 14, 28, 35, 42, 56, 84, 180. MVA-MERS results are captured on the CRF in ELISA Assay Results and ELISA Units/mL based on testing performed at the Institute for Virology in Marburg, Germany.

Further immunogenicity endpoints are not planned to be analysed. The sponsor may decide at later stage to analyse one of the further immunogenicity endpoints. In this case the SAP will be amended.

The results from exploratory objectives may not be part of the clinical study report (CSR).

Only main immunogenicity endpoints will be analysed in the PP set. All values will be reported in data listings, if available.

Main immunogenicity endpoints will be summarised at study Days 0, 7, 14, 28, 35, 42, 56, 84, 180, by dose level as the mean of log (base 10), exponentiated back into the original scale. The geometric standard deviation (GSD) and 95% CI for the endpoint will be obtained by exponentiating the standard deviation and 95% CI for the mean of log (base 10) transformed endpoint values, respectively. Figures will be generated to display scatter plots for the log (base 10) transformed endpoints at baseline and analysis Days 7, 14, 28, 35, 42, 56, 84, and 180 for each vaccine dose level and will include the mean and the 95% CI. An analysis of variance (ANOVA) regression model will be used to compare log (base 10) transformed MVA-MERS titer at analysis Days 28 between the two dose levels.

Protocol: *Seroconversion will be defined as at least a two-fold rise in titers, or becoming seropositive in originally seronegative subjects.*

Seropositivity rates of MVA-MERS ELISA endpoint titers at Days 0, 7, 14, 28, 35, 42, 56, 84, 180 based on the definition of Seroconversion will be summarised separately for each dose level. Fisher's exact test will be used to compare rates between the two dose levels for Days 0, 7, 14, 28, 35, 42, 56, 84, 180.

9.8 Safety Analyses

The safety assessments will be evaluated based on the safety set with respect to the following parameters:

- Adverse Events (AEs)
  Protocol: *The adverse events will be listed per subject using MedDRA terminology (preferred term and system organ class) and will be reported in tables summarizing the frequency of subjects with adverse events and adverse events by dosage level, by dose level and injection and by body system, the number of adverse events and number of subjects with adverse events by dosage level, by dosage level and injection, and the characteristics of adverse events.[…] A change in the value of a safety laboratory investigation can represent an AE if the change is clinically relevant or if, during treatment with the investigational medicinal product, a shift of a parameter is observed from a normal to an abnormal value, or a further worsening of an already abnormal value. […] For the hematology, clinical laboratory and the urine analysis deviations from the reference ranges will be summarized in frequency tables. […] A solicited AE is a predetermined event occurring within 14 days after application of IMP, which may reflect safety concerns related to the investigational product.*
  Analyses of solicited local and systemic AEs will be performed for the time period until 14 days following vaccination. Analyses of unsolicited AEs will be performed for those events with onset within 28 days after vaccination. Solicited and unsolicited AEs are described in Section 9.3.3.3 in the Protocol.
  Additionally, AEs will be summarised by severity, grouped by system organ class (SOC) and preferred term (PT). The severity categorization of AEs are described in Section 9.7.2 in the Protocol.
  Subjects with serious adverse events (SAEs) will be listed and summarised.

- Laboratory parameters
  Protocol: *All relevant clinical variables obtained during screening, treatment phase or final examination will be reported in appropriate tables together with descriptive statistics. Clinical laboratory findings outside of the reference range will be flagged.*
  Change from baseline will be summarised and displayed graphically by dose level, if deemed appropriate.

- Physical examination
  Physical examination will be individually listed and summarised in tables.

- Vital signs
  Protocol: *For blood pressure and pulse rate descriptive statistics will be listed by sampling times (screening and follow up) according to the data reported in the source documents.*

- ECG
  Protocol: *The results of the 12 lead ECG will be listed by sampling times (screening and follow up) according to the data reported in the source documents.*
  ECG will be summarized descriptively.

9.9 Interim Analyses

Protocol: *Interim analyses may be performed upon request of the sponsor.*

9.10 Other Analyses

Not applicable.

10 Reporting Conventions

The minimum, maximum, mean, and any other statistics, will be reported to the same number of decimal places as the original data. The standard deviation will be reported to one decimal place greater than the original data. P-values will be reported to three (3) decimal places; p-values less than 0.001 will be reported as “<0.001”.

11 Technical Details

The analysis will be carried out according to SOP001_PROGRAMMING_Staburo_GmbH [3]. The statistical analysis will be performed using the SAS® 9.4 or higher.

SAS programming will be performed according to Staburo GmbH standards as defined in SOP001_PROGRAMMING_Staburo_GmbH [3] and related work instructions. Special attention will be paid to planning and performance of quality control measures as documented in the QC plan for the analysis of this study (see also SOP002_QC_SAS_Staburo_GmbH [4]).

12 Summary of Changes to the Protocol

Not applicable.

13 Contents of Tables, Figures and Listings

Based on this SAP, an excel file is developed which covers all tables, figures and listings to be included in the Clinical study report. This document is appended to this SAP and will be available upon request. The statistical output is organized in 14 Tables, figures and graphs referred to but not included in the text

14.1 Demographic data and baseline characteristics

14.2 Immunogenicity

14.3 Safety data

The listings will be available in section 16.2

16.2 Subject data listings

The Table of content of the Table, Listings and Figures will be adapted to incorporate additional data not foreseen at the moment (e.g., listings of deaths, other serious and significant AEs and narratives of deaths, other serious and certain other significant AEs), if applicable.

14 References

[1] Note for Guidance on Structure and Content of Clinical Study Reports (ICH E3) CPMP/ICH/137/95, 1996.

[2] Note for Guidance on Statistical Principles for Clinical Trials (ICH E9) CPMP/ICH/363/96, 1998.

[3] SOP001_PROGRAMMING_Staburo_GmbH, “Standard Operating Procedure for Statistical Programming”, current version

[4] SOP002_QC_SAS_Staburo_GmbH, “Standard Operating Procedure for Quality Control of SAS® programs”, current version