Janssen Vaccines & Prevention B.V.*

Clinical Protocol

A Randomized, Observer-blind, Placebo-controlled, Phase 2 Study to Evaluate the Safety, Tolerability and Immunogenicity of Three Prime-boost Regimens of the Candidate Prophylactic Vaccines for Ebola Ad26.ZEBOV and MVA-BN-Filo in Healthy Adults in Europe

Protocol VAC52150EBL2001; Phase 2
AMENDMENT 5

Innovative Medicines Initiative-2 EBOVAC2 Consortium Partners
(London School of Hygiene and Tropical Medicine,
Institut National de la Santé et de la Recherche Médicale,
University of Oxford, Le Centre MURAZ, and Janssen Vaccines & Prevention B.V.)

VAC52150 (Ad26.ZEBOV/MVA-BN-Filo [MVA-mBN226B])

*Janssen Vaccines & Prevention B.V. (formerly known as Crucell Holland B.V.) is a Janssen pharmaceutical company of Johnson & Johnson and is hereafter referred to as the sponsor of the study.

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GCP Compliance: This study will be conducted in compliance with Good Clinical Practice, and applicable regulatory requirements.

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PROTOCOL AMENDMENTS

Protocol Version	Issue Date	Amendment Type
Original Protocol	27 March 2015	-
Amendment 1	07 May 2015	Substantial
Amendment 2	18 August 2015	Substantial
Amendment 3	26 January 2016	Substantial
Amendment 4	01 September 2016	Substantial
Amendment 5	20 April 2017	Substantial

Amendments below are listed beginning with the most recent amendment.

Amendment_5 (this document)

This amendment is considered to be substantial based on the criteria set forth in Article 10(a) of Directive 2001/20/EC of the European Parliament and the Council of the European Union.

The overall reason for the amendment: This amendment was created due to significant delays in scheduled boost vaccinations caused by study pauses required for safety evaluations. Since many subjects in France have had no boost vaccination and many subjects in the United Kingdom (UK) have had a late boost vaccination, it will be very difficult to evaluate the planned dosing regimens. Therefore, no further subjects will be recruited in the entire study (ie, UK and France). Additional data on the planned dosing regimens will be obtained from other studies. Vaccinated subjects in both countries will still be followed per protocol for safety.

The changes made to the clinical protocol VAC52150EBL2001 are listed below, including the rationale of each change and a list of all applicable sections.

Rationale: Due to significant delays in scheduled boost vaccinations caused by study pauses required for safety evaluations, enrollment in the entire study (ie, UK and France) will not be resumed. Since many subjects in France have had no boost vaccinations and many subjects in the UK have had a late boost vaccination, it will be very difficult to evaluate the planned dosing regimens. Therefore, no further subjects will be recruited in the entire study (ie, UK and France).

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3 STUDY DESIGN AND RATIONALE

5 TREATMENT ALLOCATION AND BLINDING

9.1.2 Screening Phase

10.2 Discontinuation of Study Vaccination/Withdrawal From the Study

11.2 Sample Size Determination

Rationale: Subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study VAC52150EBL4001 is opened, will have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months (whichever comes first) or discontinued earlier. Information was added to clarify the procedures that need to be followed for these subjects.

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9.1.1 Overview

9.1.5 Open-label Long-term Follow-up Phase

9.2.2 Safety Assessments

10.1 Completion

Rationale: The secondary objectives and endpoints were limited to immune responses measured by ELISA at 21 days post boost. Immune responses measured by ELISA at other relevant time points were shifted to the exploratory objectives and endpoints. As the exploratory endpoints can be presented in a separate biomarker report, there will be no delay in the preparation of the final clinical study report.

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2.1 Objectives

9.3.1 Immunogenicity Endpoints

Rationale: PBMC and serum samples for exploratory immunogenicity assessments collected in France may be analyzed in the UK and vice versa. Samples will be shared between clinical sites to allow exploratory immunogenicity endpoint analyses with the reduced subject numbers.

9.3.2 Immunogenicity Assessments

Rationale: As a result of the pause, the gap between the completion dates of the 2 countries (UK and France) will be smaller than originally expected. Therefore, it is no longer necessary to unblind by country.

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3.1 Overview of Study Design

5 TREATMENT ALLOCATION AND BLINDING

9.1.5 Open-label Long-term Follow-up Phase

Rationale: A statement has been added that due to delays in scheduled boost vaccinations caused by study pauses, the primary and final statistical analysis may be combined.

11 STATISTICAL METHODS

Rationale: The statement that pre-specified decision criteria will be used for vaccination schedule selection, based on immune response results and absence of safety concerns, was removed. The majority of the studies were designed in parallel because of the outbreak of the Ebola virus disease and decisions were made based on available data instead.

3.2 Study Design Rationale

Rationale: The statistical methods for clinical laboratory tests, vital signs, ECGs, and physical examination were revised. Since only small mean fluctuations are expected, limited interest will be on the summary statistics over time and the analyses will focus on worst abnormalities and toxicity gradings.

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11.4 Safety Analyses

Rationale: Information was added regarding the immunogenicity analysis.

11.1 Analysis Sets

Rationale: Minor textual changes have been made, in addition to updates to be in line with other current protocols.

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11 STATISTICAL METHODS

Rationale: The protocol has been updated to be in line with the current protocol template (version 1 November 2016).

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12.1.1 Adverse Event Definitions and Classifications

Amendment_4 (01 September 2016)

This amendment is considered to be substantial based on the criteria set forth in Article 10(a) of Directive 2001/20/EC of the European Parliament and the Council of the European Union.

The overall reason for the amendment: The sponsor halted vaccinations following a case of Miller Fisher syndrome after receipt of MVA-BN-Filo or placebo in this clinical study, until a revised ICF containing updated safety language for the current study VAC52150EBL2001 was prepared and approval to restart the study was granted by the relevant competent authority. As a result of the pause, some subjects will be outside the protocol-defined boost vaccination window. Information was added to clarify the procedures that need to be followed for these subjects. As requested by the Agence Nationale de Sécurité du Médicament et des produits de santé (ANSM), wording on the collection of Immediate Reportable Events was added after observation of the case of Miller Fisher syndrome. Randomization to Group 3 will be stopped to focus on the schedules for which an indication will be sought.

The changes made to the clinical protocol VAC52150EBL2001 are listed below, including the rationale of each change and a list of all applicable sections.

Rationale: As requested by the ANSM, wording on the collection of "Immediate Reportable Events" was added after one subject in this study experienced a serious and very rare condition called "Miller Fisher syndrome" about a month after boost vaccination with either MVA-BN-Filo or placebo.

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12.1.1 Adverse Event Definitions and Classifications
12.2 Special Reporting Situations
12.3.1 All Adverse Events
12.3.3 Immediate Reportable Events

Rationale: As a result of the pause, some subjects will be outside the protocol-defined boost vaccination window. Information was added to clarify the procedures that need to be followed for these subjects.

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Additional Time and Events Schedule
3.1 Overview of Study Design
11.5 Immunogenicity Analyses

Rationale: As a result of the pause, subjects whose screening period was longer than the protocol-defined 12 weeks will be allowed to rescreen once, but must have safety laboratory assessments (ie, serum chemistry and hematology, troponin I testing, serology, pregnancy testing, urinalysis) within 28 days of the prime vaccination.

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9.1.2 Screening Phase

Rationale: Randomization to Group 3 will be stopped to focus on the schedules for which an indication will be sought.

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3.2 Study Design Rationale
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10.2 Discontinuation of Study Vaccination/Withdrawal From the Study
11.2 Sample Size Determination

Rationale: The percentage of subjects who will have blood samples taken for assessment of cellular immune responses was decreased from a target of 50% to 10% of all subjects.

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Time and Events Schedule
3.1 Overview of Study Design
9.3.2 Immunogenicity Assessments

Rationale: It was clarified that in France, blood samples will be collected from and analyzed in no more than 25 subjects in Groups 1 and 2. Collection and analysis of blood samples in France in Group 3 was deleted, because randomization to this group will be stopped.

Additional Time and Events Schedule
9.3.2 Immunogenicity Assessments

Rationale: Adverse events of special interest (cardiovascular events) will no longer be collected as no cardiovascular events have been associated with the current MVA-BN-Filo vaccine. Information was added on the procedure that needs to be followed in case any cardiac sign or symptom develops after the boost vaccination.

1.2.5 Overall Benefit/Risk Assessment
9.2.2 Safety Assessments
12.2 Special Reporting Situations

Rationale: Further details regarding enrollment into the VAC52150 roll-over study have been added, such as the inclusion of placebo subjects before unblinding of the current study.

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3.1 Overview of Study Design
9.1.6 VAC52150 Vaccine Development Roll-over Study
12.3.4 Pregnancy

Rationale: All secondary immunogenicity objectives and endpoints were shifted to exploratory objectives and endpoints, except for immune responses measured by ELISA.

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2.1 Objectives
9.3.1 Immunogenicity Endpoints

Rationale: Safety information following MVA-BN-Filo vaccine administration based on the pooled safety data from studies VAC52150EBL1001 and VAC52150EBL1002 has been included.

1.1.2.2 Safety Profile of MVA-BN-based Vaccines
REFERENCES

Rationale: The name of the sponsor changed from Crucell Holland B.V. to Janssen Vaccines & Prevention B.V. (formerly known as Crucell Holland B.V.).

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Rationale: Minor textual changes have been made, in addition to updates to be in line with other current protocols.

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12.1.1 Adverse Event Definitions and Classifications

Rationale: The protocol has been updated to be in line with the current protocol template (version 6 June 2016).

4.1 Inclusion Criteria
17.5 Case Report Form Completion

Amendment_3 (26 January 2016)

This amendment is considered to be substantial based on the criteria set forth in Article 10(a) of Directive 2001/20/EC of the European Parliament and the Council of the European Union.

The overall reason for the amendment: This amendment includes the request of the Center for Biologics Evaluation and Research (CBER, a division of US Food and Drug Administration [FDA]) to extend the safety follow-up to 6 months post-boost. This request was originally made for protocols VAC52150EBL3002 and VAC52150EBL3003 and has now also been implemented for this protocol.

The changes made to the clinical protocol VAC52150EBL2001 are listed below, including the rationale of each change and a list of all applicable sections.

Rationale: As requested by the CBER (US FDA), the 6-month visit has been changed to 6-month post-boost visit. In addition, a statement has been added that subjects who already attended the 6-month post-prime visit (which has been replaced with the 6-month post-boost visit per the current protocol amendment) will be required to also attend the 6-month post-boost visit.

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5 TREATMENT ALLOCATION AND BLINDING
9.1.1 Overview
9.1.4 Post-vaccination Phase
9.1.5 Open-label Long-term Follow-up Phase
11.5 Immunogenicity Analyses

Rationale: As requested by the CBER (US FDA), the safety laboratory assessments at screening are to be performed within 28 days prior to the prime vaccination and may be repeated if they fall outside this time window.

Time and Events Schedule
4.1 Inclusion Criteria
9.1.2 Screening Phase

Rationale: The time of unblinding and database lock for primary analysis has been changed to when all subjects have completed the 6-month post-boost visit or discontinued earlier. Subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit.

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Additional Time and Events Schedule
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9.1.5 Open-label Long-term Follow-up Phase
10.1 Completion
11 STATISTICAL METHODS

Rationale: The statement that a male subject's study vaccine should be permanently discontinued if his partner becomes pregnant, has been removed. The current biodistribution and reprotoxicity data support the recommendation that there is a negligible risk to the partner of a male vaccinated subject if she becomes pregnant.

10.2 Discontinuation of Study

Rationale: The statement that the birth control method used by the female partner of a male subject should be documented, has been removed. The current biodistribution data suggest that there is no vaccine present in tissues other than the inoculated muscle and regional lymph nodes. Therefore, the risk to the partner of a male participant is negligible and it is unnecessary to confirm additional contraceptive use by the partner. Consistent condom use in the male participant will continue to be emphasized.

4.1 Inclusion Criteria

4.3 Prohibitions and Restrictions

Rationale: A statement has been added to allow enrollment of site staff or their family members, provided this is governed by institutional procedures and that there is no direct involvement in the proposed study or relationship with the investigator.

4.2 Exclusion Criteria

Rationale: One case of chest pain that might be indicative of pericarditis observed in the MVA-BN clinical trial program has been added to the Potential Risks section.

1.2.4 Potential Risks

Rationale: The analysis to estimate survival based on immunobridging methodology, linking NHP immune response data with human immune response data, will be described in a separate SAP.

11 STATISTICAL METHODS

Rationale: The analysis of response patterns over time for the immunologic parameters was deleted, since this analysis will not be performed.

SYNOPSIS

11.5 Immunogenicity Analyses

Rationale: Information regarding the marketing authorization of MVA-BN and the Phase 3 clinical study POX-MVA-013 has been updated.

1.1 Background

REFERENCES

Rationale: The VAC52150 Vaccine Development Registry was replaced by a roll-over study. Further details regarding enrollment have been added.

SYNOPSIS

3.1 Overview of Study Design

9.1.6 VAC52150 Vaccine Development Roll-over Study

12.3.4 Pregnancy

Rationale: Immunogenicity objectives, endpoints and assessments have been revised and corrected.

SYNOPSIS

2.1 Objectives

9.3 Immunogenicity Evaluations

Rationale: The protocol has been updated to be in line with the current protocol template (version 14 October 2015).

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9.1.4 Post-vaccination Phase
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17.10 On-site Audits
17.11 Use of Information and Publication

Rationale: Minor textual changes have been made, in addition to modifications for clarity and updates to be in line with other current protocols.

Amendment_2 (18 August 2015)

This amendment is considered to be substantial based on the criteria set forth in Article 10(a) of Directive 2001/20/EC of the European Parliament and the Council of the European Union.

The overall reason for the amendment: This amendment was created to implement site-specific requests and additional changes.

The table below gives an overview of the rationale for each change and all applicable sections.
The following changes were made on site-specific request:

Rationale: In the amendment, a statement was added to clarify that subjects who received placebo and reach the Day 180 visit prior to unblinding will be required to attend the Day 180 visit. After unblinding, only subjects who received active vaccine will enter a long-term follow-up phase.

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Time and Events Schedule
Additional Time and Events Schedule
3.1 Overview of Study Design
5 TREATMENT ALLOCATION AND BLINDING
9.1.5 Open-label Long-term Follow-up Phase

Rationale: In the amendment, a statement was added to clarify that the 42-days post-boost (Cohorts I and II), Day 180 and Day 365 visits (Cohort I) may be replaced by a telephone call if the subject is not able to come to the site.

Time and Events Schedule
9.1.1 Overview
9.1.4 Post-vaccination Phase
9.1.5 Open-label Long-term Follow-up Phase

Rationale: In the amendment, the Day 180 visit window has been extended from 15 to 30 days.

9.1.1 Overview

Rationale: Modifications for clarity throughout the document.

SYNOPSIS
6.1 General Instructions and Procedures

The following additional changes were made:

Rationale: In the amendment, a statement was added to clarify that unblinding and clinical database lock will be done by country.

SYNOPSIS
3.1 Overview of Study Design
5 TREATMENT ALLOCATION AND BLINDING
9.1.5 Open-label Long-term Follow-up Phase

Rationale: Modifications for clarity throughout the document.

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Additional Time and Events Schedule
1.2.4 Potential Risks
1.2.5 Overall Benefit/Risk Assessment
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Amendment_1 (7 May 2015)

This amendment is considered to be substantial based on the criteria set forth in Article 10(a) of Directive 2001/20/EC of the European Parliament and the Council of the European Union.

The overall reason for the amendment: This amendment was created upon request of the Medicines and Healthcare Products Regulatory Agency (MHRA). Further modifications were made to implement a site-specific request, to update the list of references and to correct minor inconsistencies.

The table below gives an overview of the rationale for each change and all applicable sections.

Rationale: In the amendment, '*a severe (grade 3) (non-serious) adverse event considered to be at least possibly related to study vaccine (active vaccine or placebo) that persists for 3 or more days, or a severe (grade 3) (non-serious) laboratory abnormality (including unexplained hematuria) considered to be at least possibly related to study vaccine (active vaccine or placebo)*' has been added as contraindication to boost vaccination as requested by the MHRA.

6.3 Contraindications to Boost Vaccination

Rationale: In the amendment, a statement has been added as requested by the MHRA to clarify that if any pausing rule is met, and following the IDMC review it is deemed appropriate to restart dosing, a request to restart dosing with pertinent data must be submitted to the health authorities as a request for a substantial amendment. Dosing can restart only after approval of a substantial amendment by the competent authority.

9.2.3 Pausing Rules

Rationale: In the original protocol it was stated that '*a subject's study vaccine should be permanently discontinued if (followed by a list of criteria)*'. In the amendment, it is clarified as requested by the MHRA that '*a subject's study vaccine will be permanently discontinued in case any of the following listed criteria is met*'.

10.2 Discontinuation of Study

Rationale: Wording was modified in the amendment following a site-specific request to allow study vaccine administration by unblinded study-site personnel. The definition has been modified accordingly and wording has been added to indicate that the study vaccine administrator will have no other study function.

Definitions of Terms
3.2 Study Design Rationale
5 TREATMENT ALLOCATION AND BLINDING
6.1 General Instructions and Procedures
7 TREATMENT COMPLIANCE
9.1.3 Vaccination Phase
14.3 Preparation, Handling, and Storage

Rationale: The reference list was updated to include the addendum to the Ad26.ZEBOV Investigator's Brochure Edition 2 that was issued in April 2015.

1 INTRODUCTION
1.1 Background
REFERENCES

Rationale: Modifications to correct minor inconsistencies.

1.3 Overall Rationale for the Study
11.1 Analysis Sets

SYNOPSIS

A Randomized, Observer-blind, Placebo-controlled, Phase 2 Study to Evaluate the Safety, Tolerability and Immunogenicity of Three Prime-boost Regimens of the Candidate Prophylactic Vaccines for Ebola Ad26.ZEBOV and MVA-BN-Filo in Healthy Adults in Europe

EudraCT number: 2015-000596-27

The sponsor, in collaboration with Bavarian Nordic (BN) and in conjunction with an Innovative Medicines Initiative (IMI) consortium led by the Institut National de la Santé et de la Recherche Médicale (INSERM), is investigating the potential of a prophylactic Ebola vaccine regimen comprised of the following 2 candidate Ebola vaccines:

Ad26.ZEBOV is a monovalent vaccine expressing the full length Ebola virus (EBOV, formerly known as *Zaire ebolavirus*) Mayinga glycoprotein (GP), and is produced in the human cell line PER.C6®.

MVA-mBN226B, further referred to as Modified Vaccinia Ankara (MVA)-BN-Filo®, is a multivalent vaccine expressing the Sudan virus (SUDV) GP, the EBOV GP, the Marburg virus (MARV) Musoke GP, and the Tai Forest virus (TAFV, formerly known as *Côte d'Ivoire ebolavirus*) nucleoprotein (NP), and is produced in chicken embryo fibroblast cells. The EBOV GP expressed by MVA-BN-Filo has 100% homology to the one expressed by Ad26.ZEBOV.

In this Phase 2 study, the sponsor's adenovirus serotype 26 (Ad26) vector expressing the EBOV Mayinga GP (Ad26.ZEBOV) and the MVA-BN vector with EBOV, SUDV and MARV GP inserts and TAFV NP insert (MVA-BN-Filo) will be evaluated as a heterologous prime-boost regimen, in which one study vaccine (Ad26.ZEBOV) is used to prime a filovirus-specific immune response and the other study vaccine (MVA-BN-Filo) is used to boost the immune response 28, 56 or 84 days later. The EBOV GP that is currently circulating in West Africa has 97% homology to the EBOV GP used in this vaccine regimen.

OBJECTIVES AND HYPOTHESIS

Primary Objective

The primary objective is to assess the safety and tolerability of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered intramuscularly (IM) as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.

Secondary Objective

The secondary objective is to assess humoral immune responses, as measured by ELISA, to the EBOV GP 21 days post boost of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.

Exploratory Objectives

The exploratory objectives are:

- To assess humoral immune responses, as measured by ELISA, to the EBOV GP at other relevant time points of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.
- To assess the neutralizing capacity of the EBOV GP-specific humoral immune response, as measured by virus neutralization assay, at selected time points of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.
- To further explore humoral immune responses at selected time points to different EBOV GPs, filovirus GPs and/or TAFV NP as well as the adenovirus and/or MVA backbones of the various vaccination schedules tested, if assays are available.
- To explore cellular immune responses at selected time points to different EBOV GPs, filovirus GPs and/or TAFV NP of the various vaccination schedules tested, if assays are available.

HYPOTHESIS

As this study is designed to provide descriptive information regarding safety and immunogenicity without formal treatment comparisons, no formal statistical hypothesis testing is planned.

OVERVIEW OF STUDY DESIGN

This study is a randomized, observer-blind, placebo-controlled, parallel-group, multicenter, Phase 2 study to evaluate the safety, tolerability and immunogenicity of 3 heterologous prime-boost regimens using Ad26.ZEBOV at a dose of $5x10^{10}$ viral particles (vp) as prime and MVA-BN-Filo at a dose of $1x10^{8}$ infectious units (Inf.U, nominal titer) as boost at a 28-, 56- or 84-day interval in healthy adult subjects in Europe (United Kingdom [UK] and France). The 3 prime-boost regimens will only differ in the timing of the boost vaccination (ie, 28, 56 or 84 days after prime, respectively referred to as Groups 1, 2 and 3), while the dose of each study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo) and the sequence of vaccination will be identical.

The subject population will consist of healthy men and women aged between 18 and 65 years (inclusive), who never received a candidate Ebola vaccine before and have no prior exposure to Ebola virus (including travel to West Africa less than 1 month prior to screening) or a diagnosis of Ebola virus disease.

At study entry, subjects were offered the option to enroll into Cohorts I, II, or III in the UK or into Cohorts II or III in France. In Cohorts II and III in both countries, core immunogenicity assessments (humoral and cellular assays) will be performed. In Cohort II in both countries, additional immunogenicity assessments will be done. In Cohort I, plasmablast response kinetics will be evaluated for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK. In the UK, Cohorts I and III may start in parallel, while Cohort II in the UK can only start when the peak of B-cell response after prime vaccination in Cohort I is identified. In France, Cohorts II and III may start in parallel (as there will be no Cohort I) and are independent of Cohort I in the UK.

Study Cohorts	Randomization Ratio (Active:Placebo)	Group 1 N=204	Group 2 N=204	Group 3* N=204	Cohort Total N=612	UK** N=321	France** N=291
Cohort I	-	10/0	10/0	10/0	**30**	30	-
Cohort II	14:1	84/6	84/6	84/6	**270**	135	135
Cohort III	10:3	80/24	80/24	80/24	**312**	156	156

Groups 1, 2, and 3: prime on Day 1, followed by boost 28, 56 or 84 days after prime, respectively; N: number of subjects to receive study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo)
* Randomization to Group 3 was stopped per Amendment 4 to focus on the schedules for which an indication will be sought.
** Enrollment in the entire study (ie, UK and France) will be stopped per Amendment 5.

Subjects in each cohort were randomized at baseline (on Day 1) in a 1:1:1 ratio to Groups 1, 2, and 3 (ie, the vaccination schedule). Within each group in Cohort I, subjects will receive Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion. Randomization in Cohorts II and III was stratified by country. Within each group in Cohorts II and III, subjects were randomized to receive the prime-boost vaccination with either Ad26.ZEBOV followed by MVA-BN-Filo, or placebo in a 14:1 and 10:3 ratio, respectively. Randomization in each group (for Cohorts II and III) was stratified further according to age at randomization (≤50 years, >50 years), with at least 20% (of the total of both cohorts) of subjects in the >50 years category. Note that enrollment will not be resumed.

Study-site personnel (except for those with primary responsibility for study vaccine preparation and dispensing), sponsor personnel and subjects in Cohorts II and III will be blinded to the study vaccine allocation until all subjects have completed the 6-month post-boost visit or discontinued earlier and the clinical database is locked. In case interim analyses will be performed, study-site personnel (except for those with primary responsibility for study vaccine preparation and dispensing), the sponsor (except for programming, statistics, clinical and clinical immunology personnel involved in the analysis, and the sponsor committee involved in making future decisions for the program) and subjects will remain blinded to study vaccine allocation until the primary analysis.

All subjects will receive the study vaccine through IM injection in the deltoid, either Ad26.ZEBOV ($5x10^{10}$ vp) on Day 1, followed by a boost vaccination of MVA-BN-Filo ($1x10^{8}$ Inf.U, nominal titer) on Days 29, 57 or 85; or placebo (0.9% saline) on Day 1, followed by a boost vaccination of placebo (0.9% saline) on Days 29, 57 or 85.

The study consists of a screening phase of up to 12 weeks (starting from the moment the subject signs the informed consent form [ICF]), a vaccination phase in which subjects will be vaccinated at baseline (Day 1), followed by a boost vaccination on Days 29, 57 or 85, and a post-vaccination phase and long-term follow-up phase until Day 365 (or until the start of the roll-over study or for an additional 12 months [whichever comes first] for subjects in France who agree to continue the long-term follow-up after Day 365, see details below). After unblinding, the subjects who received placebo in Cohorts II and III will be contacted by the site to communicate that they have completed the study and no further follow-up is required. Unblinding will be done when the last subject in the study completes the 6-month post-boost visit or discontinues earlier and the clinical database is locked. However, subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit. After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will continue the study until the Day 365 (or until the start of the roll-over study or for an additional 12 months [whichever comes first] for subjects in France who agree to continue the long-term follow-up after Day 365, see details below) visit to assess long-term safety and immunogenicity.

An Independent Data Monitoring Committee (IDMC) will be commissioned for this study.

The sponsor halted all vaccinations in this study due to the occurrence of a serious and very rare condition, Miller Fisher syndrome, reported in the current study VAC52150EBL2001, until a revised ICF containing updated safety language was prepared and approval to restart the study was granted by the relevant competent authority. The study pause interrupted dosing of subjects, some awaiting prime vaccination and some awaiting boost vaccination. When approval was granted in the UK to restart the

study under Amendment 4, the sponsor offered a late boost vaccination to those subjects who did not receive it yet, unless participants had withdrawn from the trial or were not eligible to receive the boost. Vaccinated subjects have been following the same post-boost vaccination schedule as those subjects unaffected by the pause. Subjects who did not receive the late boost were encouraged to return for the 1-year post-prime visit. After restart of the study in the UK under Amendment 4, screening for Groups 1 and 2 restarted, but randomization to Group 3 was stopped to focus on the schedules for which an indication will be sought. After approval of Amendment 5, enrollment in the entire study will be stopped.

Subjects in the UK and France who reach the Day 365 visit prior to unblinding will be approached to consent for enrollment into the VAC52150 Vaccine Development Roll-over study (VAC52150EBL4001) for long-term surveillance (for a total of up to 60 months after the prime vaccination). After unblinding, only subjects who received Ad26.ZEBOV and/or MVA-BN-Filo will remain in the VAC52150 Vaccine Development Roll-over study for long-term surveillance. After unblinding, subjects who received placebo and have already been enrolled into the VAC52150 Vaccine Development Roll-over study will be discontinued from further participation in the roll-over study. The parent(s)/legal guardian of children born to vaccinated female subjects who became pregnant with estimated conception within 28 days after vaccination with MVA-BN-Filo or within 3 months after vaccination with Ad26.ZEBOV, will also be approached to consent for enrollment of their offspring into the roll-over study, according to the same rules that apply for the other subjects. Subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study is opened, will have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months (whichever comes first) or discontinued earlier. These subjects will be contacted by telephone every 3 months after Day 365 (subjects will only visit the site when clinically indicated) for the collection of information on adverse events, serious adverse events, and immediate reportable events (IREs) that were ongoing at the Day 365 visit, and to report and follow up on new serious adverse events, pregnancies, and IREs.

SUBJECT POPULATION

Screening of subjects for eligibility was performed within 12 weeks before administration of the study vaccine on Day 1. The subject population will consist of healthy (on the basis of medical history, physical examination, electrocardiogram (ECG), vital signs, clinical laboratory testing, and clinical judgment) men and women aged between 18 and 65 years (inclusive), who have no prior exposure to Ebola virus (including travel to West Africa less than 1 month prior to screening) or a diagnosis of Ebola virus disease. Subjects who received a candidate Ebola vaccine or any experimental candidate Ad26- or MVA-based vaccine in the past or with known allergy or history of anaphylaxis or other serious adverse reactions to vaccines or vaccine products, including known allergy to egg, egg products and aminoglycosides will be excluded.

DOSAGE AND ADMINISTRATION

Study vaccines (Ad26.ZEBOV, MVA-BN-Filo or placebo) will be administered as 0.5-mL IM injections into the deltoid muscle. The boost vaccination should be administered in the opposite deltoid from the prime vaccination.

All subjects will receive a vaccination, according to randomization, on Day 1 (Groups 1 to 3) and on Day 29 (Group 1), Day 57 (Group 2), or Day 85 (Group 3), at the following dose levels:

- Ad26.ZEBOV: $5x10^{10}$ vp, supplied in a single use vial (0.5 mL extractable);
- MVA-BN-Filo: $1x10^{8}$ Inf.U (nominal titer); target fill $1.9x10^{8}$ Inf.U per dose (range 1.27 to 2.67 $x10^{8}$ Inf.U), supplied in a single use vial (0.5 mL extractable);
- Placebo: 0.9% saline, 0.5 mL extractable.

After each vaccination, subjects will remain at the site for a total of 60 (±15) minutes post-vaccination to monitor for the development of any acute reactions, or longer if deemed necessary by the investigator (eg, in case of grade 3 adverse events).

Criteria for postponement of vaccination at the scheduled time for vaccine administration and contraindications to boost vaccination have been defined and will be applied by the investigator.

SAFETY EVALUATIONS

All safety evaluations will be performed as specified in the Time and Events Schedule. Safety will be evaluated in all subjects from the 3 cohorts.

Safety will be assessed by collection of solicited local and systemic adverse events (reactogenicity), unsolicited adverse events, IREs, and serious adverse events. The subjects will be closely observed by study-site personnel for the first 30 (±10) minutes after each vaccination and again at 60 (±15) minutes post-vaccination, and any unsolicited, solicited local or systemic adverse events will be documented during this period. Upon discharge from the site, subjects will receive a diary, a thermometer, and a ruler to measure solicited local reactions and body temperature. Subjects will record symptoms of solicited local and systemic adverse events in the diary in the evening after each vaccination and then daily for the next 7 days. The investigator will document unsolicited adverse events from signing of the ICF onwards until 42 days post-boost, and serious adverse events and IREs from signing of the ICF onwards until the end of the study. The primary endpoints are adverse events, serious adverse events, IREs, and solicited local and systemic adverse events. Adverse events that are ongoing at 42 days post-boost vaccination will be followed until resolution or stabilization. For subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study is opened, the investigator will document adverse events, serious adverse events, and IREs that were ongoing at the Day 365 visit, and new serious adverse events, pregnancies, and IREs from Day 365 onwards until the start of the roll-over study or for an additional 12 months (whichever comes first) (or discontinued earlier).

Other safety assessments include clinical laboratory testing, ECGs, vital signs (blood pressure, pulse/heart rate, and body temperature), physical examination and pregnancy testing at the time points indicated in the Time and Events Schedule.

The investigators, together with the sponsor's medical monitor, will be responsible for the safety monitoring of the study, and will halt vaccination of further subjects in case any of the pre-specified pausing rules have been met.

IMMUNOGENICITY EVALUATIONS

All immunogenicity evaluations will be performed as specified in the Time and Events Schedule and Additional Time and Events Schedule.

Blood samples for core immunogenicity assessments (humoral and cellular assays) for the evaluation of secondary and exploratory endpoints as planned by the sponsor will be collected at the time points indicated in the Time and Events Schedule. Blood samples for assessment of humoral immune responses will be obtained from all subjects in Cohorts II and III. Blood samples for assessment of cellular immune responses will be obtained from subjects in Cohorts II and III at selected sites with the capabilities to process peripheral blood mononuclear cells (PBMC) (targeted at 10% of all subjects).

Blood samples for additional immunogenicity assessments for the evaluation of additional exploratory endpoints as planned by the academic consortium partners will be collected from subjects in Cohort II at the time points indicated in the Additional Time and Events Schedule. Blood samples for plasmablast response kinetics will be collected in Cohort I at the time points indicated in the Additional Time and Events Schedule for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK.

A blood sample for deoxyribonucleic acid (DNA) collection will be collected at baseline from subjects in Cohort II at selected sites (as part of the additional immunogenicity assessments by the academic consortium partners) and who consent separately to this component of the study. Subject participation in genomic research is optional. The goal of this genomic research is to allow the identification of genetic factors that may influence the immunogenicity, safety, or tolerability of Ad26.ZEBOV and MVA-BN-Filo. Since the sample size is small, analysis will focus on validation of genes identified as possibly associated with these factors in previous (non-Ebola) studies and control of gene expression.

Optional blood samples for transcriptomics will be collected from subjects in Cohorts I and II (as part of the specific and additional immunogenicity assessments by the academic consortium partners, respectively) and who consent separately to the transcriptomics component of the study. Blood samples will be collected at the specified time points (see Additional Time and Events Schedule). The goal is to assess the ribonucleic acid (RNA) transcriptome (gene expression) in a systems vaccinology approach to identify differentially expressed genes which relate to the immunologic and safety phenotype of the response to Ad26.ZEBOV and MVA-BN-Filo, and for analysis of antibody genes.

If subjects provide separate consent, leftover blood samples will be stored for possible future scientific/genetic research. Subjects can withdraw consent for their samples to be used for future research at any time.

STATISTICAL METHODS

An originally planned sample size of 612 subjects (pooled across the 3 cohorts) includes 492 subjects randomized to receive Ad26.ZEBOV and MVA-BN-Filo (in Cohorts II and III) to substantially contribute to an overall safety database of the Ad26.ZEBOV and MVA-BN-Filo prime-boost regimen. In Cohort I, 30 subjects will receive Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion. Recruitment in Group 3 was stopped per Amendment 4 to focus on the schedules for which an indication will be sought. Enrollment in the entire study will be stopped per Amendment 5. Details on the impact on the power will be described in the Statistical Analysis Plan (SAP) and/or Clinical Study Report. In case a specific adverse event is not observed, the one-sided 97.5% upper confidence limit of the true incidence rate of this adverse event is less than 3.3%, 2.2% and 0.8% for sample sizes of 121, 174 and 499 subjects (ie, total number of subjects in Cohorts I, II, and III to receive Ad26.ZEBOV and/or MVA-BN-Filo), respectively.

Interim analyses may be performed during the study for regulatory purposes or for the purpose of informing future vaccine-related decisions in a timely manner. The primary analysis will be performed when all subjects have completed the 6-month post-boost visit or discontinued earlier and the clinical database is locked. This analysis will include 6-month post-boost safety and all available immunogenicity data up to this point. The final analysis will be performed when all subjects have completed the last study-related visit or discontinued earlier. Specific details will be provided in the SAP.

An IDMC will be established to monitor data on a regular basis to ensure the continuing safety of the subjects enrolled in the study.

Safety Analyses

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively (including 95% confidence intervals, if applicable), by cohort and overall. For each adverse event, the number and percentage of subjects who experience at least 1 occurrence of the given event will be summarized by group. Summaries, listings, datasets, or subject narratives may be provided, as appropriate, for those subjects who die, who discontinue study vaccine due to an adverse event, or who experience a severe or a serious adverse event. The most severe laboratory abnormalities following vaccination will be tabulated. Abnormalities in vital signs, ECG parameters, and physical examination following vaccination will be tabulated by most severe abnormality grade.

Immunogenicity Analyses

Descriptive statistics (actual values and changes from baseline, including 95% confidence intervals, if applicable) will be calculated for continuous immunologic parameters at each time point analyzed. Graphical representations of changes in immunologic parameters will be prepared, as applicable. Frequency tabulations will be calculated for discrete (qualitative) immunologic parameters at each time point analyzed. In addition, differences between the vaccination schedules will be evaluated at the 21-day post-boost, 6-month post-boost and Day 365 visits.

TIME AND EVENTS SCHEDULE

Cohorts I, II and III	**Screening Phase[a] (≤12 weeks)**	**Vaccination and Post-vaccination Phase[b,c]**									**Long-term Follow-up Phase[c,d]**	
Group 1		**Day 1**	**Day 4**	**Day 8**	**-**	**Day 29**	**Day 32**	**Day 36**	**Day 50**	**Day 71[b]**	**Day 209**	**Day 365[e]**
Group 2					**-**	**Day 57**	**Day 60**	**Day 64**	**Day 78**	**Day 99[b]**	**Day 237**	
Group 3					**Day 15[f]**	**Day 85**	**Day 88**	**Day 92**	**Day 106**	**Day 127[b]**	**Day 265**	
Study Procedures		**Prime**	+3d pp	+7d pp	+14d pp	**Boost**	+3d pb	+7d pb	+21d pb	+42d pb	+6m pb[g]	+1y pp
Screening/Administrative												
Informed consent[h]	X											
Inclusion/exclusion criteria	X[i]											
Medical history and demographics	X											
Prestudy therapies[j]	X											
Serum pregnancy test[k]	X											
Serology (HIV-1/2, hepatitis B/C)	X											
Follicle-stimulating hormone (FSH)[l]	X											
Check clinical status + available data		X[m]										
Randomization		X										
Study vaccine administration[n]		▲				▼						
Safety Assessments												
Urine pregnancy test[k]		X[o]				X[o]						
Physical examination[p]	X[p]	X[o]	X	X	X[f]	X[o]	X	X	X	X	X	X
Electrocardiogram[q] (ECG)	X					X[o]		X				
Vital signs[r]	X	X[o]				X[o]						
Distribution of subject diary[s]		X				X						
Review of subject diary by site staff			X	X			X	X				
Adverse events[t]	Continuous											
Serious adverse events and immediate reportable events[u]	Continuous											
Concomitant medications	X	X	X	X	X[f]	X	X	X	X	X	X[v]	X[v]
Clinical Laboratory Assessments												
Hematology, chemistry	X	X[o]		X		X[o]		X				
Troponin I	X											
Urinalysis	X											
Core Immunogenicity Assessments (Cohorts II and III)[w]												
Blood sampling for humoral assays		X[o]			X[f]	X[o,x]		X[x]	X[x]		X[x]	X[x]
Blood sampling for cellular assays		X[o]			X[f]	X[o,x]		X[x]	X[x]		X[x]	X[x]

Additional (Cohort II) & Specific (Cohort I) Immunogenicity Assessments[y]	See Additional Time and Events Schedule

d pp: days post-prime; d pb: days post-boost; m pb: months post-boost; y pp: year post-prime;
▲ *Ad26.ZEBOV 5x10^{10} vp or placebo* ▼ *MVA-BN-Filo 1x10^{8} Inf.U (nominal titer) or placebo.*

NOTE: *If a subject withdraws early from the study, early withdrawal assessments should be obtained per the assessments for the 21-day post-boost visit. A subject who wishes to withdraw consent from participation in the study will be offered an optional visit for safety follow-up (before formal withdrawal of consent), but the subject has the right to refuse.*

[a] Screening may be split into multiple days or visits. Retesting of values (eg, safety laboratory) that lead to exclusion is allowed once using an unscheduled visit during the screening phase, also for subjects who are rescreened due to a pause. The safety laboratory assessments at screening are to be performed within 28 days prior to the prime vaccination and may be repeated if they fall outside this time window. Similarly, subjects who are rescreened due to a pause must have new safety laboratory assessments within 28 days of the prime vaccination.

[b] In addition to the assessments scheduled for the 42-day post-boost visit, subjects will be instructed to contact the investigator before the next visit if they experience any adverse event or intercurrent illness that they perceive as relevant and/or can be possibly related to study vaccine in their opinion. After unblinding, subjects who received placebo will be contacted by the site to communicate that they have completed the study and no further follow-up is required. Unblinding will be done when the last subject in the study completes the 6-month-post-boost visit or discontinues earlier and the clinical database is locked. However, subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit. The 42-days post-boost visit may be replaced by a telephone call if the subject is not able to come to the site (only applies to Cohorts I and II).

[c] For subjects who receive the boost vaccination outside the protocol-defined boost vaccination window due to pausing of the study, the timings of the next visits post boost vaccination will be determined relative to the actual day of boost vaccination. Subjects who did not receive the late boost vaccination were encouraged to return for the 1-year post-prime visit.

[d] After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will be followed up for the collection of serious adverse event information, immediate reportable events and for blood draws for immunogenicity assessments after the 6-month post-boost visit. The 6-month post-boost and Day 365 visits may be replaced by a telephone call if the subject is not able to come to the site (only applies to Cohort I).

[e] Subjects in France who agree to continue the long-term follow-up after Day 365 and who reach the Day 365 visit before the roll-over study VAC52150EBL4001 is opened, will have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months (whichever comes first) or discontinued earlier. These subjects will be contacted by telephone every 3 months after Day 365 (subjects will only visit the site when clinically indicated) for the collection of information on adverse events, serious adverse events, and immediate reportable events that were ongoing at the Day 365 visit, and to report and follow up on new serious adverse events, pregnancies, and immediate reportable events. Concomitant therapies are only to be recorded if given in conjunction with serious adverse events and immediate reportable events.

[f] Day 15 visit only for subjects in Group 3 of Cohort III.

[g] Subjects who already attended the 6-month post-prime visit (which has been replaced with the 6-month post-boost visit per protocol amendment 3) will be required to also attend the 6-month post-boost visit.

[h] Signing of the informed consent form (ICF) needs to be done before the first study-related activity.

[i] The investigators should ensure that all study enrollment criteria have been met at the end of the screening phase. Minimum criteria for the availability of documentation supporting the eligibility criteria are described in Section 17.4.

[j] Prestudy therapies up to 30 days prior to the start of screening and previous vaccinia/smallpox vaccination at any time prior to study entry must be recorded in the case report form (CRF).

[k] For women of childbearing potential.

[l] For women >45 years of age with amenorrhea for less than 2 years or ≤45 years of age with amenorrhea for more than 6 months.

[m] If a subject's clinical status changes (including available laboratory results or receipt of additional medical records) after screening so the subject no longer meets eligibility criteria, the subject should be excluded from further participation in the study.

[n] After each vaccination, subjects will remain at the site for a total of 60 (±15) minutes post-vaccination to monitor for the development of any acute reactions, or longer if deemed necessary by the investigator. Solicited and unsolicited adverse events emerging during the observation period at the site will be recorded in the CRF.

[o] Prior to study vaccine administration.

[p] A full physical examination, including height and body weight, will be carried out at screening. At other visits, an abbreviated, symptom-directed examination will be performed as indicated by the investigator.

[q] A single, 12-lead ECG (supine) after at least 5 minutes rest will be performed and interpreted locally. Additional ECG monitoring may be done at other time points during the study if clinically indicated based on signs and symptoms. If blood sampling or vital sign measurement is scheduled for the same time point as ECG recording, the procedures should be performed in the following order: vital signs, ECG(s), blood draw.

[r] Includes blood pressure, pulse/heart rate (at rest) and body temperature to be assessed prior to study vaccine administration.

[s] Subjects will use the subject diary to document solicited local and systemic adverse events (reactogenicity) in the evening after each vaccination and then daily for the next 7 days.

[t] Pregnancies will be reported from signing of the ICF until the end of the study.

[u] For reporting of immediate reportable events, refer to Section 12.3.3.

[v] Concomitant therapies must be recorded from screening onwards until 42 days after the boost vaccination. Thereafter, concomitant therapies are only to be recorded if given in conjunction with serious adverse events and immediate reportable events.

[w] Only including the core immunogenicity assessments. Blood samples will be collected for humoral assays from all subjects in Cohorts II and III and for cellular assays from subjects in Cohorts II and III at selected sites with the capabilities to process PBMC (targeted at 10% of all subjects).

[x] Subjects who receive the boost vaccination outside the protocol-defined boost vaccination window due to pausing of the study, will only have blood samples taken to assess humoral immune responses measured at selected time points by ELISA. In addition, blood samples will be taken prior to the boost vaccination and at the 7-day post-boost visit for safety assessments.

[y] For potential additional immunogenicity assessments in Cohort II and specific immunogenicity assessments in Cohort I, see Additional Time and Events Schedule.

ADDITIONAL TIME AND EVENTS SCHEDULE

Cohort I	**Vaccination and Post-vaccination Phase[a,b]**									
Group 1	**Day 1**	**Day 9**	**Day 11**	**Day 13**	**Day 15**	**Day 29**	**Day 32**	**Day 34**	**Day 36**	**Day 38**
Group 2						**Day 57**	**Day 60**	**Day 62**	**Day 64**	**Day 66**
Group 3						**Day 85**	**Day 88**	**Day 90**	**Day 92**	**Day 94**
	Prime	+8d pp	+10d pp	+12d pp	+14d pp	**Boost[l]**	+3d pb[l]	+5d pb[l]	+7d pb[l]	+9d pb[l]
Study vaccine administration	▲					▼				
Specific Immunogenicity Assessments by University of Oxford – Blood sampling in UK for:										
Determination of kinetics of B-cell response (B-cell ELISpot)	X[c]	X	X	X	X	X[c]	X	X	X	X
Transcriptomics[d]	X[c]	X	X	X	X	X[c]	X	X	X	X

Cohort II[e]	**Vaccination and Post-vaccination Phase[a,b]**											**LT FU Phase[b,f]**
Group 1	**Day 1**	**Day 2**	**Day 8**	**Day 15**	**TBD[g]**	**Day 29**	**Day 30**	**Day 36**	**Day 43**	**TBD[g]**	**Day 50**	**Day 209**
Group 2						**Day 57**	**Day 58**	**Day 64**	**Day 71**		**Day 78**	**Day 237**
Group 3						**Day 85**	**Day 86**	**Day 92**	**Day 99**		**Day 106**	**Day 265**
	Prime	+1d pp	+7d pp	+14d pp	tbd	**Boost[l]**	+1d pb[l]	+7d pb[l]	+14d pb[l]	tbd[l]	+21d pb[l]	+6m pb[h,l]
Study vaccine administration	▲					▼						
Additional Immunogenicity Assessments by University of Oxford – Blood sampling in UK for:												
Repertoire sequencing, isolation of monoclonal Ab, plasmablast phenotyping	X[c]				X[g]	X[c]				X[g]		
Memory B-cell characterization	X[c]					X[c]					X	X
DNA isolation[i]	X[c]											
Fresh intracellular cytokine staining (ICS)	X[c]					X[c]					X	
Transcriptomics[d]	X[c]				X[g]	X[c]				X[g]		
Additional Immunogenicity Assessments by INSERM – Blood sampling in France for:												
Natural killer (NK) cell analysis[j]	X[c]					X[c]			X			X
Transcriptomics[d]	X[k]	X	X			X[k]	X	X				
Fresh ICS	X[c]					X[c]					X	
T cell proliferation	X[c]			X		X[c]			X			X
T cell phenotyping (including TfH)	X[c]		X			X[c]		X				
Cytokine profiling	X[c]	X	X			X[c]	X	X				
EBOV neutralization	X[c]					X[c]					X	X

Ab: antibodies; d pp: days post-prime; d pb: days post-boost; LT FU: long-term follow-up; m pb: months post-boost; TBD: to be determined; TfH: T follicular helper;
▲ *Ad26.ZEBOV 5x10^{10} vp or placebo* ▼ *MVA-BN-Filo 1x10^{8} Inf.U (nominal titer) or placebo.*

NOTE: *In Cohort I, plasmablast response kinetics will be evaluated for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK.*

[a] After unblinding, subjects who received placebo will be contacted by the site to communicate that they have completed the study and no further follow-up is required. Unblinding will be done when the last subject in the study completes the 6-month-post-boost visit or discontinues earlier and the clinical database is locked. However, subjects who received placebo and reach the Day 365visit prior to unblinding will be required to attend the Day 365 visit.

[b] For subjects who receive the boost vaccination outside the protocol-defined boost vaccination window due to pausing of the study, the timings of the next visits post boost vaccination will be determined relative to the actual day of boost vaccination.

[c] Prior to study vaccine administration.

[d] Optional blood sample for transcriptomics only to be obtained from subjects who consent separately.

[e] Blood samples will be collected for DNA isolation (in the UK), transcriptomics (in both countries), and EBOV neutralization (in France) from all subjects in Cohort II. For the other assays, blood samples will be collected from all subjects in Cohort II in the UK, but initially only 25 subjects per group will be targeted for analysis, while in France, blood samples will be collected from and analyzed in no more than 25 subjects in Groups 1 and 2 in Cohort II at selected sites. In addition, these 'other' assays have to be performed within the same 25 subjects per group in the UK and in France.

[f] After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will be followed up for the collection of serious adverse event information, immediate reportable events, and for blood draws for immunogenicity assessments after the 6-month post-boost visit.

[g] At plasmablast peak time point determined in Cohort I (only applies to Cohort II in the UK).

[h] Subjects who already attended the 6-month post-prime visit (which has been replaced with the 6-month post-boost visit per protocol amendment 3) will be required to also attend the 6-month post-boost visit.

[i] Optional blood sample for DNA collection only to be obtained from subjects who consent separately. The DNA sample should be collected at the specified time point; however, if necessary it may be collected at a later time point without constituting a protocol deviation.

[j] Blood sampling done in France, but analysis performed at London School of Hygiene and Tropical Medicine.

[k] Prior to study vaccine administration and at 3-4 hours after study vaccine administration.

[l] Subjects who receive the boost vaccination outside the protocol-defined boost vaccination window due to pausing of the study, will not have specific or additional immunogenicity assessments at these visits.

ABBREVIATIONS

Ad26	adenovirus serotype 26 (vector)
Ad26.ZEBOV	adenovirus serotype 26 expressing the Ebola virus Mayinga glycoprotein
Ad35	adenovirus serotype 35 (vector)
ALT	alanine aminotransferase
AST	aspartate aminotransferase
β-hCG	β-human chorionic gonadotropin
BN	Bavarian Nordic
BUN	blood urea nitrogen
CRF	case report form
DNA	deoxyribonucleic acid
EBOV	Ebola virus
ECG	electrocardiogram
eDC	electronic data capture
EDTA	ethylenediaminetetraacetic acid
ELISA	enzyme-linked immunosorbent assay
ELISpot	enzyme-linked immunospot assay
EU	European Union
FSH	follicle stimulating hormone
GCP	Good Clinical Practice
GMP	Good Manufacturing Practice
GP	glycoprotein
HBsAg	hepatitis B surface antigen
HCV	hepatitis C virus
HIV	human immunodeficiency virus
ICF	informed consent form
ICH	International Council for Harmonisation
ICS	intracellular cytokine staining
IDMC	Independent Data Monitoring Committee
IEC	Independent Ethics Committee
IFN	interferon
Ig	immunoglobulin
IL	interleukin
IM	intramuscular(ly)
IMI	Innovative Medicines Initiative
Inf.U	infectious units
INSERM	Institut National de la Santé et de la Recherche Médicale
IRB	Institutional Review Board
IRE	Immediate Reportable Event
IUD	intrauterine device
IUS	intrauterine system
IWRS	interactive web response system
kb	kilobase
MARV	Marburg virus
MedDRA	Medical Dictionary for Regulatory Activities
MVA	Modified Vaccinia Ankara
MVA-BN-Filo	Modified Vaccinia Ankara Bavarian Nordic vector expressing multiple filovirus proteins
NHP	nonhuman primates
NK	natural killer (cell)
NP	nucleoprotein
NSAID	nonsteroidal anti-inflammatory drug
PBMC	peripheral blood mononuclear cell(s)
PQC	product quality complaint
QTcB	QT interval corrected according to the Bazett formula
QTcF	QT interval corrected according to the Fridericia formula
RBC	red blood cell
RNA	ribonucleic acid

SAP	Statistical Analysis Plan
SUDV	Sudan virus
SUSAR	suspected unexpected serious adverse reaction
TAFV	Tai Forest virus
$TCID_{50}$	50% tissue culture infective dose
TfH	T follicular helper
THAM	tris (hydroxymethyl)-amino methane
TNF	tumor necrosis factor
TOPS	The Over-volunteering Prevention System
UK	United Kingdom
US	United States
VISP	vaccine-induced seropositivity
vp	viral particles
WBC	white blood cell
WHO	World Health Organization

DEFINITIONS OF TERMS

Study vaccine administrator	A trained study nurse, medical doctor, or otherwise qualified health care provider who will have no other study function.
Independent study vaccine monitor	An unblinded study vaccine monitor assigned to the study who is responsible for the unblinded interface between the sponsor and the investigational site pharmacy.
Solicited adverse events (reactogenicity)	Local and systemic adverse events that are common and known to occur after vaccination and that are usually collected in a standard, systematic format in vaccine clinical studies. For the list of solicited adverse events in this study, see Section 9.2.2. For the purpose of vaccine clinical studies, all other adverse events are considered unsolicited; however, this definition should be distinguished from definitions based on pharmacovigilance guidelines.
Study vaccine	MVA-BN-Filo, Ad26.ZEBOV or placebo

1. INTRODUCTION

Janssen Vaccines & Prevention B.V. (formerly known as Crucell Holland B.V.) (hereafter referred to as the sponsor), in collaboration with Bavarian Nordic (BN), and in conjunction with an Innovative Medicines Initiative (IMI) consortium led by the Institut National de la Santé et de la Recherche Médicale (INSERM), is investigating the potential of a prophylactic Ebola vaccine regimen comprised of the following 2 candidate Ebola vaccines:

Ad26.ZEBOV is a monovalent vaccine expressing the full length Ebola virus (EBOV, formerly known as *Zaire ebolavirus*) Mayinga glycoprotein (GP), and is produced in the human cell line PER.C6®.

MVA-mBN226B, further referred to as Modified Vaccinia Ankara (MVA)-BN®-Filo, is a multivalent vaccine expressing the Sudan virus (SUDV) GP, the EBOV GP, the Marburg virus (MARV) Musoke GP, and the Tai Forest virus (TAFV, formerly known as *Côte d'Ivoire ebolavirus*) nucleoprotein (NP), and is produced in chicken embryo fibroblast cells. The EBOV GP expressed by MVA-BN-Filo has 100% homology to the one expressed by Ad26.ZEBOV.

For the most up-to-date nonclinical and clinical information regarding Ad26.ZEBOV and MVA-BN-Filo, refer to the latest versions of the Investigator's Brochures and Addenda (if applicable).[12,13,14] A brief summary of the nonclinical and clinical information is provided below.

The term "sponsor" used throughout this document refers to the entities listed in the Contact Information page(s), which will be provided as a separate document.

1.1. Background

Ebola viruses belong to the Filoviridae family and cause Ebola virus disease, which can induce severe hemorrhagic fever in humans and nonhuman primates (NHPs). Case fatality rates in Ebola disease range from 25% to 90% (average: 50%), according to the World Health Organization (WHO).[26] These viruses are highly prioritized by the United States (US) Government, who has defined them as 'Category A' agents, due to the high mortality rate of infected individuals. Currently, no licensed vaccine, treatment or cure exists for this disease.

Filoviruses are named for their long, filamentous shape. Within this filamentous virus, a single 19-kilobase (kb) negative-sense ribonucleic acid (RNA) genome encodes 7 proteins: the GP, the polymerase, the NP, the secondary matrix protein, the transcriptional activator, the polymerase cofactor, and the matrix protein. The virion surface is covered by homotrimers of the viral GP, which is believed to be the sole host attachment factor for filoviruses. Following cell entry, the viruses replicate their genomes and viral proteins in the cytoplasm using an RNA-dependent RNA polymerase, which is carried into the cell together with the virus.[9]

1.1.1. Nonclinical Studies

Immunogenicity and Efficacy

Immunogenicity and efficacy of the vaccine combination Ad26.ZEBOV and MVA-BN-Filo was evaluated in an NHP model (ie, Cynomolgus macaques, *Macaca fascicularis*). The combination

was assessed in a multivalent filovirus setting in a small number (2 per regimen) of animals and the study included heterologous prime-boost regimens of adenovirus serotype 26 (Ad26), Ad35 and MVA-BN-Filo vectors expressing different Ebola and Marburg proteins. Full protection from Ebola virus disease and death after wild-type EBOV Kikwit 1995 challenge was obtained with all heterologous regimens, including the Ad26 and MVA vaccine regimen. All heterologous prime-boost regimens induced comparable immune responses against the EBOV Mayinga GP. Independently of the vaccine regimen, a strong boost effect was seen after heterologous prime-boost immunization. Two additional studies involving more animals are ongoing, to strengthen the robustness of the nonclinical efficacy data, and also to optimize the prime-boost schedule so as to obtain induction of protective immunity as quickly as possible, to specifically respond to the Ebola virus disease outbreak in West Africa.

Toxicology

A repeated-dose toxicity study in rabbits was performed with prime-boost combinations of Ad26.ZEBOV and MVA-BN-Filo. The different dose regimens were well tolerated when administered twice by intramuscular (IM) injection to New Zealand White rabbits with a 14-day interval period. Additionally, the objective was to assess the persistence, reversibility or delayed onset of any effects after a 14-day treatment-free period. In the heterologous prime-boost regimen, either vector or both were used to prime a filovirus-specific immune response and the other/same vector or both were used to boost the immune response 2 weeks later. All vaccine dosing regimens resulted in detectable EBOV GP-specific antibody titers. No significant toxicological effects (no adverse effects) were observed. The immune response was associated with transient increases in fibrinogen, C-reactive protein, globulin, decreases in hematocrit and hemoglobin, and microscopic findings in draining iliac lymph nodes, spleen and at the injection sites. The findings were noted to be recovering over a 2-week treatment-free period and were considered to reflect a physiological response associated with vaccination. There were no effects noted that were considered to be adverse.

Biodistribution

Single-dose biodistribution studies in rabbits were performed using the MVA-BN vector or the Ad26 vector in combination with another insert (Ad26.ENVA.01: an experimental, prophylactic Ad26 vector expressing the human immunodeficiency virus [HIV] type 1, Clade A envelope protein). MVA-BN distributed to the skin, muscle, blood, spleen, lung, liver, and pooled lymph nodes and was rapidly cleared (within 48 hours following vaccination). Ad26.ENVA.01 was primarily localized in the injection site muscle, the regional lymph nodes and the spleen. Three months after the single IM injection of Ad26.ENVA.01, the vaccine was cleared from most of the examined tissues. As biodistribution is dependent on the vector platform (MVA or Ad26) and not on the insert, it can be assumed that recombinant MVA-BN-Filo or Ad26.ZEBOV is distributed in the same way as the MVA-BN vector or Ad26.ENVA.01 vector, respectively.

1.1.2. Clinical Studies

1.1.2.1. Safety Profile of Ad26-based Vaccines

To date, no human clinical studies have been completed with Ad26.ZEBOV or MVA-BN-Filo. The safety/tolerability and immunogenicity of the Ad26.ZEBOV and MVA-BN-Filo vaccines are being assessed in the ongoing Phase 1 studies (VAC52150EBL1001 and VAC52150EBL1002), where monovalent Ad26.ZEBOV and multivalent MVA-BN-Filo are combined in homologous or heterologous prime-boost regimens in which each vector is used to prime a filovirus-specific immune response followed by a boost immunization with the same or the other vector 2 to 8 weeks later. In addition, 2 other Phase 1 studies are planned to be conducted in Africa (VAC52150EBL1003 and VAC52150EBL1004). Two additional Phase 1 studies investigating MVA-BN-Filo are also ongoing (EBL01 and CVD-Mali Ebola Vaccine #1000). Refer to the latest versions of the Ad26.ZEBOV and MVA-BN-Filo Investigator's Brochures and Addenda (if applicable) for more details.[12,13,14]

Limited data from the ongoing Phase 1 studies with Ad26.ZEBOV and MVA-BN-Filo are available. Study VAC52150EBL1001 completed enrollment of 87 subjects and the blinded phase of the study is ongoing. Study VAC52150EBL1002 completed enrollment of 92 subjects and the blinded phase of the study is also ongoing.

Safety data generated with the 2 vectors containing different inserts are provided below:

Safety Data From Other Ad26-based Vaccine Programs

Ad26.ZEBOV is a monovalent recombinant, replication-incompetent Ad26-based vector. Only limited clinical data are available for Ad26.ZEBOV. However, adenovirus vaccine programs with other gene inserts revealed no significant safety issues. The data described below are based on the evaluation of the prototype vaccine Ad26.ENVA.01, which expresses the HIV envelope gene.[15]

Three randomized, placebo-controlled, Phase 1 studies (IPCAVD-001, IPCAVD-003, IPCAVD-004) have evaluated the safety and immunogenicity of the prototype vaccine Ad26.ENVA.01. This prototype vaccine has been administered to more than 200 healthy, HIV-negative subjects between the ages of 18 and 50 years in the United States and Africa.[15,16,17]

- In the dose-escalation study IPCAVD-001 (n=60), 2 or 3 IM doses of Ad26.ENVA.01 ($1x10^9$, $1x10^{10}$, $5x10^{10}$, $1x10^{11}$ viral particles [vp]) were given to Ad26 seronegative subjects. There were no deaths or vaccine-related serious adverse events. Ad26.ENVA.01 was generally well tolerated at all 4 dose levels with minimal reactogenicity observed in the $1x10^9$ and $1x10^{10}$ vp dose groups. Moderate to severe malaise, myalgia, fatigue and chills occurred in the majority of subjects 12 to 18 hours after the first dose of $1x10^{11}$ vp, but were resolved within 24 to 36 hours and were not seen after the second injection at this dose level. Two subjects in the $1x10^{11}$ vp dose group chose not to have the second injection, however, one of them decided to have the 6-month injection. Envelope-specific humoral and cell-mediated immune responses were induced at all 4 dose levels of vaccine.[2,4]

- In the single-dose study IPCAVD-003 (n=24), an IM dose of Ad26.ENVA.01 ($5x10^{10}$ vp) or placebo was given to subjects, who were stratified according to baseline Ad26 immune status, to evaluate the safety, mucosal immunogenicity and innate immune responses. Local reactogenicity comprised moderate injection site pain/tenderness and/or moderate to severe erythema which resolved within 3 days of vaccination. Transient systemic reactogenicity comprised headache, chills, joint pain, myalgia, malaise/fatigue, and fever. No deaths or vaccine-related serious adverse events were observed. Vaccination elicited both systemic and mucosal envelope-specific humoral and cellular immune responses. No increased activated total or vector-specific mucosal $CD4^+$ T-lymphocytes following vaccination were detected in the colorectal mucosa, indicating that vaccination with Ad26 did not increase mucosal inflammation.[1]
- In study IPCAVD-004 (n=217), the safety and immunogenicity of IM doses of Ad26.ENVA.01 and Ad35.ENV (an Ad35 vector expressing an HIV envelope GP used in that study at a dose of $5x10^{10}$ vp), given in heterologous and homologous prime-boost regimens at 3- versus 6-month intervals, was evaluated. There were 452 adverse events reported by 84 of 176 Ad26-vaccine recipients (47.7%), the majority being mild (75.5%) in severity. The proportion of subjects with moderate or severe symptoms was not statistically significantly different between vaccine and placebo. There were 3 serious adverse events: 2 serious adverse events in placebo recipients (grade 3 peritonsillar abscess and grade 4 migraine headache, both resolved with no residual effects) and 1 serious adverse event in an Ad35/Ad26 vaccine recipient (grade 4 acute myelogenous leukemia, resolved with sequelae). No deaths or vaccine-related serious adverse events were reported. Overall, 97% to 100% of subjects developed anti-envelope binding antibodies (enzyme-linked immunosorbent assay [ELISA]) after a second dose, with heterologous and homologous regimens being comparable. Immune responses in groups who received 3-month and 6-month schedules were comparable. Four weeks post-vaccination, interferon (IFN)-γ enzyme-linked immunospot (ELISpot) assay showed response rates between 44% and 100%. The heterologous and homologous regimens were comparable. There was induction of Ad26-neutralizing antibodies in the majority of vaccine recipients after 2 immunizations with Ad26.ENVA.01.[19]

In addition, the sponsor performed a Phase 1/2a double-blind, randomized, placebo-controlled, dose-escalation study (MAL-V-A001) to evaluate the safety, tolerability and immunogenicity of 2 dose levels ($1x10^{10}$ and $5x10^{10}$ vp) of Ad35.CS.01/Ad26.CS.01 (both expressing the malaria *Plasmodium falciparum* circumsporozoite antigen) prime-boost regimens in healthy subjects. The dose-escalation phase was followed by an evaluation of efficacy of the higher dose level in an experimental malaria challenge. A total of 42 subjects were enrolled and were vaccinated. The analysis of adverse events did not show any consistent pattern suggestive of an association of Ad35.CS.01 or Ad26.CS.01 with specific adverse events. There were no serious adverse events reported during the study. No subject discontinued during a study phase (vaccination or challenge) due to adverse events. One subject in the high-dose group completed the vaccination phase and the final safety follow-up visit but did not take part in any challenge phase activities because of ongoing dyspnea. The most common related adverse events after each vaccination were injection site pain, malaise, headache, myalgia and chills. The incidence of vaccine-related adverse events was generally higher in the high-dose group than in the low-dose group. In general, incidence of malaise, headache, and myalgia were higher after the third dose (Ad26) than after the first or second doses (Ad35). Injection site pain was more commonly reported in

the low and high-dose groups than by placebo subjects. There were no clinically significant changes in laboratory test parameters or vital signs data.[5]

Recent data indicate that administration of a deoxyribonucleic acid (DNA) vaccine expressing EBOV Mayinga GP, the same GP as in the Ad26.ZEBOV component, was safe, well tolerated and immunogenic in a Phase 1 clinical study. During this study, 9 subjects received three 4-weekly IM doses of vaccine (4 mg/dose), followed by a boost at ≥32 weeks in 8 subjects.[22]

Based on the previous clinical experience of Ad26 vector with different inserts, there has been no impact of Ad26 seropositivity on subjects' safety and only limited impact on immunogenicity results. Therefore, there are no safety concerns with regard to the inclusion of Ad26 seropositive subjects in the study, and the study subjects will not be screened for Ad26 seropositivity as part of the study eligibility criteria. The purpose of the Ad26 seropositivity assessment at baseline is to evaluate its impact, if any, on vaccine immunogenicity.

1.1.2.2. Safety Profile of MVA-BN-based Vaccines

MVA-BN is a further attenuated version of the MVA virus, which in itself is a highly attenuated strain of the poxvirus Chorioallantois Vaccinia Virus Ankara. MVA-BN induces strong cellular activity as well as a humoral (antibody) immune response and has demonstrated an ability to stimulate a response even in individuals with pre-existing immunity against Vaccinia. One of the advantages of MVA-BN is the virus' inability to replicate in a vaccinated individual. The replication cycle is blocked at a very late stage, which ensures that new viruses are not generated and released. This means that the virus cannot spread in the vaccinated person and none of the serious side effects normally associated with replicating Vaccinia viruses have been seen with MVA-BN.

MVA-BN (MVA-BN®, trade name IMVAMUNE® outside the European Union [EU], invented name IMVANEX® in the EU) has received marketing authorization in the EU for active immunization against smallpox in adults, and in Canada for persons 18 years of age and older who have a contraindication to the first or second generation smallpox vaccines including individuals with immune deficiencies and skin disorders.[11] A Phase 3 clinical study (POX-MVA-013) has been completed (ClinicalTrials.gov Identifier: NCT01144637).[6] Results of completed and ongoing clinical studies of MVA-BN-based vaccines in more than 8,100 individuals, including elderly, children and immunocompromised subjects in whom replicating vaccines are contraindicated, have shown that the platform displays high immunogenicity and a favorable safety profile.[20] Across all clinical studies, no trends for unexpected or serious adverse reactions due to the product were detected.

Safety information was combined from the first 2 studies of MVA-BN-Filo (VAC52150EBL1001 and VAC52150EBL1002). In general, MVA-BN-Filo has been shown to be well tolerated.[14]

Three fifths of the subjects reported at least one local site reaction (injection site pain, tenderness, warmth, redness, swelling and/or itching) following administration of MVA-BN-

Filo; mostly of mild severity. The most commonly reported local site reaction was pain at the injection site. All the local reactions resolved to normal without any lasting effects.

At least one general symptom was reported in two fifths of the subjects following MVA-BN-Filo administration. The most common general symptoms were fatigue, headache, myalgia (muscle pain) and nausea. All general symptoms were transient and resolved without lasting effects.

Changes in laboratory tests were reported following MVA-BN-Filo administration which included hypokalemia and decreased numbers of neutrophils. Both changes in laboratory tests were seen in similar numbers of participants following MVA-BN-Filo and the dummy (placebo) vaccine. Less frequently, events of decreased hemoglobin levels were reported. The changes in laboratory tests were not associated with any complaints or symptoms.

Extensive nonclinical studies support the safety profile of the MVA-BN strain.[23,24]

1.1.2.3. Relevant Safety Information from Ongoing VAC52150 Studies

One subject in this study experienced a serious and very rare condition called “Miller Fisher syndrome”. This condition consists of double vision, pain on moving the eye, and difficulty with balance while walking. Miller Fisher syndrome most commonly occurs following a recent infection. The subject experienced these symptoms about a week after suffering from a common cold and fever. The event happened about a month after boost vaccination with either MVA-BN-Filo or placebo. This subject had to go to the hospital for treatment and has recovered. After an extensive investigation, the event has been considered to be doubtfully related to vaccine and most likely related to the previous common cold.

In the ongoing clinical studies with more than 2,000 participants, there have been a few reports of mild to moderate tingling especially in the hands and feet or a sensation of mild to moderate muscle weakness in subjects vaccinated with Ad26.ZEBOV or placebo. These symptoms have been observed to last no more than 24 to 48 hours in the majority of cases but can last for several weeks before going away on their own. These types of symptoms have also been reported following administration of other licensed vaccines and following acute viral infections of various types. One serious case of probable peripheral sensory neuropathy of moderate severity has occurred and has been ongoing for several months, interfering with some of the subject’s daily activities.

1.1.2.4. Viral Shedding

Viral shedding information is available from 6 clinical studies with Ad-vectored vaccines against HIV type 1 (using Ad26 and Ad35: Ad26.ENVA.01 and Ad35.ENV) and *Mycobacterium tuberculosis* (using Ad35: AERAS-402). Viral shedding was not observed in any of these clinical studies. In a clinical study evaluating viral shedding of Ad26.ENVA.01 and Ad35.ENV (Study IPCAVD-004), all cultures from oropharyngeal swabs and urine were negative for adenovirus; in 5 clinical studies evaluating viral shedding of AERAS-402 (Studies C-001-402, C-003-402, C-008-402, C-009-402, C-017-402), no shedding of AERAS-402 was seen in any of the urine or throat cultures.[12]

MVA-BN-Filo is an attenuated recombinant MVA incapable of replication in human cells with a block in the late stage of virus replication. In human cells, upon infection, viral genes are expressed, but no infectious progeny virus is produced. Given the inability of virus assembly and very limited host range of the vector, no viral shedding studies were performed.

1.2. Risk Benefit Section

1.2.1. Known Benefits

The clinical benefit of prime-boost combinations of Ad26.ZEBOV and MVA-BN-Filo is to be established.

1.2.2. Potential Benefits

There is no known potential individual benefit from vaccination for the subjects at the current stage; others may benefit from knowledge gained in this study that may aid in the development of an Ebola vaccine.

1.2.3. Known Risks

To date, there are only limited data from the Phase 1 studies with Ad26.ZEBOV and MVA-BN-Filo available. However, Ad26- and Ad35-based vaccines with other gene inserts have been administered to a limited number of human volunteers in clinical studies. These other vaccines mainly elicited some solicited local and systemic reactions, as expected with injectable vaccines, and no serious safety concerns in study participants. MVA-BN-based vaccines have been administered to more than 8,100 individuals without unexpected or serious adverse reactions reported. For details, see the safety data presented in Section 1.1.

1.2.4. Potential Risks

The following potential risks for Ad26.ZEBOV and MVA-BN-Filo will be monitored during the study and are specified in the protocol:

Risks Related to Vaccines

Subjects may exhibit general signs and symptoms associated with administration of a vaccine, or a placebo vaccination, including nausea/vomiting, headache, myalgia, arthralgia, fever, fatigue/malaise and chills. In addition, subjects may experience local (injection site) reactions such as pain/tenderness, erythema, induration/swelling, and itching at the injection site. These events will be monitored, but are generally short-term and do not require treatment.

Subjects may have an allergic reaction to the vaccination. An allergic reaction may cause a rash, hives or even difficulty breathing (anaphylaxis). Severe reactions are rare. Medications must be available in the clinic to treat serious allergic reactions.

The risks related to vaccine-induced seropositivity (VISP) are discussed in Section 9.3.2.

Risk of Myo/Pericarditis

While replicating smallpox vaccines have been associated with an increased risk to develop myo/pericarditis,[21] this has not been observed with MVA-BN and is not expected with this highly attenuated, non-replicating vaccine. Based on observations with first- and second-generation replication-competent smallpox vaccines, particular attention has been placed on the monitoring for cardiac signs and symptoms in all clinical studies using MVA-BN. Despite the close cardiac monitoring, no event indicating a case of pericarditis has been observed in any completed MVA-BN study. There has been 1 case of chest pain that might be indicative of pericarditis (consisting of chest pain only with no other cardiac findings suggestive of pericarditis) with previous MVA use although this diagnosis was not finally confirmed and the subject fully recovered. In a review of prospective surveillances for cardiac adverse events in 6 different clinical studies in 382 subjects receiving MVA vaccines, only 1 (0.3%) subject met the criteria for vaccine-induced myocarditis and eventually the subject was found to suffer from exercise-induced palpitations. Self-limited mild elevations in troponin I were recorded in 3 (0.8%) subjects without evidence of myo/pericarditis.[7] Based on the current exposure data in more than 8,100 subjects vaccinated with MVA-BN and other MVA-BN recombinant products, the safety profile of MVA-BN has shown to be comparable with other licensed, live attenuated vaccines.

Pregnancy and Birth Control

The effect of the study vaccines on a fetus or nursing baby is unknown, as well as the effect on semen, so women of childbearing potential, and men having sexual intercourse with women, are required to agree to practice adequate birth control measures for sexual intercourse from at least 28 days before the prime vaccination (or prior to enrollment for men) until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer), see Sections 4.1 and 4.3. Women who are pregnant or breast-feeding, or are planning to become pregnant while enrolled in the study until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer) will be excluded from enrollment into the study.

Women of childbearing potential must also agree not to donate eggs (ova, oocytes) for the purposes of assisted reproduction from the start of screening onwards until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer). Men must also agree not to donate sperm from the start of screening onwards until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer).

After unblinding for the primary analysis, placebo subjects may stop birth control measures or resume their pre-study birth control measures, and may donate eggs (women) or sperm (men).

Risks from Blood Draws

Blood draws may cause pain/tenderness, bruising, bleeding, and, rarely, infection at the site where the blood is taken.

Unknown Risks

There are no clinical data on the use of either vaccine (Ad26.ZEBOV or MVA-BN-Filo) in:

- Pediatric subjects (<18 years);
- Pregnant or nursing women;
- Adults >50 years;
- Immunocompromised subjects (including those with HIV infection).

There may be other serious risks that are not known.

1.2.5. Overall Benefit/Risk Assessment

Based on the available data and proposed safety measures, the overall benefit/risk assessment for this clinical study is considered acceptable for the following reasons:

- Preliminary safety data from the ongoing Phase 1 studies and safety data generated with the 2 vaccines with different inserts revealed no significant safety issues (see Section 1.1 and 1.2.3). Further experience from Ad26.ZEBOV or MVA-BN-Filo will be gained from currently ongoing clinical studies.
- Only subjects who meet all inclusion criteria and none of the exclusion criteria (specified in Section 4) will be allowed to participate in this study. The selection criteria include adequate provisions to minimize the risk and protect the well-being of subjects in the study.
- Safety will be closely monitored throughout the study:
  - After each vaccination, subjects will remain at the site for a total of 60 (±15) minutes post-vaccination to monitor the development of any acute reactions, or longer if deemed necessary by the investigator (eg, in case of grade 3 adverse events). The subjects will be closely observed by study-site personnel for the first 30 (±10) minutes after each vaccination and again at 60 (±15) minutes post-vaccination, and any unsolicited, solicited local or systemic adverse events will be documented during this period. Subjects will use a diary to document symptoms of solicited local and systemic adverse events in the evening after each vaccination and then daily for the next 7 days.
  - The investigator or the designee will document unsolicited adverse events from signing of the informed consent form (ICF) onwards until 42 days post-boost, and serious adverse events and immediate reportable events (IREs) from signing of the ICF onwards until the end of the study.
  - Safety measures, including clinical laboratory testing, electrocardiogram (ECG) and vital sign measurements, physical examinations and pregnancy testing, will be performed at scheduled visits during the study, which lasts up to 1 year after the prime vaccination in subjects who received Ad26.ZEBOV or MVA-BN-Filo.

  - Any clinically significant abnormalities (including those persisting at the end of the study/early withdrawal) will be followed by the investigator until resolution or until a clinically stable endpoint is reached.

- Several safety measures are included in this protocol to minimize the potential risk to subjects, including the following:
  - The neuroinflammatory disorders listed in Section 12.1.1 should be categorized as IREs and should be reported to the sponsor as described in Section 12.3.3.
  - There are pre-specified pausing rules that would result in pausing of further vaccination if predefined conditions occur, preventing exposure of new subjects to study vaccine until an Independent Data Monitoring Committee (IDMC) reviews all safety data (see Section 9.2.3 for further details).
  - Subjects will discontinue study vaccine for the reasons included in Section 10.2.
  - If acute illness (excluding minor illnesses such as diarrhea or mild upper respiratory tract infection) or fever (body temperature ≥38.0°C) occur at the scheduled time for vaccination, the subject may be vaccinated up to 10 days beyond the window allowed for the scheduled vaccination, or be withdrawn from that vaccination at the discretion of the investigator and after consultation with the sponsor (see Section 6.2).
  - Contraindications to boost vaccinations are included in Section 6.3.
  - If a subject withdraws from the study (withdrawal of consent), he/she maintains the option to participate in the safety follow-up.

1.3. Overall Rationale for the Study

In nonclinical studies in the Cynomolgus macaque model, heterologous prime-boost regimens of a multivalent mixture of Ad26 vectors (each expressing EBOV Mayinga, SUDV or MARV GP) and MVA-BN-Filo provided complete protection against the highly pathogenic wild-type EBOV Kikwit 1995 variant (report pending). Further nonclinical studies are ongoing to evaluate the protection of the multivalent vaccine regimen in additional animals and to assess the protective efficacy of a combination regimen of Ad26.ZEBOV and MVA-BN-Filo (either a simultaneous administration or as prime-boost regimen).

In humans, both Ad26- and MVA-based vaccines containing various antigenic inserts have been shown to be safe and immunogenic (see Section 1.1). To date, more than 230 subjects have received the sponsor's Ad26-based vaccines in completed clinical studies (based on the adenoviral vaccine safety database report [dated 20 March 2015]). Up to 28 October 2015, 227 subjects received Ad26.ZEBOV in ongoing studies. The MVA-BN platform is the basis of the non-replicating smallpox vaccine registered in Canada and Europe, and has been safely used in more than 7,600 humans.[20] Although routinely used by the subcutaneous route, MVA-BN at a dose of $1x10^8$ $TCID_{50}$ (50% Tissue Culture Infective Dose) has been demonstrated to be as safe and immunogenic when used by the IM route.[8,25] The IM route has been chosen for the present study.

The unprecedented size and scale of the ongoing Ebola virus disease outbreak that started in December 2013 in Guinea and subsequently spread to Sierra Leone, Nigeria and Liberia, led to the outbreak being declared a public health emergency of international concern in August 2014 by the WHO. This study is one of a series of studies to evaluate the heterologous combination of Ad26.ZEBOV and MVA-BN-Filo as a possible vaccine regimen to prevent Ebola virus disease. It will test schedules evaluated in ongoing NHP challenge and Phase 1 clinical studies.

In this Phase 2 study, the sponsor's Ad26 vector expressing the EBOV Mayinga GP (Ad26.ZEBOV) and the MVA-BN vector with EBOV, SUDV and MARV GP inserts and TAFV NP insert (MVA-BN-Filo) will be evaluated as a heterologous prime-boost regimen, in which one study vaccine (Ad26.ZEBOV) is used to prime a filovirus-specific immune response and the other study vaccine (MVA-BN-Filo) is used to boost the immune response 28, 56 or 84 days later. The EBOV GP that is currently circulating in West Africa has 97% homology to the EBOV GP used in this vaccine regimen.[3] The concept of a prime-boost regimen that will be evaluated in the Phase 2 studies with the candidate prophylactic Ebola vaccines Ad26.ZEBOV and MVA-BN-Filo is supported by the results of clinical studies with candidate malaria vaccines which have demonstrated that Ad-based priming immunization followed by MVA vector boost induced high levels of immunity.

For the prevention of Ebola virus disease, shorter vaccination schedules may be relevant in the context of the epidemic and suitable for use during acute outbreaks of Ebola. When the current outbreak is under control, longer vaccination schedules may be more relevant than the shorter prime-boost intervals provided a more persistent immune response is observed. The 14-, 28- and 56-day prime-boost intervals are being evaluated in the current Phase 1 studies. An interval longer than 56 days may provide better protection than shorter schedules. Therefore, the objective of this study is to evaluate the immunogenicity of intervals that are slightly shorter as well as slightly longer relative to the 56-day schedule (ie, the schedule that yielded 100% protection in NHP).

The 3 different time intervals (of 28, 56 or 84 days) between the prime and the boost vaccination will be evaluated for safety and tolerability as well as for immunogenicity. These 3 prime-boost regimens will only differ in the timing of the boost vaccination, while the dose of each vaccine and the sequence of vaccination will be identical. The prime vaccine consists of Ad26.ZEBOV at a dose of $5x10^{10}$ vp and the boost vaccine consists of MVA-BN-Filo at a dose of $1x10^{8}$ infectious units (Inf.U, nominal titer). The MVA-BN-Filo dose to be used corresponds to the dose of $1x10^{8}$ $TCID_{50}$ that is used in the current Phase 1 studies.

This study aims to evaluate the safety, tolerability and immunogenicity of the 3 prime-boost regimens, only different in terms of vaccination schedule with a time interval of 28, 56 or 84 days between the prime and the boost vaccination, and to enlarge the safety and immunogenicity database for the selected Ad26.ZEBOV and MVA-BN-Filo prime-boost regimen(s).

2. OBJECTIVES AND HYPOTHESIS

2.1. Objectives

Primary Objective

The primary objective is to assess the safety and tolerability of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.

Secondary Objective

The secondary objective is to assess humoral immune responses, as measured by ELISA, to the EBOV GP 21 days post boost of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.

Exploratory Objectives

The exploratory objectives are:

- To assess humoral immune responses, as measured by ELISA, to the EBOV GP at other relevant time points of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.
- To assess the neutralizing capacity of the EBOV GP-specific humoral immune response, as measured by virus neutralization assay, at selected time points of 3 vaccination schedules of Ad26.ZEBOV and MVA-BN-Filo administered IM as heterologous prime-boost regimens on Days 1 and 29, Days 1 and 57, or Days 1 and 85.
- To further explore humoral immune responses at selected time points to different EBOV GPs, filovirus GPs and/or TAFV NP as well as the adenovirus and/or MVA backbones of the various vaccination schedules tested, if assays are available.
- To explore cellular immune responses at selected time points to different EBOV GPs, filovirus GPs and/or TAFV NP of the various vaccination schedules tested, if assays are available.

2.2. Hypothesis

As this study is designed to provide descriptive information regarding safety and immunogenicity without formal treatment comparisons, no formal statistical hypothesis testing is planned.

3. STUDY DESIGN AND RATIONALE

3.1. Overview of Study Design

This is a randomized, observer-blind, placebo-controlled, parallel-group, multicenter, Phase 2 study to evaluate the safety, tolerability and immunogenicity of 3 heterologous prime-boost regimens using Ad26.ZEBOV at a dose of $5x10^{10}$ vp as prime and MVA-BN-Filo at a dose of

$1x10^8$ Inf.U (nominal titer) as boost at a 28-, 56- or 84-day interval in healthy adult subjects in Europe (United Kingdom [UK] and France). The 3 prime-boost regimens will only differ in the timing of the boost vaccination (ie, 28, 56 or 84 days after prime, respectively referred to as Groups 1, 2 and 3), while the dose of each study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo) and the sequence of vaccination will be identical.

The subject population will consist of healthy men and women aged between 18 and 65 years (inclusive), who have no prior exposure to Ebola virus (including travel to West Africa less than 1 month prior to screening) or a diagnosis of Ebola virus disease. Subjects who received a candidate Ebola vaccine or any experimental candidate Ad26- or MVA-based vaccine in the past or with known allergy or history of anaphylaxis or other serious adverse reactions to vaccines or vaccine products, including known allergy to egg, egg products and aminoglycosides will be excluded.

At study entry, subjects were offered the option to enroll into Cohorts I, II, or III in the UK or into Cohorts II or III in France. In Cohorts II and III in both countries, core immunogenicity assessments (humoral and cellular assays) will be performed. In Cohort II in both countries, additional immunogenicity assessments will be done. In Cohort I, plasmablast response kinetics will be evaluated for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK. In the UK, Cohorts I and III may start in parallel, while Cohort II in the UK can only start when the peak of B-cell response after prime vaccination in Cohort I is identified. In France, Cohorts II and III may start in parallel (as there will be no Cohort I) and are independent of Cohort I in the UK. A schematic overview of the study design, cohorts and groups is provided in Table 1.

Table 1: Schematic Overview of Study Design, Cohorts and Groups

Study Cohorts	Randomization Ratio (Active:Placebo)	Group 1 N=204	Group 2 N=204	Group 3* N=204	Cohort Total N=612	UK** N=321	France** N=291
Cohort I	-	10/0	10/0	10/0	**30**	30	-
Cohort II	14:1	84/6	84/6	84/6	**270**	135	135
Cohort III	10:3	80/24	80/24	80/24	**312**	156	156

Groups 1, 2 and 3: prime on Day 1, followed by boost 28, 56 or 84 days after prime, respectively; N: number of subjects to receive study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo)

* Randomization to Group 3 was stopped per Amendment 4 to focus on the schedules for which an indication will be sought.

** Enrollment in the entire study (ie, UK and France) will be stopped per Amendment 5.

Subjects in each cohort were randomized at baseline (on Day 1) in a 1:1:1 ratio to Groups 1, 2 and 3 (ie, the vaccination schedule). A schematic overview of the study vaccination schedules is provided in Table 2. Within each group in Cohort I, subjects will receive Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion. Randomization in Cohorts II and III was stratified by country. Within each group in Cohorts II and III, subjects were randomized to receive the prime-boost vaccination with either Ad26.ZEBOV followed by MVA-BN-Filo, or placebo in a 14:1 and 10:3 ratio, respectively (see Table 1). Randomization in each group (for Cohorts II and III) was stratified further according to age at randomization (≤50 years, >50 years), with at least 20%

(of the total of both cohorts) of subjects in the >50 years category. Note that enrollment will not be resumed.

Study-site personnel (except for those with primary responsibility for study vaccine preparation and dispensing), sponsor personnel and subjects in Cohorts II and III will be blinded to the study vaccine allocation until all subjects have completed the 6-month post-boost visit or discontinued earlier and the clinical database is locked. Refer to Section 5 for further details on blinding in case of interim analyses.

Table 2: Schematic Overview of Study Vaccination Schedules in the Three Groups Pooled Across Cohorts I, II and III

Group*	N		Prime	Boost		
			Day 1	Day 29	Day 57	Day 85
1	204	174	Ad26.ZEBOV	MVA-BN-Filo		
		30	placebo	placebo		
2	204	174	Ad26.ZEBOV		MVA-BN-Filo	
		30	placebo		placebo	
3**	204	174	Ad26.ZEBOV			MVA-BN-Filo
		30	placebo			placebo

N: number of subjects to receive study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo)
Ad26.ZEBOV dose level is $5x10^{10}$ vp in all groups, MVA-BN-Filo dose level is $1x10^{8}$ Inf.U (nominal titer) in all groups, placebo is 0.9% saline
* Enrollment in the entire study (ie, UK and France) will be stopped per Amendment 5.
** Randomization to Group 3 was stopped per Amendment 4 to focus on the schedules for which an indication will be sought.

All subjects will receive the study vaccine (Ad26.ZEBOV, MVA-BN-Filo or placebo) through IM injection (0.5 mL) in the deltoid muscle:

- Ad26.ZEBOV ($5x10^{10}$ vp) on Day 1, followed by a boost vaccination of MVA-BN-Filo ($1x10^{8}$ Inf.U, nominal titer) on Days 29, 57 or 85; *OR*
- Placebo (0.9% saline) on Day 1, followed by a boost vaccination of placebo (0.9% saline) on Days 29, 57 or 85.

Refer to Section 6 for further details on dosage and administration. After each vaccination, subjects will remain at the site for a total of 60 (±15) minutes post-vaccination to monitor for the development of any acute reactions, or longer if deemed necessary by the investigator (eg, in case of grade 3 adverse events).

The study consists of a screening phase of up to 12 weeks (starting from the moment the subject signs the ICF), a vaccination phase in which subjects will be vaccinated at baseline (Day 1), followed by a boost vaccination on Days 29, 57 or 85, and a post-vaccination phase and long-term follow-up phase until Day 365 (or until the start of the roll-over study or for an additional 12 months [whichever comes first] for subjects in France who agree to continue the long-term follow-up after Day 365, see details below and Section 9.1.5). After unblinding, the subjects who received placebo in Cohorts II and III will be contacted by the site to communicate that they have completed the study and no further follow-up is required. Unblinding will be done when the last subject in the study completes the 6-month post-boost visit or discontinues earlier and the

clinical database is locked. However, subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit. After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will continue the study until Day 365 (or until the start of the roll-over study or for an additional 12 months [whichever comes first] for subjects in France who agree to continue the long-term follow-up after Day 365, see details below and Section 9.1.5) visit to assess long-term safety and immunogenicity. Refer to Section 9.1.2 for subjects whose screening period was longer than the protocol-defined 12 weeks as a result of a pause.

The baseline visit may be scheduled as soon as the results of all screening assessments are known (but should occur within 12 weeks from screening, see Section 9.1.2) and show that the subject is eligible for inclusion. The administration of the prime vaccination will occur on Day 1 (baseline), after the completion of all baseline assessments.

The investigators, together with the sponsor's medical monitor, will be responsible for the safety monitoring of the study, and will halt vaccination of further subjects in case any of the pre-specified pausing rules described in Section 9.2.3 have been met. In addition, discontinuation of study vaccine should occur in any subject meeting the criteria outlined in Section 10.2. Criteria for postponement of vaccination at the scheduled time for vaccine administration and contraindications to boost vaccination have been defined and will be applied by the investigator (see Sections 6.2 and 6.3, respectively).

Safety will be assessed in all subjects from the 3 cohorts by collection of solicited local and systemic adverse events (reactogenicity), unsolicited adverse events, IREs and serious adverse events. The subjects will be closely observed by study-site personnel for the first 30 (±10) minutes after each vaccination and again at 60 (±15) minutes post-vaccination, and any unsolicited, solicited local or systemic adverse events will be documented during this period. Upon discharge from the site, subjects will receive a diary, a thermometer and a ruler to measure solicited local reactions and body temperature. Subjects will record symptoms of solicited local and systemic adverse events in the diary in the evening after each vaccination and then daily for the next 7 days. The investigator will document unsolicited adverse events from signing of the ICF onwards until 42 days post-boost, and serious adverse events and IREs from signing of the ICF onwards until the end of the study. The primary endpoints are adverse events, serious adverse events, IREs, and solicited local and systemic adverse events, see Section 9.2.1.

Other safety assessments include clinical laboratory testing, ECGs, vital signs (blood pressure, pulse/heart rate, and body temperature), physical examination and pregnancy testing at the time points indicated in the Time and Events Schedule.

Blood samples for core immunogenicity assessments (humoral and cellular assays) for the evaluation of secondary and exploratory endpoints as planned by the sponsor (see Table 4 in Section 9.3.2) will be collected at the time points indicated in the Time and Events Schedule. Blood samples for assessment of humoral immune responses will be obtained from all subjects in Cohorts II and III. Blood samples for assessment of cellular immune responses will be obtained

from subjects in Cohorts II and III at selected sites with the capabilities to process peripheral blood mononuclear cells (PBMC) (targeted at 10% of all subjects).

Blood samples for additional immunogenicity assessments for the evaluation of additional exploratory endpoints as planned by the academic consortium partners will be collected from subjects in Cohort II (see Table 6 in Section 9.3.2) at the time points indicated in the Additional Time and Events Schedule. Blood samples for plasmablast response kinetics will be collected in Cohort I (see Table 5) at the time points indicated in the Additional Time and Events Schedule for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK.

A blood sample for DNA collection will be collected at baseline from subjects in Cohort II at selected sites (as part of the additional immunogenicity assessments by the academic consortium partners, see Table 6) and who consent separately to this component of the study. Subject participation in genomic research is optional. The goal of this genomic research is to allow the identification of genetic factors that may influence the immunogenicity, safety, or tolerability of Ad26.ZEBOV and MVA-BN-Filo. Since the sample size is small, analysis will focus on validation of genes identified as possibly associated with these factors in previous (non-Ebola) studies and control of gene expression.

Optional blood samples for transcriptomics will be collected from subjects in Cohorts I and II (as part of the specific and additional immunogenicity assessments by the academic consortium partners, respectively, see Table 5 and Table 6) and who consent separately to the transcriptomics component of the study. Blood samples will be collected at the specified time points (see Additional Time and Events Schedule). The goal is to assess the RNA transcriptome (gene expression) in a systems vaccinology approach to identify differentially expressed genes which relate to the immunologic and safety phenotype of the response to Ad26.ZEBOV and MVA-BN-Filo, and for analysis of antibody genes.

If subjects provide separate consent, leftover blood samples will be stored for possible future scientific/genetic research (see Section 16.2.5). Subjects can withdraw consent for their samples to be used for future research at any time.

Interim analyses may be performed as detailed in Section 11.6. The primary analysis will be performed when all subjects have completed the 6-month post-boost visit or discontinued earlier and the clinical database is locked. This analysis will include 6-month post-boost safety and all available immunogenicity data up to this point. The final analysis will be performed when all subjects have completed the last study-related visit (see Section 10.1) or discontinued earlier.

An IDMC will be commissioned for this study. Refer to Section 11.7 for details.

The sponsor halted all vaccinations in this study due to the occurrence of a serious and very rare condition, Miller Fisher syndrome, reported in the current study VAC52150EBL2001 (described in Section 1.1.2.3) until a revised ICF containing updated safety language was prepared and approval to restart the study was granted by the relevant competent authority. The study pause

interrupted dosing of subjects, some awaiting prime vaccination and some awaiting boost vaccination. When approval was granted in the UK to restart the study under Amendment 4, the sponsor offered a late boost vaccination to those subjects who did not receive it yet, unless participants had withdrawn from the trial or were not eligible to receive the boost. Vaccinated subjects have been following the same post-boost vaccination schedule as those subjects unaffected by the pause. Subjects who did not receive the late boost were encouraged to return for the 1-year post-prime visit. After restart of the study in the UK under Amendment 4, screening for Groups 1 and 2 restarted, but randomization to Group 3 was stopped to focus on the schedules for which an indication will be sought. After approval of Amendment 5, enrollment in the entire study will be stopped.

Subjects in the UK and France who reach the Day 365 visit prior to unblinding will be approached to consent for enrollment into the VAC52150 Vaccine Development Roll-over study (VAC52150EBL4001) for long-term surveillance (for a total of up to 60 months after the prime vaccination) (see Section 9.1.6 for details). After unblinding, only subjects who received Ad26.ZEBOV and/or MVA-BN-Filo will remain in the VAC52150 Vaccine Development Roll-over study for long-term surveillance. After unblinding, subjects who received placebo and have already been enrolled into the VAC52150 Vaccine Development Roll-over study will be discontinued from further participation in the roll-over study. The parent(s)/legal guardian of children born to vaccinated female subjects who became pregnant with estimated conception within 28 days after vaccination with MVA-BN-Filo or within 3 months after vaccination with Ad26.ZEBOV, will also be approached to consent for enrollment of their offspring into the roll-over study, according to the same rules that apply for the other subjects. Subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study is opened, will have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months [whichever comes first] or discontinued earlier. These subjects will be contacted by telephone every 3 months after Day 365 (subjects will only visit the site when clinically indicated) for the collection of information on adverse events, serious adverse events, and IREs that were ongoing at the Day 365 visit, and to report and follow up on new serious adverse events, pregnancies, and IREs.

3.2. Study Design Rationale

The study design is largely driven by the accelerated development approach of the Ad26.ZEBOV and MVA-BN-Filo prime-boost combination. Based on the growing availability of data from the entire development program as the study proceeds, modifications to the study design may be considered.

Due to significant delays in scheduled boost vaccinations caused by study pauses required for safety evaluations, enrollment in the entire study will not be resumed. Since many subjects in France have had no boost vaccinations and many subjects in the UK have had a late boost vaccination, it will be very difficult to evaluate the planned dosing regimens. Therefore, no further subjects will be recruited in the entire study (ie, UK and France). Additional data on the planned dosing regimens will be obtained from other studies. Vaccinated subjects in both countries will still be followed per protocol for safety.

The elimination of one or more post-boost sampling time points for immunogenicity assessments may be considered to reduce blood sample volume in these subjects if preliminary findings suggest that these have no added informative value for the study.

Control and Blinding

At study entry, subjects were offered the option to enroll into Cohorts I, II or III (as applicable). Within each cohort, subjects were randomized at baseline (on Day 1) in a 1:1:1 ratio to one of the 3 vaccination schedules (Groups 1, 2 and 3). All groups in Cohort I will receive an IM injection with Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion. The rationale for this cohort is to identify the peak of the B-cell response for the determination of the optimal sampling time points regarding B-cell responses as part of the additional immunogenicity assessments in Cohort II in the UK. Within each group in Cohorts II and III, subjects were randomized in a ratio indicated in Table 1 to receive the prime-boost vaccination with either Ad26.ZEBOV followed by MVA-BN-Filo, or placebo. Randomization (to vaccination schedule and to study vaccine) was used to minimize bias in the assignment of subjects to groups, to increase the likelihood that known and unknown subject attributes (eg, demographic and baseline characteristics) are evenly balanced across groups, and to enhance the validity of possible statistical comparisons across groups. Note that randomization to Group 3 was not resumed per Amendment 4 and that enrollment in entire study will be stopped per Amendment 5.

A placebo control will be used (in Cohorts II and III) to establish the frequency and magnitude of changes in clinical and immunologic endpoints that may occur in the absence of Ad26.ZEBOV and MVA-BN-Filo. Blind treatment will be used to reduce potential bias during data collection and evaluation of clinical safety endpoints. Blinding will be guaranteed by the preparation of study vaccine by unblinded qualified study-site personnel not involved in any other study-related procedure and by the administration of vaccine in a masked syringe by a study vaccine administrator (see Definitions of Terms).

The rationale for defining the study as an observer-blind study is that the subjects and study-site personnel will not be blinded to the vaccination schedule and no additional placebo injections will be administered to mask the regimens across groups; blinding will only mask the administration (and observations) of Ad26.ZEBOV, MVA-BN-Filo or placebo within groups.

Study Groups

The 3 vaccination schedules (Groups 1, 2, and 3) will only differ in the timing of the boost vaccination (ie, 28-, 56- or 84-day interval), while the dose of each vaccine and the sequence of vaccination will be identical. The prime vaccine consists of Ad26.ZEBOV at a dose of $5x10^{10}$ vp and the boost vaccine consists of MVA-BN-Filo at a dose of $1x10^{8}$ Inf.U (nominal titer), which corresponds to the dose of $1x10^{8}$ $TCID_{50}$ that is used in the current Phase 1 studies. The safety, tolerability and immunogenicity results for the 3 vaccination schedules will be evaluated in the study.

DNA Collection

An optional blood sample for DNA collection will be collected at baseline from subjects in Cohort II at selected sites and who consent separately to this component of the study. It is recognized that genetic variation can be an important contributory factor to interindividual differences in vaccine response (both safety and immunogenicity) and can also serve as a marker for disease susceptibility and prognosis. Genomic research in vaccine studies may help to explain interindividual variability in clinical and immunologic outcomes and may help to identify population subgroups that respond differently to a vaccine. The goal of the genomic component is to collect DNA to allow the identification of genetic factors that may influence the immunogenicity, safety, or tolerability of Ad26.ZEBOV and MVA-BN-Filo. DNA samples may be used to help address emerging issues and to enable the development of safer, more effective vaccines. Since the sample size is small, analysis will focus on validation of genes identified as possibly associated with these factors in previous (non-Ebola) studies and control of gene expression.

Transcriptomics

Optional blood samples for transcriptomics will be collected at the specified time points from subjects in Cohorts I and II and who consent separately to the transcriptomics component of the study. The goal is to assess the RNA transcriptome (gene expression) in a systems vaccinology approach to identify differentially expressed genes which relate to the immunologic and safety phenotype of the response to Ad26.ZEBOV and MVA-BN-Filo (taking advantage of the rich phenotypic data being collected in the safety, and cellular and humoral immunologic studies) and for analysis of antibody genes.

Future Research

If subjects provide separate consent, leftover blood samples will be stored for possible future scientific/genetic research (see Section 16.2.5). Future scientific research may be conducted to further investigate Ebola vaccine- and disease-related questions. This may include the development of new or the improvement of existing techniques to characterize EBOV-directed immune responses or diagnostic tests. Subjects can withdraw consent for their samples to be used for future research at any time.

4. SUBJECT POPULATION

Screening of subjects for eligibility was performed within 12 weeks before administration of the study vaccine on Day 1. The inclusion and exclusion criteria for enrolling subjects in this study are described in the following 2 subsections. If there is a question about the inclusion or exclusion criteria below, the investigator must consult with the appropriate sponsor representative and resolve any issues before enrolling a subject in the study. Waivers are not allowed.

For a discussion of the statistical considerations of subject selection, refer to Section 11.2, Sample Size Determination.

4.1. Inclusion Criteria

Each potential subject must satisfy all of the following criteria to be enrolled in the study.

1. Signed an ICF indicating that he/she understands the purpose of, and procedures required for, the study and is willing to participate in the study.

2. Man or woman, between 18 and 65 years of age, inclusive, at randomization.

3. Healthy in the investigator's clinical judgment on the basis of medical history, physical examination, ECG, and vital signs performed at screening.

4. Criterion modified per Amendment 4

4.1. Healthy on the basis of clinical laboratory tests performed at screening. If the results of the laboratory screening tests are outside the normal reference ranges, the subject may be included only if the investigator judges the abnormalities or deviations from normal to be not clinically significant or to be appropriate and reasonable for the population under study. This determination must be recorded in the subject's source documents and initialed by the investigator.

Note: The safety laboratory assessments at screening are to be performed within 28 days prior to the prime vaccination on Day 1 (including Day 1 before vaccination) and may be repeated if they fall outside this time window.

Note: In case of menstruation, urinalysis must be postponed but a result should be available before the prime vaccination.

Note: If laboratory screening tests are out of range and deemed clinically significant, repeat of screening tests is permitted once using an unscheduled visit during the screening period to assess eligibility.

5. Criterion modified per Amendment 4

5.1. Contraceptive use by men and women should be consistent with local regulations regarding the use of contraceptive methods for subject participating in clinical studies.

Before randomization, a woman must be either:

- Of childbearing potential and practicing (or intending to practice) a highly effective method of birth control consistent with local regulations regarding the use of birth control methods for subjects participating in clinical studies, beginning at least 28 days prior to vaccination. The sponsor considers the following methods of birth control to be highly effective: established use of oral, injected or implanted hormonal methods of contraception; placement of an intrauterine device (IUD) or intrauterine system (IUS); barrier methods: condom or occlusive cap (diaphragm or cervical/vault caps) with spermicidal foam/gel/film/cream/suppository; male partner sterilization (the vasectomized partner should be the sole partner for that subject); true abstinence (when this is in line with the preferred and usual lifestyle of the subject); *OR*

- Not of childbearing potential: postmenopausal (>45 years of age with amenorrhea for at least 2 years or ≤45 years of age with amenorrhea for at least 6 months, and a serum follicle stimulating hormone (FSH) level >40 IU/L or mIU/mL); permanently sterilized (eg, bilateral tubal occlusion [which includes tubal ligation procedures as consistent with local regulations], hysterectomy, bilateral salpingectomy, bilateral oophorectomy); or otherwise be incapable of pregnancy.

Note: If the social situation of a woman of childbearing potential changes (eg, woman who is not heterosexually active becomes active), she must begin a highly effective method of birth control, as described above.

Note: Women ≤45 years of age with amenorrhea for ≤6 months are considered of childbearing potential and do not need FSH testing.

6. Woman of childbearing potential must have a negative serum (β-human chorionic gonadotropin [β-hCG]) at screening and a negative urine β-hCG pregnancy test immediately prior to each study vaccine administration.
7. Man who is sexually active with a woman of childbearing potential and has not had a vasectomy performed more than 1 year prior to screening must be willing to use condoms for sexual intercourse beginning prior to enrollment.
8. Available and willing to participate for the duration of the study and follow-up visits.
9. Willing and able to comply with the protocol requirements, including the prohibitions and restrictions specified in Section 4.3.
10. Willing to provide verifiable identification.
11. Having a means to be contacted.
12. Having coverage by a social security insurance (for sites in France) and allowing use and storage of National Insurance Number on The Over-volunteering Prevention System (TOPS) database and bank account information for purposes of reimbursement (for sites in UK).

4.2. Exclusion Criteria

Any potential subject who meets any of the following criteria will be excluded from participating in the study.

1. Having received any candidate Ebola vaccine.
2. Diagnosed with Ebola virus disease, or prior exposure to Ebola virus, including travel to West Africa less than 1 month prior to screening. West Africa includes but is not limited to the countries of Guinea, Liberia, Mali, and Sierra Leone.

   Note: Participation of international volunteers to Ebola operations is allowed, but they should comply with the prohibitions and restrictions as specified in Section 4.3.

3. Having received any experimental candidate Ad26- or MVA-based vaccine in the past.

   Note: Receipt of any approved vaccinia/smallpox vaccine or experimental Ad-vector vaccine other than Ad26 at any time prior to study entry is allowed.

4. Known allergy or history of anaphylaxis or other serious adverse reactions to vaccines or vaccine products (including any of the constituents of the study vaccines [eg, polysorbate 80, ethylenediaminetetraacetic acid (EDTA) or L-histidine for Ad26.ZEBOV vaccine; and tris (hydroxymethyl)-amino methane (THAM) for MVA-BN-Filo vaccine]), including known allergy to egg, egg products, and aminoglycosides.

5. Presence of acute illness (this does not include minor illnesses such as diarrhea or mild upper respiratory tract infection) or temperature ≥38.0ºC on Day 1. Subjects with such symptoms will be excluded from enrollment at that time, but may be rescheduled for enrollment at a later date.

6. Positive hepatitis B surface antigen (HBsAg) or hepatitis C virus (HCV) antibody at screening.

7. HIV type 1 or type 2 infection.

8. Pregnant, breast-feeding, or planning to become pregnant while enrolled in this study until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer).

9. Presence of significant conditions or clinically significant findings during screening of medical history, physical examination, ECG, vital signs, or laboratory testing for which, in the opinion of the investigator, participation would not be in the best interest of the subject (eg, compromise the safety or well-being) or that could prevent, limit, or confound the protocol-specified assessments.

10. History of or underlying liver or renal insufficiency, or significant cardiac, vascular, pulmonary (eg, persistent asthma), gastrointestinal, endocrine, neurologic, hematologic, rheumatologic, psychiatric, or metabolic disturbances.

11. History of malignancy other than squamous cell or basal cell skin cancer, unless there has been surgical excision, that is considered cured.

12. Major surgery (per the investigator's judgment) within the 4 weeks prior to screening, or planned major surgery during the study (from the start of screening onwards).

13. Post-organ and/or stem cell transplant whether or not with chronic immunosuppressive therapy.

14. Received any disallowed therapies as noted in Section 8 before the planned first administration of the prime vaccine on Day 1.

15. Received an investigational drug or investigational vaccines or used an invasive investigational medical device within 3 months prior to screening, or current or planned participation in another clinical study during the study.

    Note: Participation in an observational clinical study is allowed.

16. Donation of a unit of blood within 12 weeks before Day 1 or plans to donate blood during participation in the study (from the start of screening onwards).

17. Receipt of blood products or immunoglobulin within 3 months prior to screening and during participation in the study.

18. Current or past abuse of alcohol, recreational or narcotic drugs, which in the investigator's opinion would compromise the subject's safety and/or compliance with the study procedures.

19. History of chronic urticaria (recurrent hives).

20. Unable to communicate reliably with the investigator.

21. Unlikely to adhere to the requirements of the study in the opinion of the investigator.

22. Employee of the investigator or study site, with direct involvement in the proposed study or other studies under the direction of that investigator or study site, as well as family members of the employees or the investigator. Site staff, or their family members, may be enrolled provided this is governed by institutional procedures and that there is no direct involvement in the proposed study or relationship with the investigator.

23. Under legal guardianship or incapacitation.

NOTE: Investigators should ensure that all study enrollment criteria have been met at screening. If a subject's clinical status changes (including any available laboratory results or receipt of additional medical records) after screening but before the prime vaccination on Day 1 such that he/she no longer meets all eligibility criteria, then the subject should be excluded from further participation in the study. Section 17.4 describes the required documentation to support meeting the enrollment criteria.

4.3. Prohibitions and Restrictions

Potential subjects must be willing and able to adhere to the following prohibitions and restrictions during the course of the study to be eligible for participation:

1. Woman of childbearing potential must remain on a highly effective method of birth control consistent with local regulations regarding the use of birth control methods for subjects participating in clinical studies (see inclusion criteria) until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer). If the social situation of a woman of childbearing potential changes (eg, woman who is not heterosexually active becomes active), she must begin a highly effective method of birth control, as described above in Section 4.1, until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer). *****

   Note: A period of 3 months after vaccination with Ad26.ZEBOV and 1 month after vaccination with MVA-BN-Filo should be respected.

   Note: Prior to each study vaccine administration, a urine pregnancy test should be performed for women of childbearing potential.

2. Woman must agree not to donate eggs (ova, oocytes) for the purposes of assisted reproduction from the start of screening onwards until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer). *****

3. Man who has not had a vasectomy performed more than 1 year prior to screening and is sexually active with a woman of childbearing potential must use condoms for sexual intercourse until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer), and must also not donate sperm from the start of screening onwards until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer). *****

   ***After unblinding for the primary analysis, placebo subjects may stop birth control measures or resume their pre-study birth control measures, and may donate eggs (women) or sperm (men).**

4. Woman should not breast-feed while enrolled in the study until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer).

5. Not travel to epidemic Ebola areas while enrolled in the study from the start of screening onwards until the 42-day post-boost visit. Subjects who traveled to these areas should have returned at least 1 month before the long-term follow-up visits (6 months post-boost and Day 365). Any traveling to epidemic Ebola areas should be documented in the case report form (CRF).

   Note: Subjects travelling to epidemic Ebola areas will be excluded from follow-up collection of blood for immunogenicity assessments if they contract Ebola virus disease (see also exclusion criterion #2 in Section 4.2.)

6. Not use any disallowed concomitant therapies as described in Section 8.

5. TREATMENT ALLOCATION AND BLINDING

Vaccine Schedule and Study Vaccine Allocation

Based on subject's availability and informed consent, a subject was enrolled in one of 3 cohorts at study entry. Central randomization was implemented in this study. At baseline, subjects were randomly assigned to 1 of 3 groups in a 1:1:1 ratio in Cohorts I, II, and III, and within groups randomly assigned to Ad26.ZEBOV and MVA-BN-Filo, or placebo in a 14:1 and 10:3 ratio in Cohorts II and III, respectively, based on a computer-generated randomization schedule prepared before the study by or under the supervision of the sponsor. The randomization within each group was balanced by using randomly permuted blocks and was stratified by country in Cohorts II and III. Randomization in each group (for Cohorts II and III) was stratified further according to age at randomization (≤50 years, >50 years), with at least 20% (of the total of both cohorts) of subjects in the >50 years category. Note that randomization to Group 3 was not resumed per Amendment 4 and that enrollment in the entire study will be stopped per Amendment 5. The interactive web response system (IWRS) assigned a unique code, which dictated the assignment and matching vaccination schedule for the subject. The requestor must use his or her own user identification and personal identification number when contacting the IWRS, and will then give the relevant subject details to uniquely identify the subject.

Blinding

Blinding procedures are not applicable for Cohort I (as all subjects will receive Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion).

In Cohorts II and III, subjects and study-site personnel will be blinded to the study vaccine allocation within groups until the last subject of the study completes the 6-month post-boost visit or discontinues earlier and the clinical database is locked, except for unblinded qualified study-site personnel with primary responsibility for study vaccine preparation and dispensing, and not involved in any other study-related procedure. The study vaccines will be administered by a study vaccine administrator (see Definitions of Terms).

In Cohorts II and III, sponsor personnel will be blinded to study vaccine allocation within groups until all subjects of the study have their 6-month post-boost visit or discontinue earlier and the clinical database is locked. At that time, the subjects enrolled in the study will be unblinded and the subjects who received placebo will be contacted by the site to communicate that they have completed the study and no further follow-up is required. However, subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit. After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will be followed up in an open-label fashion until the Day 365 visit. The primary analysis will be performed when all subjects in the study have completed the 6-month post-boost visit or discontinued earlier and the clinical database is locked. In case interim analyses will be performed, study-site personnel (except for those with primary responsibility for study vaccine preparation and dispensing), the sponsor (except for programming, statistics, clinical and clinical immunology personnel involved in the analysis, and the sponsor committee involved in making

future decisions for the program) and subjects will remain blinded to study vaccine allocation until the primary analysis (see below).

The investigator will not be provided with randomization codes. The codes will be maintained within the IWRS, which has the functionality to allow the investigator to break the blind for an individual subject (in case of a medical emergency requiring unblinding).

Data that may potentially unblind the study vaccine assignment (ie, antibodies to study vaccine, study vaccine preparation/accountability data, or other specific laboratory data) will be handled with special care to ensure that the integrity of the blind is maintained and the potential for bias is minimized. This can include making special provisions, such as segregating the data in question from view by the investigators, clinical team, or others as appropriate until the time of database lock and unblinding. The pharmacy and preparation of study vaccines will be monitored by an independent study vaccine monitor (see Definitions of Terms and Section 17.8).

Under normal circumstances, the blind should not be broken until the primary analysis. Otherwise, the blind should be broken only if specific emergency treatment/course of action would be dictated by knowing the treatment status of the subject. In such cases, the investigator may in an emergency determine the identity of the study vaccine by contacting the IWRS. It is recommended that the investigator contacts the sponsor or its designee if possible to discuss the particular situation, before breaking the blind. Telephone contact with the sponsor or its designee will be available 24 hours per day, 7 days per week. In the event the blind is broken, the sponsor must be informed as soon as possible. The date and reason for the unblinding must be documented by the IWRS, in the appropriate section of the CRF, and in the source document. The documentation received from the IWRS indicating the code break must be retained with the subject's source documents in a secure manner.

In general, randomization codes will be disclosed fully for the study only if all subjects in the study completed the 6-month post-boost visit (or discontinued earlier) and the clinical database is locked. However, if an interim analysis is specified, the randomization codes and, if required, the translation of randomization codes into treatment and control groups will be disclosed to those authorized and only for those subjects included in the interim analysis.

If the randomization code is broken by the investigator or the study-site personnel, the subject must discontinue study vaccine and must be followed as appropriate (see Section 10.2 for further details). If the randomization code is broken by the sponsor for safety reporting purposes, the subject should not discontinue study vaccine and may remain in the study (if the randomization code is still blinded to the study-site personnel and subject).

6. DOSAGE AND ADMINISTRATION

An overview of the study vaccination schedules is provided in Table 2.

6.1. General Instructions and Procedures

All subjects will receive a vaccination, according to randomization, on Day 1 (Groups 1 to 3) and on Day 29 (Group 1), Day 57 (Group 2) or Day 85 (Group 3) at the following dose levels:

- Ad26.ZEBOV: $5x10^{10}$ vp, supplied in a single use vial (0.5 mL extractable);
- MVA-BN-Filo: $1x10^{8}$ Inf.U (nominal titer); target fill $1.9x10^{8}$ Inf.U per dose (range 1.27 to 2.67 $x10^{8}$ Inf.U), supplied in a single use vial (0.5 mL extractable);
- Placebo: 0.9% saline, 0.5 mL extractable.

Ad26.ZEBOV and MVA-BN-Filo, or placebo, will be administered as 0.5-mL IM injections into the deltoid, by a study vaccine administrator (see Definitions of Terms). The injection site should be free from any injury, local skin problem, significant tattoo or other issue that might interfere with evaluating the arm after injection (eg, subjects with a history of skin cancer must not be vaccinated at the previous tumor site), per exclusion criterion #9 in Section 4.2. In each subject, the boost vaccination should be administered in the opposite deltoid from the prime vaccination (unless the opposite arm has a condition that prevents evaluating the arm after injection) and it should be recorded in the CRF in which arm the vaccination has been administered. No local or topical anesthetic will be used prior to the injection.

Discontinuation of study vaccine administration should occur in any subject meeting the criteria outlined in Section 10.2. Criteria for postponement of vaccination at the scheduled time for vaccine administration and contraindications to boost vaccination have been defined in Sections 6.2 and 6.3, respectively. Refer to Section 9.2.3 for details on the pre-specified pausing rules to halt vaccination of further subjects.

After each vaccination, subjects will remain at the site for a total of 60 (±15) minutes post-vaccination to monitor for the development of any acute reactions, or longer if deemed necessary by the investigator (eg, in case of grade 3 adverse events).

The Site Investigational Product Procedures Manual specifies the maximum time that will be allowed between preparation and administration of the study vaccine. For storage conditions, please refer to Section 14.3.

6.2. Criteria for Postponement of Vaccination

A subject will not be given the prime or boost vaccination if he/she experiences any of the following events at the scheduled time for vaccination:

- Acute illness at the time of vaccination (this does not include minor illnesses such as diarrhea or mild upper respiratory tract infection);
- Fever (body temperature ≥38.0°C) at the time of vaccination.

Subjects experiencing any of the events described above may be vaccinated up to 10 days beyond the window allowed for the scheduled vaccination, or be withdrawn from that vaccination at the discretion of the investigator and after consultation with the sponsor (see Section 10.2).

Note: In case the boost vaccination is postponed, the timing of the post-boost visits will be planned relative to the actual vaccination day (see Section 9.1.1).

6.3. Contraindications to Boost Vaccination

A subject will not be given the boost vaccination if he/she experiences any of the following events at any time after the prime vaccination:

1. Anaphylaxis clearly attributable to vaccination with the study vaccine; *OR*
2. Generalized urticaria within 72 hours of vaccination considered to be at least possibly related to the study vaccine; *OR*
3. A serious adverse event considered to be at least possibly related to the study vaccine; *OR*
4. A severe (grade 3) (non-serious) adverse event considered to be at least possibly related to the study vaccine that persists for 3 or more days, or a severe (grade 3) (non-serious) laboratory abnormality (including unexplained hematuria) considered to be at least possibly related to the study vaccine; *OR*
5. Injection site ulceration, abscess or necrosis considered to be at least possibly related to the study vaccine; *OR*
6. Any other safety concern threatening the subject's safety or persisting clinically significant abnormality considered to be related to prime vaccination.

Subjects experiencing any of the events described above must not receive any further study vaccine, but should be monitored for safety and for immunogenicity according to the protocol as described in Section 10.2.

An ad hoc IDMC meeting may be requested via the sponsor for any single event or combination of multiple events which are considered to jeopardize the safety of the subjects.

7. TREATMENT COMPLIANCE

All study vaccines will be administered on site by a study vaccine administrator (see Definitions of Terms). The date and time of each study vaccine administration will be recorded in the CRF.

8. PRESTUDY AND CONCOMITANT THERAPY

Prestudy therapies administered up to 30 days prior to the start of screening and previous vaccinia/smallpox vaccination at any time prior to study entry must be recorded in the CRF.

Concomitant therapies must be recorded from screening onwards until 42 days after the boost vaccination. Thereafter, concomitant therapies are only to be recorded if given in conjunction with serious adverse events and IREs that meet the criteria outlined in Sections 12.3.2 and 12.3.3, respectively.

All therapies (prescription or over-the-counter medications, including vaccines, vitamins, herbal supplements; non-pharmacologic therapies such as electrical stimulation, acupuncture, special diets, exercise regimens) must be recorded in the CRF. Recorded information will include a description of the type of the drug, treatment period, dosing regimen, route of administration, and its indication.

Subjects must use adequate birth control measures prior to randomization as described in Section 4.1.

Subjects are allowed to receive all routine immunizations according to local schedules, taking into consideration the following restrictions. These routine immunizations should be administered at least 15 days before (or at least 15 days after) administration of any study vaccine to avoid any potential interference in efficacy of the routine immunizations or the interpretation of immune responses to study vaccines, as well as to avoid potential confusion with regard to attribution of adverse events. Vaccination with live attenuated vaccines is prohibited in the period from 30 days before Day 1 (prime vaccination) to 30 days after the boost vaccination. However, if a vaccine is indicated in a post-exposure setting (eg, rabies or tetanus), it must take priority over the study vaccine.

Analgesic/antipyretic medications and non-steroidal anti-inflammatory drugs (NSAIDs) may be used post-vaccination only in case of medical need (eg, body temperature ≥38.5°C or pain) and their use must be documented. Use of these medications as routine prophylaxis prior to study vaccine administration is prohibited. Chronic or recurrent use of medications that modify the host immune response (eg, cancer chemotherapeutic agents, systemic corticosteroids, immunomodulators) are prohibited.

The sponsor must be notified in advance (or as soon as possible thereafter) of any instances in which prohibited therapies are administered. Prohibited therapies will be captured as protocol deviations.

9. STUDY EVALUATIONS

9.1. Study Procedures

9.1.1. Overview

The Time and Events Schedule and Additional Time and Events Schedule summarize the frequency and timing of safety, tolerability and immunogenicity measurements and evaluations applicable to this study. Details for all study procedures are provided in the following sections. Additional unscheduled study visits may be required if in the investigator's opinion, further clinical or laboratory evaluation is needed.

In summary, safety will be evaluated in all subjects from the 3 cohorts. In Cohort I (in the UK), only plasmablast response kinetics (and transcriptomics in subjects who consented separately) will be evaluated in addition to the safety assessments. In Cohorts II and III in both countries, core immunogenicity assessments (humoral and cellular assays) will be performed. In Cohort II in both countries, additional immunogenicity assessments will be done.

Visit Windows

The screening visit had to be performed within 12 weeks prior to the baseline visit (ie, the day of the subject's prime vaccination, Day 1). Visit windows that will be allowed are summarized in Table 3. The subject should be encouraged to come in the exact day planned and use the visit window only if absolutely necessary. If a subject did not receive study vaccine on the planned day of vaccination, the timings of the next visits post vaccination (see Time and Events Schedule and Additional Time and Events Schedule) will be determined relative to the actual day of vaccination.

Table 3: Visit Windows

Visit Description	Group 1	Group 2	Group 3	Allowed Window
Prime Vaccination	***Day 1***	***Day 1***	***Day 1***	**-**
One Day Post-Prime – Cohort II only	Day 2	Day 2	Day 2	±1 day
Three Days Post-Prime	Day 4	Day 4	Day 4	±1 day
Seven Days Post-Prime	Day 8	Day 8	Day 8	±1 day
Eight Days Post-Prime – Cohort I only	Day 9	Day 9	Day 9	±1 day
Ten Days Post Prime – Cohort I only	Day 11	Day 11	Day 11	±1 day
Twelve Days Post-Prime – Cohort I only	Day 13	Day 13	Day 13	±1 day
Fourteen Days Post-Prime – Cohorts I and II and Group 3 in Cohort III only	Day 15	Day 15	Day 15	±1 day
To be determined[a] – Cohort II only	Day *x*	Day *x*	Day *x*	±1 day
Boost Vaccination	***Day 29***	***Day 57***	***Day 85***	***±1 day***
One Day Post-Boost – Cohort II only	Day 30	Day 58	Day 86	±1 day
Three Days Post-Boost	Day 32	Day 60	Day 88	±1 day
Five Days Post-Boost – Cohort I only	Day 34	Day 62	Day 90	±1 day
Seven Days Post-Boost	Day 36	Day 64	Day 92	±1 day
Nine Days Post-Boost – Cohort I only	Day 38	Day 66	Day 94	±1 day
Fourteen Days Post-Boost – Cohort II only	Day 43	Day 71	Day 99	±1 day
To be determined[a] – Cohort II only	Day *x*	Day *x*	Day *x*	±1 day
Twenty-one Days Post-Boost	Day 50	Day 78	Day 106	±3 days
Forty-two Days Post-Boost[b]	Day 71	Day 99	Day 127	±3 days
Follow-up (6 months post-boost)[c,d]	Day 209	Day 237	Day 265	±30 days
Follow-up (1 year post-prime)[c,e]	Day 365	Day 365	Day 365	±30 days

Note: If a subject did not receive study vaccine on the planned day of vaccination, the timings of the next visits post-vaccination will be determined relative to the actual day of vaccination.

[a] At plasmablast peak time point determined in Cohort I (only applies to Cohort II in the UK).

[b] The 42-days post-boost visit may be replaced by a telephone call if the subject is not able to come to the site (only applies to Cohort I in the UK and Cohort II in the UK and France).

[c] The 6-month post-boost and Day 365 visits may be replaced by a telephone call if the subject is not able to come to the site (only applies to Cohort I in the UK).

[d] Subjects who already attended the 6-month post-prime visit (which has been replaced with the 6-month post-boost visit per protocol amendment 3) will be required to also attend the 6-month post-boost visit.

[e] Subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study VAC52150EBL4001 is opened, will have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months (whichever comes first) or discontinued earlier. These subjects will be contacted by telephone every 3 months after Day 365 (subjects will only visit the site when clinically indicated).

Blood Sampling Volumes

For subjects with core as well as exploratory immunogenicity assessments, less than 500 mL of blood will be drawn over each 60-day interval throughout the study, which is within the limits of standard blood donation.

Additional serum or urine pregnancy tests may be performed, as determined necessary by the investigator or required by local regulation, to establish the absence of pregnancy at any time during the subject's participation in the study.

Repeat or unscheduled samples may be taken for safety reasons or for technical issues with the samples.

9.1.2. Screening Phase

Up to 12 weeks before baseline (the day of the subject's prime vaccination, Day 1) and after signing and dating the ICF (see Section 16.2.3), screening assessments as indicated in the Time and Events Schedule were performed. Screening could be split into multiple days or visits. In exceptional cases, the screening phase could be extended if discussed with and approved (documented) by the sponsor, eg, if not all the test results became available during the allocated 12 weeks; this was evaluated on a case-by-case basis. Subjects whose screening period was longer than the protocol-defined 12 weeks as a result of a pause were allowed to rescreen once.

For men and for women of non-childbearing potential, there was no minimum duration of the screening phase and it lasted only for the time required to verify eligibility criteria. For women of childbearing potential, it should be confirmed that adequate birth control measures were used from at least 28 days before the prime vaccination with a negative serum β-hCG pregnancy test at screening and a negative urine pregnancy test immediately prior to each study vaccination (see Sections 4.1 and 4.2). All men and women, except for women of non-childbearing potential, will be asked to use adequate birth control for sexual intercourse until at least 3 months after the prime vaccination or up to 1 month after the boost vaccination (whichever takes longer, see Section 4.3).

Only healthy subjects complying with the criteria specified in Section 4 were included in the study. The investigator provided detailed information on the study to the subject and obtained written informed consent prior to each subject's study participation. The procedures indicated in the Time and Events Schedule and Additional Time and Events Schedule were only performed after the subject's written informed consent had been obtained.

The following was performed to determine eligibility requirements as specified in the inclusion and exclusion criteria:

- Review of all inclusion and exclusion criteria;
- Review of medical history (including concomitant diseases) and demographics;
- Review of prestudy therapies (up to 30 days prior to the start of screening), previous vaccinia/smallpox vaccination (at any time prior to study entry), and concomitant therapies;
- Serum pregnancy test (for women of childbearing potential);
- Blood sampling for hematology and chemistry (fasting or non-fasting) and troponin I;
- Urine sampling for urinalysis (dipstick);
- Serology testing (HIV type 1 or type 2, hepatitis B, hepatitis C);
- FSH assessment (for women >45 years of age with amenorrhea for less than 2 years or ≤45 years of age with amenorrhea for more than 6 months);

- Full physical examination (including height and body weight);
- ECG recording;
- Measurement of vital signs (blood pressure, pulse/heart rate, body temperature).

All adverse events and pregnancies will be collected from the time a signed and dated ICF is obtained.

The overall eligibility of the subject to participate in the study was assessed once all screening values and results of any other required evaluations were available. Retesting of values (eg, safety laboratory) that lead to exclusion was allowed once using an unscheduled visit during screening to assess eligibility, also for subjects who were rescreened due to a pause. The safety laboratory assessments (ie, serum chemistry and hematology, troponin I testing, serology, pregnancy testing, urinalysis) were to be performed within 28 days prior to the prime vaccination and might be repeated if they fall outside this time window. Similarly, subjects who were rescreened due to a pause had to have new safety laboratory assessments within 28 days of the prime vaccination. Study subjects who qualified for inclusion were contacted and scheduled for enrollment and prime vaccination within 12 weeks.

Note that enrollment will be stopped per Amendment 5.

9.1.3. Vaccination Phase

Prime Vaccination - Day 1 (All Groups)

If eligible, the subject will come for the baseline visit (Day 1). The investigator should ensure that all enrollment criteria have been met during screening. If the initial laboratory sampling occurred more than 12 weeks before baseline (Day 1), sampling will need to be repeated. If a subject's clinical status changes (including available laboratory results or receipt of additional medical records) after screening but before the prime vaccination (Day 1) such that the subject no longer meets all eligibility criteria, then the subject should be excluded from further participation in the study.

A urine pregnancy test (for women of childbearing potential), vital signs measurements, blood sampling for hematology and chemistry (fasting or non-fasting), and an abbreviated, symptom-directed physical examination (as indicated by the investigator) will be performed. The eligible subjects will be allocated (by central randomization) to a vaccination schedule as described in Section 5 and will receive the prime vaccination via IM injection as described in Section 6, unless any of the pre-specified criteria not to proceed with vaccination are met (see Sections 6.2 and 10.2 for details) or if a pause for vaccination of further subjects has been installed (see Section 9.2.3).

Blood samples for core immunogenicity assessments in Cohorts II and III will be collected before the prime vaccination as indicated in the Time and Events Schedule and Section 9.3.2. Blood samples for additional immunogenicity assessments in Cohort II and for specific immunogenicity assessments in Cohort I will be collected as indicated in the Additional Time and Events Schedule and Section 9.3.2.

Study vaccine will be prepared on-site by unblinded qualified study-site personnel not involved in any other study-related procedure who will place a blinding tape on the syringe to mask its content and send the study vaccine to a study vaccine administrator (see Definitions of Terms) for administration to the subject (see Section 14.3 for details). Refer to Section 6 for further details on dosage and administration and post-vaccination monitoring.

All adverse events, serious adverse events and pregnancies will be collected and documented on the CRF, together with the information on any concomitant therapies. For reporting of IREs, refer to Section 12.3.3.

Upon discharge from the site, subjects will be provided with a diary, a thermometer, and a ruler to measure and record solicited local adverse events and body temperature. Subjects will also record symptoms of solicited local and systemic adverse events in the diary in the evening after vaccination and then daily for the next 7 days. Subjects will be instructed to notify the investigator immediately for any unsolicited adverse event (not listed on the diary card) or for any severe (grade 3) solicited adverse event (listed on the diary card).

Boost Vaccination - Day 29 (Group 1), Day 57 (Group 2), Day 85 (Group 3)

The subjects will receive the boost vaccination via IM injection as described in Section 6, unless any of the pre-specified criteria not to proceed with vaccination are met (see Sections 6.2, 6.3, and 10.2 for details) or if a pause for vaccination of further subjects has been installed (see Section 9.2.3).

Before the boost vaccination, a urine pregnancy test (women of childbearing potential), ECG and vital sign measurements, blood sampling for hematology and chemistry (fasting or non-fasting), and an abbreviated, symptom-directed physical examination (as indicated by the investigator) will be performed.

Blood samples for core immunogenicity assessments in Cohorts II and III will be collected before the boost vaccination as indicated in the Time and Events Schedule and Section 9.3.2. Blood samples for additional immunogenicity assessments in Cohort II and for specific immunogenicity assessments in Cohort I will be collected as indicated in the Additional Time and Events Schedule and Section 9.3.2.

The same procedures are applicable as for the prime vaccination regarding vaccine preparation; collection of adverse events, serious adverse events, IREs and pregnancies, together with the information on concomitant therapies; and procedures for subjects after discharging.

9.1.4. Post-vaccination Phase

The subjects will come to the site at 3 and 7 days after each vaccination and also at 21 and 42 days after the boost vaccination as indicated in the Time and Events Schedule. In Cohorts I and II, the subjects will come to the site for additional post-vaccination visits as indicated in the Additional Time and Events Schedule. The 42-days post-boost visit (Cohorts I and II) may be replaced by a telephone call if the subject is not able to come to the site.

The subject's diary will be reviewed by study-site personnel at 3 and 7 days after each vaccination. The investigator will examine the injection site for occurrences of erythema, induration/swelling, and pain/tenderness, and will ask if itching at the injection site occurred at these visits in order to complete the relevant parts of the CRF.

Blood sampling for hematology and chemistry (fasting or non-fasting) at 7 days after each vaccination and an abbreviated, symptom-directed physical examination (as indicated by the investigator) will be performed. An ECG will be recorded at 7 days after the boost vaccination. Adverse events, serious adverse events and pregnancies will be collected and documented on the CRF, together with the information on any concomitant therapies. For reporting of IREs, refer to Section 12.3.3.

Blood samples for core immunogenicity assessments in Cohorts II and III will be collected as indicated in the Time and Events Schedule and Section 9.3.2. Blood samples for additional immunogenicity assessments in Cohort II and for specific immunogenicity assessments in Cohort I will be collected as indicated in the Additional Time and Events Schedule and Section 9.3.2.

Subjects will be instructed to contact the investigator before the next visit (ie, 6 months post-boost) if they experience any adverse event or intercurrent illness that they perceive as relevant and/or can be possibly related to study vaccine in their opinion.

9.1.5. Open-label Long-term Follow-up Phase

When all subjects have had their 6-month post-boost visit (Day 209 in Group 1; Day 237 in Group 2; Day 265 in Group 3) or discontinued earlier and the clinical database is locked, the subjects will be unblinded. After unblinding, only subjects who received Ad26.ZEBOV or MVA-BN-Filo will be followed up in an open-label fashion until the Day 365 visit. Subjects who received placebo will be contacted by the site to communicate that they have completed the study and no further follow-up is required. However, subjects who received placebo and reach the Day 365 visit prior to unblinding will be required to attend the Day 365 visit. The 6-month post-boost and Day 365 visits (Cohort I) may be replaced by a telephone call if the subject is not able to come to the site.

Serious adverse event information and IREs will be collected until the end of the study, and concomitant therapies should only be recorded if given in conjunction with serious adverse events and IREs. Pregnancies will be reported until the end of the study.

Blood samples for core immunogenicity assessments in Cohorts II and III will be collected at both visits as indicated in the Time and Events Schedule and Section 9.3.2. In Cohort II, follow-up blood samples will be collected for additional immunogenicity assessments on the 6-month post-boost visit as indicated in the Additional Time and Events Schedule and Section 9.3.2.

Subjects in France who agree to continue the long-term follow-up after Day 365 and reach the Day 365 visit before the roll-over study VAC52150EBL4001 (see Section 9.1.6) is opened, will

have further follow-ups in the current study until they have started in the roll-over study or for an additional 12 months (whichever comes first) or discontinued earlier. These subjects will be contacted by telephone every 3 months after Day 365 (subjects will only visit the site when clinically indicated) for the collection of information on adverse events, serious adverse events, and IREs that were ongoing at the Day 365 visit, and to report and follow up on new serious adverse events, pregnancies, and IREs.

9.1.6. VAC52150 Vaccine Development Roll-over Study

Subjects in the UK and France who reach the Day 365 visit prior to unblinding will be approached to consent for enrollment into the VAC52150 Vaccine Development Roll-over study for long-term surveillance (for a total of up to 60 months after the prime vaccination). After unblinding, only subjects enrolled in this study who received Ad26.ZEBOV and/or MVA-BN-Filo will remain in the VAC52150 Vaccine Development Roll-over study for long-term safety surveillance. After unblinding, subjects who received placebo and have already been enrolled into the VAC52150 Vaccine Development Roll-over study will be discontinued from further participation in the roll-over study. The parent(s)/legal guardian of children born to vaccinated female subjects who became pregnant with estimated conception within 28 days after vaccination with MVA-BN-Filo or within 3 months after vaccination with Ad26.ZEBOV, will also be approached to consent for enrollment of their offspring into the roll-over study, according to the same rules that apply for the other subjects.

9.2. Safety Evaluations

9.2.1. Safety Endpoints

Primary Endpoints:

- Adverse events, collected from signing of the ICF onwards until the 42-day post-boost visit (Day 71 in Group 1; Day 99 in Group 2; and Day 127 in Group 3).
- Serious adverse events and IREs, collected from signing of the ICF onwards until the end of the study.
- Solicited local and systemic adverse events (reactogenicity) until 7 days after each study vaccine administration.

9.2.2. Safety Assessments

The study will include the following evaluations of safety and tolerability as described below and according to the time points provided in the Time and Events Schedule. Any clinically significant abnormalities occurring from signing of the ICF onwards until 42 days after the boost vaccination must be recorded on the Adverse Event section of the CRF. Thereafter, reporting will be limited to all serious adverse events. Any clinically significant abnormalities (including those persisting at the end of the study/early withdrawal) will be followed by the investigator until resolution or until a clinically stable endpoint is reached (see Section 12). For reporting of IREs, refer to Section 12.3.3. Refer to Section 9.1.5 for more information for subjects in France who have further follow-ups from Day 365 onwards until the roll-over study is opened.

The investigators, together with the sponsor's medical monitor, will be responsible for the safety monitoring of the study, and will halt vaccination of further subjects in case any of the pre-specified pausing rules described in Section 9.2.3 have been met.

An IDMC will be appointed by the sponsor with recommendations from the Clinical Steering Committee (see Section 11.8) before the start of the study to perform regular review of the safety data during the study. Details regarding the IDMC are provided in Section 11.7.

Adverse Events

All adverse events, whether serious or non-serious, will be reported by the subject from signing of the ICF onwards until the 42-day post-boost visit. Thereafter, reporting will be limited to all serious adverse events and IREs up to the subject's last study-related procedure. Solicited local and systemic adverse events (reactogenicity, see below) will be reported by the subject until 7 days after each study vaccine administration. Adverse events will be followed by the investigator as specified in Section 12. Refer to Section 9.1.5 for more information for subjects in France who have further follow-ups from Day 365 onwards until the roll-over study is opened.

Solicited Adverse Events

Solicited adverse events (see Definitions of Terms) are precisely defined events that subjects are specifically asked about and which are noted by subjects in the diary. The subjects will be closely observed by study-site personnel for the first 30 (±10) minutes after each study vaccine administration and again at 60 (±15) minutes post-vaccination, and any unsolicited, solicited local or systemic adverse events will be documented during this period. Upon discharge from the site, subjects will receive a diary, a thermometer, and a ruler to measure solicited local reactions and body temperature. Subjects will record symptoms of solicited local and systemic adverse events in the diary in the evening after each study vaccine administration and then daily for the next 7 days (until Day 8) to serve as a reminder to the subject for the next visit. On Day 8, the diary needs to be completed on site before the subject leaves the site. The investigator should discuss the information from the diary with the subject and document the relevant information in the clinic chart.

On-site and diary reported solicited adverse events will be captured on a separate CRF page as described in the CRF Completion Guidelines, in contrast to the unsolicited adverse events which will be reported on the Adverse Event page of the CRF. The investigator must record in the CRF his/her opinion concerning the relationship of the adverse event to study vaccine.

Solicited Local (Injection Site) Adverse Events

Subjects will also be instructed on how to note occurrences of erythema, induration/swelling (measured using the ruler supplied), pain/tenderness and itching at the injection site in the evening after each study vaccine administration and then daily for the next 7 days in the diary.

Solicited Systemic Adverse Events

Subjects will be instructed on how to record daily temperature using a thermometer provided for home use. Subjects should record the body temperature in the evening after each study vaccine

administration and then daily for the next 7 days in the diary. Temperature should be measured at approximately the same time each day. If more than one measurement is made on any given day, the highest value will be recorded in the CRF.

Subjects will also be instructed on how to note the following symptoms in the evening after each study vaccine administration and then daily for the next 7 days in the diary:

- nausea/vomiting
- headache
- myalgia
- arthralgia
- fatigue/malaise
- chills
- fever

If a ***solicited local or systemic adverse event*** is not resolved on Day 8, the follow-up will be captured in the diary. The subject will be instructed to record the date of last symptoms and maximum severity in the diary after resolution.

Cardiac Events

In case any cardiac sign or symptom develops after the boost vaccination, an ECG and troponin I test should be obtained and the subject should be referred to a local cardiologist.

Clinical Laboratory Tests

Blood samples for serum chemistry and hematology and for troponin I, serology and pregnancy testing (~15 mL) and a urine sample for urinalysis (dipstick) will be collected at screening. Blood samples for serum chemistry and hematology (~7 mL) will also be taken at additional time points as indicated in the Time and Events Schedule. Samples can be taken in fasting or non-fasting conditions but should be documented in the CRF. The investigator must review the laboratory results, document this review and record any clinically relevant changes occurring during the study in the Adverse Event section of the CRF up to the 42-day post-boost visit. Thereafter, only serious adverse events need to be recorded. The laboratory reports must be filed with the source documents. For reporting of IREs, refer to Section 12.3.3.

The following tests will be performed by the central laboratory. Parameters marked with an asterisk (*) will only be measured at screening:

- Hematology and Coagulation Panel

  - hemoglobin
  - white blood cell (WBC) count with differential
  - platelet count
  - fibrinogen*
  - prothrombin time
  - activated partial thromboplastin time

  Blood smear: A WBC evaluation may include any abnormal cells, which will then be reported by the laboratory. A hematology evaluation may include abnormalities in the red blood cell (RBC) count and/or RBC parameters and/or RBC morphology, which will then be reported by the laboratory. In addition, any other abnormal cells in a blood smear will also be reported. Only clinically significant abnormal WBC, abnormal RBC, or any other abnormal cells in a blood smear will be reported as adverse events.

- Serum Chemistry Panel

  - sodium
  - potassium
  - blood urea nitrogen (BUN)
  - aspartate aminotransferase (AST)
  - alanine aminotransferase (ALT)
  - glucose (fasting or non-fasting)*
  - albumin*
  - total protein*
  - total bilirubin*
  - creatinine
  - FSH*
    (only in women >45 years of age with amenorrhea <2 years or ≤45 years of age with amenorrhea >6 months)

- Urinalysis – Dipstick for:

  - glucose*
  - ketones*
  - protein*
  - blood*

  In case of positive urinalysis dipstick results, the sediment will be examined microscopically (only RBC will be documented).

Additional clinical laboratory assessments to be performed by the central laboratory (unless otherwise specified) are as follows:

- Serum pregnancy test for women of childbearing potential at screening;
- Urine pregnancy test for women of childbearing potential before each study vaccination (by the **local** laboratory);
- Serology (HIV type 1 and type 2 antibody, HBsAg, and HCV antibody) at screening;
- Troponin I testing at screening.

Laboratory values will be determined according to the toxicity grading tables in Attachment 1 if applicable for laboratory tests.

Any clinically significant abnormalities occurring from signing of the ICF onwards until 42 days after the boost vaccination must be recorded on the Adverse Event section of the CRF. Thereafter, reporting will be limited to all serious adverse events. All events should be followed (by repeat testing) to resolution, or until reaching a clinically stable endpoint. For reporting of IREs, refer to Section 12.3.3.

Electrocardiogram

A single, 12-lead ECG will be performed at the time points indicated in the Time and Events Schedule and interpreted locally. Additional ECG monitoring may be done at other time points during the study if clinically indicated based on signs and symptoms.

During the collection of ECGs, subjects should be in a quiet setting without distractions (eg, television, cell phones). Subjects should rest in a supine position for at least 5 minutes before ECG collection and should refrain from talking or moving arms or legs. If blood sampling or vital sign measurement is scheduled for the same time point as ECG recording, the procedures should be performed in the following order: vital signs, ECG(s), blood draw.

Any clinically significant abnormalities occurring from signing of the ICF onwards until 42 days after the boost vaccination must be recorded on the Adverse Event section of the CRF. Thereafter, reporting will be limited to all serious adverse events. All events should be followed to resolution, or until reaching a clinically stable endpoint. For reporting of IREs, refer to Section 12.3.3.

Vital Signs (blood pressure, pulse/heart rate and body temperature)

Blood pressure and pulse/heart rate measurements will be assessed (at rest) with a completely automated device. Manual techniques will be used only if an automated device is not available.

Body temperature will be assessed at screening and prior to study vaccine administration at the site. In addition, subjects will measure and record body temperature at home in the diary (see Solicited Systemic Adverse Events).

Any clinically significant abnormalities occurring from signing of the ICF onwards until 42 days after the boost vaccination must be recorded on the Adverse Event section of the CRF. Thereafter, reporting will be limited to all serious adverse events. All events should be followed to resolution, or until reaching a clinically stable endpoint. For reporting of IREs, refer to Section 12.3.3.

Physical Examination

A full physical examination, including height and body weight, will be performed by the investigator at screening. At other visits, an abbreviated, symptom-directed physical examination will be performed as indicated by the investigator based on any clinically relevant issues, clinically relevant symptoms and medical history. The symptom-directed physical examination may be repeated if deemed necessary by the investigator.

A full physical examination includes the following: general appearance, eyes, ears, nose, throat, cardiovascular system, respiratory system, gastrointestinal system, and skin and mucous membranes. A neurological and musculoskeletal examination as well as an examination of the lymph nodes will also be performed. An abbreviated, symptom-directed physical examination includes inspection of the vaccination site(s).

The height should be measured barefooted at the screening visit. To obtain the actual body weight, subjects must be weighed lightly clothed.

Any clinically significant abnormalities occurring from signing of the ICF onwards until 42 days after the boost vaccination must be recorded on the Adverse Event section of the CRF. Thereafter, reporting will be limited to all serious adverse events. All events should be followed to resolution, or until reaching a clinically stable endpoint. For reporting of IREs, refer to Section 12.3.3.

9.2.3. Pausing Rules

The investigators and the sponsor's medical monitor will review the safety of enrolled subjects on an ongoing basis. The sponsor's medical monitor will be involved in all discussions and decisions (see Section 12.4).

If any of the following events occur in any subject who received at least one dose of study vaccine in the study (at any site), that site investigator will halt the vaccination of further subjects and the sponsor's medical monitor will be notified immediately. The sponsor's medical monitor will then also inform all the other investigators to halt further vaccination as well.

1. Death in any subject considered at least possibly related to the study vaccine; *OR*

2. An anaphylactic reaction within 24 hours of vaccination or the presence of generalized urticaria within 72 hours of vaccination in any subject considered at least possibly related to the study vaccine; *OR*

3. A life-threatening or other serious adverse event in any subject considered at least possibly related to the study vaccine.

If any of the following events occur in subjects who received at least one dose of study vaccine in the study (across all sites), the sponsor's medical monitor will notify all investigators to halt vaccination of further subjects.

4. Three or more subjects experience a severe (grade 3) (non-serious) unsolicited adverse event (of the same type) considered to be related to any of the study vaccines that persists for 3 or more days; *OR*

5. Three or more subjects experience a persistent (upon repeat testing) severe (grade 3) (non-serious) abnormality (including unexplained hematuria) related to the same laboratory parameter and considered to be related to any of the study vaccines; *OR*

6. Three or more subjects experience the same severe (grade 3) (non-serious) solicited systemic reaction considered to be related to any of the study vaccines that persists for 3 or more days (subjective systemic reaction corroborated by study personnel).

For the events described above, the sponsor's medical monitor notifies the IDMC immediately. Dosing will be halted and health authorities will be notified. Within 3 business days, the IDMC will convene to review the available safety data as outlined in the charter and to discuss study suspension or discontinuation of further vaccination or to decide that vaccination may resume. If any pausing rule is met, and following the IDMC review it is deemed appropriate to restart dosing, a request to restart dosing with pertinent data must be submitted to health authorities as a request for a substantial amendment. Dosing can restart only after approval of a substantial amendment by the competent authority. The sites will be allowed to resume activities upon receipt of a written notification from the sponsor. The criteria for pausing will be re-set each time and the same criteria have to be met again to halt further vaccination. The communications from

the IDMC will be forwarded by the investigator to the Independent Ethics Committee (IEC)/Institutional Review Board (IRB) according to local standards and regulations and by the sponsor to relevant health authorities.

9.3. Immunogenicity Evaluations

9.3.1. Immunogenicity Endpoints

Secondary Endpoints:

Humoral immune response

- Binding antibody levels elicited by vaccination using EBOV GP ELISA at 21 days post boost.

Exploratory Endpoints:

Exploratory analyses may be performed to further investigate study vaccine-elicited immune responses. These may include, but might not be limited to, the following assays at selected time points:

Humoral immune response

- Binding antibody levels elicited by vaccination using EBOV GP ELISA at other relevant time points.
- Neutralizing antibody responses against the EBOV GP defined as the serum titer that is able to inhibit viral infection by a certain percentage ($IC_{50,}$ IC_{80} and/or IC_{90}) using virus neutralization assay.
- Binding and/or neutralizing antibody responses against Ad26 and/or MVA vector in ELISA and/or neutralization assays and against EBOV isolate in a neutralization test, if assays are available.
- Humoral immune responses to different EBOV GPs and/or filovirus GPs and/or TAFV NP, if assays are available.
- Molecular and functional characterization of study vaccine-elicited antibodies, which may include, but will not be limited to: kinetic studies, repertoire analysis, Fc characterization, isotype analysis and epitope mapping, if assays are available.

Cellular immune response

- The presence and functional capacity of T cells may be determined using pathogen-specific stimulation of PBMC with EBOV GP peptides using IFN-γ ELISpot assays. Cytokine-producing T cells can be quantified using ELISpot technology.
- Activation of CD4+ and CD8+ T cell subsets and their cytokine expression patterns may be determined by flow cytometry (intracellular cytokine staining [ICS]) after EBOV GP-specific stimulation of fresh and/or frozen PBMC (including, but not limited to, IFN-γ, interleukin [IL]-2, and tumor necrosis factor [TNF]-α).

- Cellular responses to filovirus GPs and/or TAFV NP, if assays are available.
- Phenotypic and functional analysis of B, T and/or natural killer (NK) cells and other immune cell populations.

9.3.2. Immunogenicity Assessments

Details on sample collection and handling are provided in Section 9.5. In case of insufficient blood sample volume to perform assays, the samples will be analyzed according to the following priority ranking (ranked in order of decreasing priority): 1) core secondary endpoints, 2) core exploratory endpoints and 3) additional exploratory endpoints.

Core Immunogenicity Assessments in Cohorts II and III

Table 4 provides an overview of the core immunogenicity assessments (humoral and cellular assays) in Cohorts II and III. Blood samples for core immunogenicity assessments for the evaluation of secondary and exploratory endpoints as planned by the sponsor will be collected at the time points indicated in the Time and Events Schedule.

Blood samples (10 mL) for assessment of humoral immune responses will be obtained from all subjects in Cohorts II and III. Blood samples (40 mL) for assessment of cellular immune responses will be obtained from subjects in Cohorts II and III at selected sites with the capabilities to process PBMC (targeted at 10% of all subjects).

Table 4: Overview of Core Immunogenicity Assessments in Cohorts II and III

Sample	Core Immunogenicity Assessments (non-exhaustive)	Assays (non-exhaustive)
Serum, all subjects from Cohorts II and III	Analysis of antibodies binding to EBOV GPs Analysis of neutralizing antibodies to EBOV GPs Analysis of binding and/or neutralizing antibodies to adenovirus and/or MVA Analysis of EBOV GP, Filovirus GPs and/or TAFV NP antibody characteristics, including IgG subtyping	ELISA Virus neutralization assay Adenovirus and/or MVA ELISA and/or neutralization assay Molecular antibody characterization
PBMC, subjects from Cohorts II and III at selected sites[a]	T-cell IFN-γ responses to EBOV GP Analysis of T-cell responses to EBOV GP, Filovirus GPs and/or TAFV NP (including CD4/8, IL-2, IFN-γ, TNF-α and/or activation markers)	ELISpot ICS

ELISA: enzyme-linked immunosorbent assay; ELISpot: enzyme-linked immunospot; ICS: intracellular cytokine staining; IFN: interferon; IgG: immunoglobulin G; IL: interleukin; TNF: tumor necrosis factor
[a] With the capabilities to process PBMC (targeted at 10% of all subjects).

Specific Immunogenicity Assessments in Cohort I and Additional Immunogenicity Assessments in Cohort II

Table 5 provides an overview of the specific immunogenicity assessments in Cohort I. Blood samples for plasmablast response kinetics will be collected in Cohort I at the time points indicated in the Additional Time and Events Schedule for the determination of the optimal sampling time points of the B-cell response as part of the additional immunogenicity assessments in Cohort II in the UK.

Table 5: Overview of Specific Immunogenicity Assessments by Academic Consortium Partners in Cohort I

Academic Consortium Partner/Laboratory	Specific Immunogenicity Assessments	Max. Blood Volume per Sampling Point	No. of Subjects Targeted per Group
University of Oxford	Kinetics of B-cell response (B-cell ELISpot)	10 mL, PBMC	10
	Transcriptomics[a]	3 mL, Tempus™ tube	All[a]

[a] Optional blood samples (for transcriptomics) only to be obtained from subjects who consent separately.

Table 6 provides an overview of the additional immunogenicity assessments in Cohort II. Blood samples for additional immunogenicity assessments for the evaluation of additional exploratory endpoints as planned by the academic consortium partners will be collected from subjects in Cohort II at the time points indicated in the Additional Time and Events Schedule. Blood samples will be collected for DNA isolation (in the UK), transcriptomics (in both countries), and EBOV neutralization (in France) from all subjects in Cohort II. For the other assays listed in Table 6, blood samples will be collected from all subjects in Cohort II in the UK, but initially only 25 subjects per group will be targeted for analysis, while in France, blood samples will be collected from and analyzed in no more than 25 subjects in Groups 1 and 2 in Cohort II at selected sites. In addition, these 'other' assays have to be performed within the same 25 subjects per group in the UK and in France. PBMC and serum samples for exploratory immunogenicity assessements collected in France may be analyzed in the UK and vice versa.

Table 6: Overview of Additional Immunogenicity Assessments by Academic Consortium Partners in Cohort II

Academic Consortium Partner/Laboratory[a]	Potential Additional Immunogenicity Assessments	Max. Blood Volume per Sampling Point	No. of Subjects Targeted per Group
University of Oxford	DNA isolation[b]	2 mL, whole blood	All[b]
	Transcriptomics[c]	3 mL, Tempus™ tube	All[c]
	Repertoire sequencing Isolation of monoclonal Ab Plasmablast phenotyping	25 mL (10 mL pre-prime), PBMC	25[d]
	Memory B-cell characterization	25 mL, PBMC	
	Fresh ICS	10 mL, whole blood/PBMC	
INSERM	EBOV neutralization	3 mL, serum	All
	Transcriptomics[c]	3 mL, Tempus™ tube	All[c]
	Fresh ICS	10 mL, whole blood/PBMC	25[e,f]
	T cell proliferation	10 mL, PBMC	
	T cell phenotyping (including TfH)	4 mL, PBMC	
	Cytokine profiling	3 mL, serum	
London School of Hygiene and Tropical Medicine[f]	NK cell analysis[f]	4 mL, PBMC and 2 mL, serum[f]	

Ab: antibody; No.: number; TfH: T follicular helper

[a] PBMC and serum samples for exploratory immunogenicity assessments collected in France may be analyzed in the UK and vice versa.

[b] Optional blood sample (for DNA) only to be obtained from subjects who consent separately.

[c] Optional blood samples (for transcriptomics) only to be obtained from subjects who consent separately.

[d] Blood samples to be collected from all subjects in Cohort II in the UK, but initially only 25 subjects per group will be targeted for analysis.

[e] Blood samples to be collected from and analyzed in no more than 25 subjects in Groups 1 and 2 in Cohort II at selected sites in France.

[f] Blood sampling for NK cell analysis done in France, but analysis performed at London School of Hygiene and Tropical Medicine.

Buffy coats will be obtained from the National Blood Transfusion Service (UK) for use of the PBMC in the validation and control of the B-cell ELISpot and memory B-cell characterization.

9.4. Vaccine-induced Seropositivity

In general, uninfected subjects who participate in Ebola vaccine studies may develop Ebola-specific antibodies as a result of an immune response to the candidate Ebola vaccine, referred to as VISP. These antibodies may be detected in Ebola serologic tests, causing the test to appear positive even in the absence of actual Ebola infection. VISP may become evident during the study, or after the study has been completed. The potential of a study participant becoming PCR-positive after vaccination is being assessed in a Phase 1 study (VAC52150EBL1002).

Subjects should not donate blood during participation in the study (from the start of screening onwards, see Section 4.2).

Consent will be obtained to contact the doctors that the subject sees regularly, to let them know that the subject is taking part in this study. It is important for all of the subject's doctors to know that the subject may be administered experimental vaccines. Subjects participating in the study

will be provided with a "wallet (study) card" and instructed to carry this card with them for the duration of the study (see Section 12.3.1).

9.5. Sample Collection and Handling

The actual dates and times of sample collection must be recorded in the CRF or laboratory requisition form. Refer to the Time and Events Schedule and the Additional Time and Events Schedule for the timing and frequency of all sample collections.

Sample collection and processing will be performed by the study-site personnel according to current versions of approved standard operating procedures.

Instructions for the collection, handling, storage, and shipment of samples are found in the Laboratory Manual that will be provided. Collection, handling, storage, and shipment of samples must be under the specified, and where applicable, controlled temperature conditions as indicated in the Laboratory Manual.

10. SUBJECT COMPLETION/DISCONTINUATION OF STUDY VACCINATION/WITHDRAWAL FROM THE STUDY

10.1. Completion

A subject will be considered to have completed the study if he or she has completed all assessments at the 6-month post-boost visit (or Day 365 if no boost vaccination was administered) for subjects who received placebo or at the Day 365 visit for subjects who received active vaccine. A subject who agrees to participate in the roll-over study will be considered to have completed the study when he or she completed the last safety-related visit in the current study (VAC52150EBL2001) or when he or she has been followed for an additional 12 months (whichever comes first).

10.2. Discontinuation of Study Vaccination/Withdrawal From the Study

Discontinuation of Study Vaccination

If a subject's study vaccine must be discontinued before the end of the vaccination schedule, this will not result in automatic withdrawal of the subject from the study.

A subject’s study vaccine (prime or boost) must be discontinued at the discretion of the investigator and after consultation with the sponsor for any of the events in Section 6.2.

A subject's study vaccine will be **permanently** discontinued in case any of the following listed criteria is met:

- The investigator believes that for safety reasons (eg, adverse event) it is in the best interest of the subject to discontinue study vaccine;
- The subject becomes pregnant;
- The subject has confirmed Ebola virus disease;

- The subject experiences any of the events in Section 6.3;
- The randomization code is broken by the investigator or the study-site personnel.

Subjects meeting any of the reasons listed above must not receive any further study vaccine, but should continue to be monitored for safety and for immunogenicity according to the protocol if this does not result in safety risks for the subject. In case of early discontinuation of study vaccine due to an adverse event, the investigator will collect all information relevant to the adverse event and safety of the subject, and will follow the subject to resolution, or until reaching a clinically stable endpoint.

Withdrawal From the Study

Each subject has the right to withdraw from the study at any time for whatever reason. The investigator should make an attempt to contact subjects who did not return for scheduled visits or follow-up. Although the subject is not obliged to give reason(s) for withdrawing early, the investigator should make a reasonable effort to ascertain the reason(s) while fully respecting the subject's rights.

A subject will be withdrawn from the study for any of the following reasons:

- Decision by the investigator to withdraw a subject for repeated failure to comply with protocol requirements;
- Decision by the sponsor to stop or cancel the study;
- Decision by local regulatory authorities and IEC/IRB to stop or cancel the study;
- Lost to follow-up;
- Withdrawal of consent;
- Death.

If a subject withdraws early from the study for any of the reasons listed above (except in case of death), early withdrawal assessments should be obtained per the assessments for the 21-day post-boost visit. A subject who wishes to withdraw consent from participation in the study will be offered an optional visit for safety follow-up (before formal withdrawal of consent), but the subject has the right to refuse.

If a subject is lost to follow-up, every reasonable effort must be made by the study-site personnel to contact the subject and determine the reason for discontinuation/withdrawal. The measures taken to follow up must be documented.

When a subject withdraws before completing the study, the reason for withdrawal is to be documented in the CRF and in the source document. Study vaccine assigned to the withdrawn subject may not be assigned to another subject. For subjects who withdraw from the study after randomization but before the prime vaccination, an additional subject will be enrolled who will receive the same vaccine regimen as the withdrawn subject. No additional subjects will be enrolled in case subjects withdraw from the study after receiving the prime vaccination.

10.3. Withdrawal From the Use of Research Samples

A subject who withdraws from the study will have the following options regarding the optional research samples or for storage of samples for future use:

- The collected samples will be retained and used in accordance with the subject's original informed consent for optional research samples or for storage of samples for future use.
- The subject may withdraw consent for optional research samples or for storage of samples for future use, in which case the samples will be destroyed and no further testing will take place. To initiate the sample destruction process, the investigator must notify the sponsor study site contact of withdrawal of consent for the optional research samples and to request sample destruction. The sponsor study site contact will, in turn, contact the biomarker representative to execute sample destruction. If requested, the investigator will receive written confirmation from the sponsor that the samples have been destroyed.

Withdrawal From the Optional Research Samples While Remaining in the Main Study

The subject may withdraw consent for optional research samples while remaining in the study. In such a case, the optional research samples will be destroyed. The sample destruction process will proceed as described above.

Withdrawal From Storage of Samples for Future Use While Remaining in the Study

The subject may withdraw consent for storage of samples for future use (refer to Section 16.2.5) while remaining in the study. In such a case, the samples will be destroyed as described above. Details of the sample retention for research are presented in the ICF.

11. STATISTICAL METHODS

Statistical analysis on subject information, safety and core immunogenicity data will be done by the sponsor or under the authority of the sponsor. A general description of the statistical methods to be used is outlined below. Specific details will be provided in the Statistical Analysis Plan (SAP). The analysis to estimate survival based on immunobridging methodology, linking NHP immune response data with human immune response data, will be described in a separate SAP.

Separately, data of specific and additional immunogenicity assessments, as outlined in Section 9.3.2, will be analyzed by the academic consortium partners. Specific details of these analyses will be provided in a separate SAP and reported separately from the Clinical Study Report.

Interim analyses may be performed as detailed in Section 11.6. The primary analysis will be performed when all subjects have completed the 6-month post-boost visit or discontinued earlier. This analysis will include 6-month post-boost safety and all available immunogenicity data up to this point. The final analysis will be performed when all subjects have completed the last study-related visit or discontinued earlier. Note: due to delays in scheduled boost vaccinations caused by study pauses, the primary and final analysis may be combined.

11.1. Analysis Sets

The Full Analysis set includes all subjects who were randomized and received at least one dose of study vaccine, regardless of the occurrence of protocol deviations. Safety data will be analyzed based on the Full Analysis set.

The Immunogenicity analysis set includes all randomized and vaccinated subjects, who have data from baseline and at least one post-vaccination immunogenicity blood draw.

The Per Protocol analysis set includes all randomized and vaccinated subjects, who received both the prime and boost (administered not more than 10 days outside the visit window) vaccinations, have immunogenicity data from baseline and at least one post-vaccination evaluable immunogenicity sample, and have no major protocol violations influencing the immune response.

The immunogenicity analysis will be based on the Per Protocol analysis set. As sensitivity analysis, the immunogenicity data will also be analyzed based on the Immunogenicity analysis set, provided more than 10% of subjects in the Immunogenicity analysis set are excluded from the Per Protocol analysis set.

11.2. Sample Size Determination

An originally planned sample size of 612 subjects (pooled across the 3 cohorts) includes 492 subjects randomized to receive Ad26.ZEBOV and MVA-BN-Filo (in Cohorts II and III) to substantially contribute to an overall safety database of the Ad26.ZEBOV and MVA-BN-Filo prime-boost regimen. In Cohort I, 30 subjects will receive Ad26.ZEBOV and MVA-BN-Filo in an open-label fashion. Randomization to Group 3 was stopped per Amendment 4 to focus on the schedules for which an indication will be sought.

As randomization to Group 3 was not resumed, the overall sample size was reduced to approximately 543 subjects (Group 1: 204, Group 2: 204, Group 3: 135), with approximately 469 subjects (Group 1: 174, Group 2: 174, Group 3: 121) to receive Ad26.ZEBOV and MVA-BN-Filo (in Cohorts II and III) prime-boost regimen. Enrollment in the entire study will be stopped per Amendment 5. Details on the impact on the power will be described in the SAP and/or Clinical Study Report.

The probability of detecting an adverse event in each of the groups, and total, is given in Table 7, for various true incidence rates. In case a specific adverse event is not observed, the one-sided 97.5% upper confidence limit of the true incidence rate of this adverse event is less than 3.3%, 2.2% and 0.8% for sample sizes of 121, 174 and 499 (ie, total number of subjects in Cohorts I, II, and III to receive Ad26.ZEBOV and/or MVA-BN-Filo) subjects, respectively.

Table 7: Probability of Observing at Least One Adverse Event Given a True Adverse Event Incidence

True Adverse Event Incidence (%)	N=30	N=121	N=174	N=469	N=499
0.1	3%	11%	16%	37%	39%
0.5	14%	45%	58%	90%	92%
1.0	26%	70%	83%	99%	99%
2.5	53%	95%	99%	100%	100%
5.0	79%	100%	100%	100%	100%
10.0	96%	100%	100%	100%	100%

Having core immunogenicity response data available for 164 subjects per group (ie, 84 from Cohort II and 80 from Cohort III), the following pairwise differences in immune response (ELISA antibody levels against EBOV GP) between vaccine schedules in Table 8 can be detected at any given time point, for a given variability in ELISA antibody levels. With the reduction in sample size in Group 3, the power to detect these pairwise differences is at least 80%.

Table 8: Magnitude of Pairwise Difference to be Detected Between Vaccine Regimens With Sample Size of 164 Subjects

Variability ELISA Antibody Levels (SD, log_{10} scale)	Detectable Difference (log_{10} scale) *
0.5	0.20
0.6	0.24
0.7	0.28
0.8	0.32
0.9	0.37
1.0	0.41
1.1	0.45
1.2	0.49

SD: standard deviation; * 90% power, 2-sided test, 5% significance level, accounting for multiple testing

11.3. Subject Information

For all subjects, demographic characteristics (eg, age, height, weight, body mass index, race, and sex) and screening/baseline characteristics (eg, physical examination, medical history) will be tabulated and summarized with descriptive statistics.

11.4. Safety Analyses

No formal statistical testing of safety data is planned. Safety data will be analyzed descriptively (including 95% confidence intervals, if applicable), by cohort and overall.

Baseline for all safety parameters will be defined as the last value before the prime vaccination.

Adverse Events (Including Reactogenicity)

The verbatim terms used in the CRF by investigators to report adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). All reported adverse events (solicited local, solicited systemic, and unsolicited) will be included in the analysis. For each

adverse event, the number and percentage of subjects who experience at least 1 occurrence of the given event will be summarized by group. Summaries, listings, datasets, or subject narratives may be provided, as appropriate, for those subjects who die, who discontinue study vaccine due to an adverse event, or who experience a severe or a serious adverse event.

The analysis for solicited adverse events will be done on those subjects in the Full Analysis set for whom reactogenicity assessments are available in the database. The analysis of unsolicited adverse events will be done on the Full Analysis set.

Clinical Laboratory Tests

Laboratory abnormalities will be determined according to the toxicity grading tables, and in accordance with the normal ranges of the clinical laboratory. The most severe laboratory abnormalities following vaccination will be tabulated per regimen.

Vital Signs and Electrocardiogram

Vital signs and ECG abnormalities following vaccination will be tabulated by most severe abnormality grade.

Physical Examination

Physical examination abnormalities following vaccination will be tabulated by most severe abnormality grade.

11.5. Immunogenicity Analyses

Descriptive statistics (actual values and changes from baseline, including 95% confidence intervals, if applicable) will be calculated for continuous immunologic parameters at each time point. Graphical representations of changes in immunologic parameters will be prepared, as applicable. Frequency tabulations will be calculated for discrete (qualitative) immunologic parameters at each time point. In addition, differences between the vaccination schedules will be evaluated at the 21-day post-boost, 6-month post-boost and Day 365 visits.

Integrative statistical analysis of the gene expression and functional assays and modeling analyses of data obtained in Cohort II will be detailed and reported separately from this study.

The SAP will provide details on how the samples from the subjects whose boost is out of window due to the pause are to be handled in the statistical analyses.

11.6. Interim Analyses

Interim analyses may be performed during the study for regulatory purposes or for the purpose of informing future vaccine-related decisions in a timely manner. The results will not influence the conduct of the study in terms of early termination or later safety or immunogenicity endpoint assessments.

A separate SAP will be prepared before the conduct of an interim analysis.

11.7. Independent Data Monitoring Committee

An IDMC will be established to monitor safety data on an ongoing basis to ensure the continuing safety of the subjects enrolled in this study. The committee will meet periodically to review interim data. Ad hoc IDMC meetings may be requested via the sponsor for any single event or combination of multiple events which are considered to jeopardize the safety of the subjects. After the review, the IDMC will make recommendations regarding the continuation of the study. The details will be provided in a separate IDMC charter.

The IDMC will be appointed by the sponsor with recommendations from the Clinical Steering Committee (see Section 11.8) before the start of the study. The IDMC will consist of at least one medical expert in the relevant therapeutic area and at least one statistician. The IDMC responsibilities, authorities, and procedures will be documented in its charter.

11.8. Trial Management Team and Clinical Steering Committee

Oversight of the study will be conducted by the Trial Management Team which will consist of members from the sponsor as well as key collaborators from participating countries. This Trial Management Team will be responsible for protocol development and ensuring proper study execution.

As part of the consortium agreement between the sponsor, INSERM and the London School of Hygiene and Tropical Medicine, a Clinical Steering Committee with representatives from each consortium partner has been established to guide the overall clinical development plan, including this study. The Clinical Steering Committee is responsible for overseeing the EBOVAC2 collaboration activities and for making decisions on specific issues.

12. ADVERSE EVENT REPORTING

Timely, accurate, and complete reporting and analysis of safety information from clinical studies are crucial for the protection of subjects, investigators, and the sponsor, and are mandated by regulatory agencies worldwide. The sponsor has established Standard Operating Procedures in conformity with regulatory requirements worldwide to ensure appropriate reporting of safety information; all clinical studies conducted by the sponsor or its affiliates will be conducted in accordance with those procedures.

12.1. Definitions

12.1.1. Adverse Event Definitions and Classifications

Adverse Event

An adverse event is any untoward medical occurrence in a clinical study subject administered a medicinal (investigational or non-investigational) product. An adverse event does not necessarily have a causal relationship with the treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal finding), symptom, or disease temporally associated with the use of a medicinal (investigational or non-investigational) product, whether or not

related to that medicinal (investigational or non-investigational) product. (Definition per International Council for Harmonisation [ICH])

This includes any occurrence that is new in onset or aggravated in severity or frequency from the baseline condition, or abnormal results of diagnostic procedures, including laboratory test abnormalities.

Note: The sponsor collects adverse events starting with the signing of the ICF (refer to Section 12.3.1, for time of last adverse event recording).

Serious Adverse Event

A serious adverse event based on ICH and EU Guidelines on Pharmacovigilance for Medicinal Products for Human Use is any untoward medical occurrence that at any dose:

- Results in death
- Is life-threatening
  (The subject was at risk of death at the time of the event. It does not refer to an event that hypothetically might have caused death if it were more severe.)
- Requires inpatient hospitalization or prolongation of existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect
- Is a suspected transmission of any infectious agent via a medicinal product
- Is Medically Important*

*Medical and scientific judgment should be exercised in deciding whether expedited reporting is also appropriate in other situations, such as important medical events that may not be immediately life threatening or result in death or hospitalization but may jeopardize the subject or may require intervention to prevent one of the other outcomes listed in the definition above. These should usually be considered serious.

If a serious and unexpected adverse event occurs for which there is evidence suggesting a causal relationship between the study vaccine and the event (eg, death from anaphylaxis), the event must be reported as suspected unexpected serious adverse reaction (SUSAR) (even after the study is over, if the sponsor, IDMC or investigator becomes aware of them) by the sponsor to the Health Authorities and by the investigator to the IEC/IRB according to regulatory and local requirements.

Unlisted (Unexpected) Adverse Event/Reference Safety Information

An adverse event is considered unlisted if the nature or severity is not consistent with the applicable product reference safety information. For Ad26.ZEBOV and for MVA-BN-Filo, the expectedness of an adverse event will be determined by whether or not it is listed in the Investigator's Brochures and Addenda, if applicable.

Adverse Event Associated With the Use of the Study Vaccine

An adverse event is considered associated with the use of the study vaccine if the attribution is **possibly**, **probably**, or **very likely** by the definitions listed in Section 12.1.2, Attribution Definitions.

Immediate Reportable Events

The following list of neuroinflammatory disorders are categorized as IREs, and should be reported to the sponsor within 24 hours of becoming aware of the event using the IRE Form. Relevant data from the IRE Form will be captured in the clinical database.

- Cranial nerve disorders, including paralyses/paresis (eg, Bell's palsy)
- Optic neuritis
- Multiple sclerosis
- Transverse myelitis
- Guillain-Barré syndrome, including Miller Fisher syndrome, Bickerstaff's encephalitis and other variants
- Acute disseminated encephalomyelitis, including site specific variants: eg, non-infectious encephalitis, encephalomyelitis, myelitis, myeloradiculomyelitis
- Myasthenia gravis and Lambert-Eaton myasthenic syndrome
- Immune-mediated peripheral neuropathies and plexopathies (including chronic inflammatory demyelinating polyneuropathy, multifocal motor neuropathy and polyneuropathies associated with monoclonal gammopathy).
- Narcolepsy
- Isolated paresthesia of >7 days duration

Symptoms, signs or conditions which might (or might not) represent the above diagnoses, should be recorded and reported as IREs even if the final or definitive diagnosis has not yet been determined, and alternative diagnoses have not yet been eliminated or shown to be less likely. Follow-up information and final diagnoses, if applicable, should be submitted as soon as they become available.

If the IRE is also serious (serious adverse event), it will also be reported using the same process as for other serious adverse events.

12.1.2. Attribution Definitions

Every effort should be made by the investigator to explain any adverse event and to assess its potential causal relationship, ie, to administration of the study vaccine or to alternative causes, eg, natural history of underlying disease(s), concomitant drug(s). This applies to all adverse events, whether serious or non-serious. Assessment of causality must be done by a licensed study physician (the investigator or designee).

The investigator will use the following guidelines to assess the causal relationship of an adverse event to study vaccine:

Not Related

An adverse event that is not related to the use of the vaccine.

Doubtful

An adverse event for which an alternative explanation is more likely, eg, concomitant drug(s), concomitant disease(s), or the relationship in time suggests that a causal relationship is unlikely.

Possible

An adverse event that might be due to the use of the vaccine. An alternative explanation, eg, concomitant drug(s), concomitant disease(s), is inconclusive. The relationship in time is reasonable; therefore, the causal relationship cannot be excluded.

Probable

An adverse event that might be due to the use of the vaccine. The relationship in time is suggestive. An alternative explanation is less likely, eg, concomitant drug(s), concomitant disease(s).

Very Likely

An adverse event that is listed as a possible adverse reaction and cannot be reasonably explained by an alternative explanation, eg, concomitant drug(s), concomitant disease(s). The relationship in time is very suggestive.

12.1.3. Severity Criteria

Adverse events and laboratory data will be coded for severity using the toxicity grading tables in Attachment 1. For adverse events not identified in the table, the following guidelines will apply:

Mild	Grade 1	Symptoms causing no or minimal interference with usual social and functional activities
Moderate	Grade 2	Symptoms causing greater than minimal interference with usual social and functional activities
Severe	Grade 3	Symptoms causing inability to perform usual social and functional activities

Note: Only clinically significant abnormalities in laboratory data occurring from signing of the ICF onwards will be reported as adverse events and graded using the table above.

12.2. Special Reporting Situations

Safety events of interest on a sponsor study vaccine that may require expedited reporting or safety evaluation include, but are not limited to:

- Administration of an overdose of study vaccine
- Accidental or occupational exposure to a study vaccine
- Administration error involving a study vaccine (with or without subject/patient exposure to the study vaccine, eg, name confusion)
- IREs

Special reporting situations should be recorded in the CRF. For reporting of IREs, refer to Section 12.3.3. Any special reporting situation that meets the criteria of a serious adverse event should be recorded on the Serious Adverse Event page of the CRF.

12.3. Procedures

Refer to Section 9.1.5 for more information for subjects in France who have further follow-ups from Day 365 onwards until the roll-over study is opened.

12.3.1. All Adverse Events

All adverse events and special reporting situations, whether serious or non-serious, will be reported from the time a signed and dated ICF is obtained until the 42-day post-boost visit. Serious adverse events and IREs will be collected from signing of the ICF onwards until the end of the study. Subjects will record symptoms of solicited local or systemic adverse events (reactogenicity) in the diary in the evening after each vaccination and then daily for the next 7 days. Serious adverse events must be reported by the investigator using the Serious Adverse Event Form. SUSARs will be reported even after the study is over, if the sponsor, the IDMC or the investigator becomes aware of them. The sponsor will evaluate any safety information that is spontaneously reported by an investigator beyond the time frame specified in the protocol.

All adverse events, regardless of seriousness, severity, or presumed relationship to study vaccine, must be recorded using medical terminology in the source document and the CRF. Whenever possible, diagnoses should be given when signs and symptoms are due to a common etiology (eg, cough, runny nose, sneezing, sore throat, and head congestion should be reported as "upper respiratory infection"). Investigators must record in the CRF their opinion concerning the relationship of the adverse event to study vaccine. All measures required for adverse event management must be recorded in the source document and reported according to sponsor instructions. For reporting of IREs, refer to Section 12.3.3.

The sponsor assumes responsibility for appropriate reporting of adverse events to the regulatory authorities. The sponsor will also report to the investigator (and the head of the investigational institute where required) all SUSARs. The investigator (or sponsor where required) must report SUSARs to the appropriate IEC/IRB that approved the protocol unless otherwise required and documented by the IEC/IRB. A SUSAR will be reported to regulatory authorities unblinded.

Participating investigators and IEC/IRB will receive a blinded SUSAR summary, unless otherwise specified.

For all studies with an outpatient phase, including open-label studies, the subject must be provided with a "wallet (study) card" and instructed to carry this card with them for the duration of the study indicating the following:

- Study number
- Statement, in the local language(s), that the subject is participating in a clinical study
- Investigator's name and 24-hour contact telephone number
- Local sponsor's name and 24-hour contact telephone number (for medical staff only)
- Site number
- Subject number
- Any other information that is required to do an emergency breaking of the blind

12.3.2. Serious Adverse Events

All serious adverse events occurring during the study must be reported to the appropriate sponsor contact person by study-site personnel within 24 hours of their knowledge of the event.

Information regarding serious adverse events will be transmitted to the sponsor using the Serious Adverse Event Form, which must be completed and signed by a physician from the study site, and transmitted to the sponsor within 24 hours. The initial and follow-up reports of a serious adverse event should be made by facsimile (fax).

All serious adverse events that have not resolved by the end of the study, or that have not resolved upon discontinuation of the subject's participation in the study, must be followed until any of the following occurs:

- The event resolves
- The event stabilizes
- The event returns to baseline, if a baseline value/status is available
- The event can be attributed to agents other than the study vaccine or to factors unrelated to study conduct
- It becomes unlikely that any additional information can be obtained (subject or health care practitioner refusal to provide additional information, lost to follow-up after demonstration of due diligence with follow-up efforts)

Suspected transmission of an infectious agent by a medicinal product will be reported as a serious adverse event. Any event requiring hospitalization (or prolongation of hospitalization) that occurs during the course of a subject's participation in a study must be reported as a serious adverse event, except hospitalizations for the following:

- Hospitalizations not intended to treat an acute illness or adverse event (eg, social reasons such as pending placement in long-term care facility)
- Surgery or procedure planned before entry into the study (must be documented in the CRF). *Note: Hospitalizations that were planned before the signing of the ICF, and where the underlying condition for which the hospitalization was planned has not worsened, will not be considered serious adverse events. Any adverse event that results in a prolongation of the originally planned hospitalization is to be reported as a new serious adverse event.*

The cause of death of a subject in a study, whether or not the event is expected or associated with the study vaccine, is considered a serious adverse event.

12.3.3. Immediate Reportable Events

One subject in the study VAC52150EBL2001 experienced a serious and very rare condition called "Miller Fisher syndrome". This condition consists of double vision, pain on moving the eye, and difficulty with balance while walking. Miller Fisher syndrome most commonly occurs following a recent infection. The subject experienced these symptoms about a week after suffering from a common cold and fever. The event happened about a month after boost vaccination with either MVA-BN-Filo or placebo. This subject had to go to the hospital for treatment and has recovered. After an extensive investigation, the event has been considered to be doubtfully related to vaccine and most likely related to the previous common cold.

Any events of neuroimmunologic significance (listed in Section 12.1.1) should be categorized as IREs and should be reported throughout the study using the IRE Form provided **within 24 hours to the sponsor**. Events suggestive of the disorders considered IREs should be reported even if the final diagnosis has not been yet determined, and follow-up information and final diagnosis should be submitted to the sponsor as soon as they become available.

If an event meets serious adverse event criteria (see above), it should be documented as such using the Serious Adverse Event Form, as well as the relevant CRF Adverse Event page and the complete IRE Form page 3 to be included as part of the Serious Adverse Event report.

12.3.4. Pregnancy

Pregnancies will be reported from signing of the ICF until the end of the study.

All initial reports of pregnancy in female subjects or partners of male subjects must be reported to the sponsor by the study-site personnel within 24 hours of their knowledge of the event using the appropriate pregnancy notification form. Abnormal pregnancy outcomes (eg, spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered serious adverse events and must be reported using the Serious Adverse Event Form. Any subject who becomes pregnant during the study must be promptly withdrawn from further study vaccination but should continue participation in the study for safety follow-up.

Because the effect of the study vaccine on sperm is unknown, pregnancies in partners of male subjects included in the study will be reported as noted above.

Follow-up information regarding the outcome of the pregnancy and any postnatal sequelae in the infant will be required.

The parent(s)/legal guardian of children born to vaccinated female subjects who become pregnant with estimated conception within 28 days after vaccination with MVA-BN-Filo or within 3 months after vaccination with Ad26.ZEBOV, will be approached to consent for enrollment of their offspring into the VAC52150 Vaccine Development Roll-over study, according to the same rules that apply for the other subjects (see Section 9.1.6).

12.4. Contacting Sponsor Regarding Safety

The names (and corresponding telephone numbers) of the individuals who should be contacted regarding safety issues or questions regarding the study are listed in the Contact Information page(s), which will be provided as a separate document.

13. PRODUCT QUALITY COMPLAINT HANDLING

A product quality complaint (PQC) is defined as any suspicion of a product defect related to manufacturing, labeling, or packaging, ie, any dissatisfaction relative to the identity, quality, durability, or reliability of a product, including its labeling or package integrity. A PQC may have an impact on the safety and efficacy of the product. Timely, accurate, and complete reporting and analysis of PQC information from studies are crucial for the protection of subjects, investigators, and the sponsor, and are mandated by regulatory agencies worldwide. The sponsor has established procedures in conformity with regulatory requirements worldwide to ensure appropriate reporting of PQC information; all studies conducted by the sponsor or its affiliates will be conducted in accordance with those procedures.

13.1. Procedures

All initial PQCs must be reported to the sponsor by the study-site personnel within 24 hours after being made aware of the event.

If the defect is combined with a serious adverse event, the study-site personnel must report the PQC to the sponsor according to the serious adverse event reporting timelines (refer to Section 12.3.2). A sample of the suspected product should be maintained for further investigation if requested by the sponsor.

13.2. Contacting Sponsor Regarding Product Quality

The names (and corresponding telephone numbers) of the individuals who should be contacted regarding product quality issues are listed in the Contact Information page(s), which will be provided as a separate document.

14. STUDY VACCINE INFORMATION

14.1. Description of Study Vaccines

Ad26.ZEBOV

Ad26.ZEBOV is a monovalent recombinant, replication-incompetent Ad26-based vector that expresses the full length EBOV Mayinga GP and is produced in the human cell line PER.C6®.

The Ad26.ZEBOV vaccine will be supplied at a concentration of $1x10^{11}$ vp/mL in 2-mL single-use glass vials as a frozen liquid suspension to be thawed before use. Each vial contains an extractable volume of 0.5 mL. Refer to the Investigator's Brochure for a list of excipients.[12]

The Ad26.ZEBOV vaccine is manufactured by IDT Biologika GmbH for Janssen Vaccines & Prevention B.V., The Netherlands.

MVA-BN-Filo

MVA-BN-Filo is a recombinant multivalent vaccine intended for active immunization against Ebola and Marburg virus infection. MVA-BN-Filo is strongly attenuated; the vaccine is propagated in primary chicken embryo fibroblast cells and does not replicate in human cells.

The MVA-BN-Filo vaccine is supplied at a concentration of $2x10^{8}$ Inf.U/mL (nominal titer) in 2-mL glass vials as a frozen liquid suspension to be thawed before use. Each vial contains an extractable volume of 0.5 mL. Refer to the Investigator's Brochure for a list of excipients.[14]

The MVA-BN-Filo vaccine is manufactured by IDT Biologika GmbH for Janssen Vaccines & Prevention B.V., The Netherlands.

Placebo

The placebo supplied for this study will be formulated as a sterile 0.9% saline for injection (as commercially available).

14.2. Packaging and Labeling

All study vaccines will be manufactured and packaged in accordance with Good Manufacturing Practice (GMP). All study vaccines will be packaged and labeled under the responsibility of the sponsor. No study vaccine can be repacked or relabeled on site without prior approval from the sponsor.

Further details for study vaccine packaging and labeling can be found in the Site Investigational Product Procedures Manual.

14.3. Preparation, Handling, and Storage

Study vaccine must be stored at controlled temperatures: Ad26.ZEBOV vials must be stored at ≤ -65°C and MVA-BN-Filo vials must be stored at ≤ -20°C.

Vials must be stored in a secured location with no access for unauthorized personnel. All study product storage equipment (including refrigerators, freezers), must be equipped with a continuous temperature monitor and alarm, and with back-up power systems. In the event that study vaccine is exposed to temperatures outside the specified temperature ranges, all relevant data will be sent to the sponsor to determine if the affected study vaccine can be used or will be replaced. The affected study vaccine must be quarantined and not used until further instruction from the sponsor is received.

Blinding will be achieved by preparation of study vaccine by unblinded qualified study-site personnel not involved in any other study-related procedure, and by the administration of vaccine in a masked syringe by a study vaccine administrator (see Definitions of Terms). To preserve blinding, the unblinded qualified study-site personnel will place a blinding tape on the syringe to mask its content.

Details on the preparation, the holding time and storage conditions from the time of preparation to administration of Ad26.ZEBOV and MVA-BN-Filo are provided in the Site Investigational Product Procedures Manual.

14.4. Study Vaccine Accountability

The investigator is responsible for ensuring that all study vaccine received at the site is inventoried and accounted for throughout the study. The study vaccine administered to the subject must be documented on the study vaccine accountability form. All study vaccine will be stored and disposed of according to the sponsor's instructions. Study-site personnel must not combine contents of the study vaccine containers.

Study vaccine must be handled in strict accordance with the protocol and the container label, and must be stored at the study site in a limited-access area or in a locked cabinet under appropriate environmental conditions. Unused study vaccine must be available for verification by the sponsor's study site monitor during on-site monitoring visits. The return to the sponsor of unused study vaccine will be documented on the study vaccine return form. When the study site is an authorized destruction unit and study vaccine supplies are destroyed on-site, this must also be documented on the study vaccine return form.

Potentially hazardous materials such as used ampules, needles, syringes and vials containing hazardous liquids should be disposed of immediately in a safe manner and therefore will not be retained for study vaccine accountability purposes.

Study vaccine should be dispensed under the supervision of the investigator or a qualified member of the study-site personnel, or by a qualified staff member. Study vaccine will be supplied only to subjects participating in the study. Returned study vaccine must not be dispensed again, even to the same subject. Study vaccine may not be relabeled or reassigned for use by other subjects. The investigator agrees neither to dispense the study vaccine from, nor store it at, any site other than the study sites agreed upon with the sponsor.

15. STUDY-SPECIFIC MATERIALS

The investigator will be provided with the following supplies:

- Investigator's Brochures and Addenda (if applicable) for Ad26.ZEBOV and MVA-BN-Filo
- Site Investigational Product Procedures Manual
- Laboratory Manual
- IWRS Manual
- Electronic Data Capture (eDC) Manual/electronic CRF Completion Guidelines and Randomization Instructions
- Sample ICF
- Subject diaries
- Rulers, thermometers
- Subject wallet cards
- Recruitment tools, as applicable

16. ETHICAL ASPECTS

16.1. Study-specific Design Considerations

Potential subjects will be fully informed of the risks and requirements of the study and, during the study, subjects will be given any new information that may affect their decision to continue participation. They will be told that their consent to participate in the study is voluntary and may be withdrawn at any time with no reason given and without penalty or loss of benefits to which they would otherwise be entitled. Only subjects who are fully able to understand the risks, benefits, and potential adverse events of the study, and provide their consent voluntarily will be enrolled.

The total blood volume to be collected is considered to be within the limits of standard blood donation.

16.2. Regulatory Ethics Compliance

16.2.1. Investigator Responsibilities

The investigator is responsible for ensuring that the study is performed in accordance with the protocol, current ICH guidelines on Good Clinical Practice (GCP), and applicable regulatory and country-specific requirements.

Good Clinical Practice is an international ethical and scientific quality standard for designing, conducting, recording, and reporting studies that involve the participation of human subjects. Compliance with this standard provides public assurance that the rights, safety, and well-being of study subjects are protected, consistent with the principles that originated in the Declaration of Helsinki, and that the study data are credible.

16.2.2. Independent Ethics Committee or Institutional Review Board

Before the start of the study, the investigator (or sponsor where required) will provide the IEC/IRB with current and complete copies of the following documents (as required by local regulations):

- Final protocol and, if applicable, amendments
- Sponsor-approved ICF (and any other written materials to be provided to the subjects)
- Investigator's Brochure (or equivalent information) and amendments/addenda
- Sponsor-approved subject recruiting materials
- Information on compensation for study-related injuries or payment to subjects for participation in the study, if applicable
- Investigator's curriculum vitae or equivalent information (unless not required, as documented by the IEC/IRB)
- Information regarding funding, name of the sponsor, institutional affiliations, other potential conflicts of interest, and incentives for subjects
- Any other documents that the IEC/IRB requests to fulfill its obligation

This study will be undertaken only after the IEC/IRB has given full approval of the final protocol, amendments (if any, excluding the ones that are purely administrative, with no consequences for subjects, data or study conduct, unless required locally), the ICF, applicable recruiting materials, and subject compensation programs, and the sponsor has received a copy of this approval. This approval letter must be dated and must clearly identify the IEC/IRB and the documents being approved.

Approval for the collection of optional samples for research and for the corresponding ICF must be obtained from the IEC/IRB. Approval for the protocol can be obtained independent of this optional research component.

During the study the investigator (or sponsor where required) will send the following documents and updates to the IEC/IRB for their review and approval, where appropriate:

- Protocol amendments (excluding the ones that are purely administrative, with no consequences for subjects, data or study conduct)
- Revision(s) to ICF and any other written materials to be provided to subjects
- If applicable, new or revised subject recruiting materials approved by the sponsor
- Revisions to compensation for study-related injuries or payment to subjects for participation in the study, if applicable
- New edition(s) of the Investigator's Brochure and amendments/addenda
- Summaries of the status of the study at intervals stipulated in guidelines of the IEC/IRB (at least annually)

- Reports of adverse events that are serious, unlisted/unexpected, and associated with the study vaccine
- New information that may adversely affect the safety of the subjects or the conduct of the study
- Deviations from or changes to the protocol to eliminate immediate hazards to the subjects
- Report of deaths of subjects under the investigator's care
- Notification if a new investigator is responsible for the study at the site
- Development Safety Update Report and Line Listings, where applicable
- Any other requirements of the IEC/IRB

For all protocol amendments (excluding the ones that are purely administrative, with no consequences for subjects, data or study conduct), the amendment and applicable ICF revisions must be submitted promptly to the IEC/IRB for review and approval before implementation of the change(s).

At least once a year, the IEC/IRB will be asked to review and reapprove this study, where required.

At the end of the study, the investigator (or sponsor where required) will notify the IEC/IRB about the study completion.

16.2.3. Informed Consent

Each subject must give written consent according to local requirements after the nature of the study has been fully explained. The ICF(s) must be signed before performance of any study-related activity. The ICF(s) that is/are used must be approved by both the sponsor and by the reviewing IEC/IRB and be in a language that the subject can read and understand. The informed consent should be in accordance with principles that originated in the Declaration of Helsinki, current ICH and GCP guidelines, applicable regulatory requirements, and sponsor policy.

Before enrollment in the study, the investigator or an authorized member of the study-site personnel must explain to potential subjects the aims, methods, reasonably anticipated benefits, and potential hazards of the study, and any discomfort participation in the study may entail. Subjects will be informed that their participation is voluntary and that they may withdraw consent to participate at any time. They will be informed that choosing not to participate will not affect the care the subject will receive. Finally, they will be told that the investigator will maintain a subject identification register for the purposes of long-term follow up if needed and that their records may be accessed by health authorities and authorized sponsor personnel without violating the confidentiality of the subject, to the extent permitted by the applicable law(s) or regulations. By signing the ICF the subject is authorizing such access. It also denotes that the subject agrees to allow his or her study physician to recontact the subject for the purpose of obtaining consent for additional safety evaluations, if needed.

The subject will be given sufficient time to read the ICF and the opportunity to ask questions. After this explanation and before entry into the study, consent should be appropriately recorded by means of the subject's personally dated signature. After having obtained the consent, a copy of the ICF must be given to the subject.

Subjects will be asked for consent to provide optional samples for research. After informed consent for the study is appropriately obtained, the subject will be asked to sign and personally date a separate tick box on the ICF indicating agreement to participate in the optional research component. Refusal to participate in the optional research will not result in ineligibility for the study. A copy of this signed ICF will be given to the subject.

If the subject is unable to read or write, an impartial witness should be present for the entire informed consent process (which includes reading and explaining all written information) and should personally date and sign the ICF after the oral consent of the subject is obtained.

16.2.4. Privacy of Personal Data

The collection and processing of personal data from subjects enrolled in this study will be limited to those data that are necessary to fulfill the objectives of the study.

These data must be collected and processed with adequate precautions to ensure confidentiality and compliance with applicable data privacy protection laws and regulations. Appropriate technical and organizational measures to protect the personal data against unauthorized disclosures or access, accidental or unlawful destruction, or accidental loss or alteration must be put in place. Sponsor personnel whose responsibilities require access to personal data agree to keep the identity of subjects confidential.

The informed consent obtained from the subject includes explicit consent for the processing of personal data and for the investigator/institution to allow direct access to his or her original medical records (source data/documents) for study-related monitoring, audit, IEC/IRB review, and regulatory inspection. This consent also addresses the transfer of the data to other entities and to other countries.

The subject has the right to request through the investigator access to his or her personal data and the right to request rectification of any data that are not correct or complete. Reasonable steps will be taken to respond to such a request, taking into consideration the nature of the request, the conditions of the study, and the applicable laws and regulations.

Exploratory immunogenicity research is not conducted under standards appropriate for the return of data to subjects. In addition, the sponsor cannot make decisions as to the significance of any findings resulting from exploratory research. Therefore, exploratory research data will not be returned to subjects or investigators, unless required by law or local regulations. Privacy and confidentiality of data generated in the future on stored samples will be protected by the same standards applicable to all other clinical data.

16.2.5. Long-term Retention of Samples for Additional Future Research

Samples collected in this study, for which consent has been obtained and for which additional material is available after study-specified testing is complete, may be stored for up to 15 years (or according to local regulations) for possible additional future scientific/genetic research. Samples will only be used to understand Ebola vaccine- and disease-related questions and to develop tests/assays related to the characterization of EBOV-directed immune responses or diagnostic tests. The research may begin at any time during the study or the post-study storage period. Applicable approvals will be sought before any such samples are used for analysis not specified in the protocol (amendment) approved by the IEC/IRB.

Stored samples will be coded throughout the sample storage and analysis process and will not be labeled with personal identifiers. Subjects may withdraw their consent for their samples to be stored for future research at any time (refer to Section 10.3, Withdrawal From the Use of Research Samples). In such case, their blood samples will be destroyed after all the tests specified for this study have been concluded.

The sponsor will be responsible for the overall management of the sample inventory, shipping plan, allocation and storage of samples.

16.2.6. Country Selection

This study will only be conducted in those countries where the intent is to launch or otherwise help ensure access to the developed product if the need for the product persists, unless explicitly addressed as a specific ethical consideration in Section 16.1.

17. ADMINISTRATIVE REQUIREMENTS

17.1. Protocol Amendments

Neither the investigator nor the sponsor will modify this protocol without a formal amendment by the sponsor. All protocol amendments must be issued by the sponsor, and signed and dated by the investigator. Protocol amendments must not be implemented without prior IEC/IRB approval, or when the relevant competent authority has raised any grounds for non-acceptance, except when necessary to eliminate immediate hazards to the subjects, in which case the amendment must be promptly submitted to the IEC/IRB and relevant competent authority. Documentation of amendment approval by the investigator and IEC/IRB must be provided to the sponsor. When the change(s) involve(s) only logistic or administrative aspects of the study, the IRB/IEC (where required) only needs to be notified.

During the course of the study, in situations where a departure from the protocol is unavoidable, the investigator or other physician in attendance will contact the appropriate sponsor representative (listed in the Contact Information page(s), which will be provided as a separate document). Except in emergency situations, this contact should be made before implementing any departure from the protocol. In all cases, contact with the sponsor must be made as soon as possible to discuss the situation and agree on an appropriate course of action. The data recorded in the CRF and source documents will reflect any departure from the protocol, and the source documents will describe this departure and the circumstances requiring it.

17.2. Regulatory Documentation

17.2.1. Regulatory Approval/Notification

This protocol and any amendment(s) must be submitted to the appropriate regulatory authorities in each respective country, if applicable. A study may not be initiated until all local regulatory requirements are met.

17.2.2. Required Prestudy Documentation

The following documents must be provided to the sponsor before shipment of study vaccine to the study site:

- Protocol and amendment(s), if any, signed and dated by the principal investigator
- A copy of the dated and signed (or sealed, where appropriate per local regulations), written IEC/IRB approval of the protocol, amendments, ICF, any recruiting materials, and if applicable, subject compensation programs. This approval must clearly identify the specific protocol by title and number and must be signed (or sealed, where appropriate per local regulations) by the chairman or authorized designee.
- Name and address of the IEC/IRB, including a current list of the IEC/IRB members and their function, with a statement that it is organized and operates according to GCP and the applicable laws and regulations. If accompanied by a letter of explanation, or equivalent, from the IEC/IRB, a general statement may be substituted for this list. If an investigator or a member of the study-site personnel is a member of the IEC/IRB, documentation must be obtained to state that this person did not participate in the deliberations or in the vote/opinion of the study.
- Regulatory authority approval or notification, if applicable
- Signed and dated statement of investigator (eg, Form FDA 1572), if applicable
- Documentation of investigator qualifications (eg, curriculum vitae)
- Completed investigator financial disclosure form from the principal investigator, where required
- Signed and dated clinical trial agreement, which includes the financial agreement
- Any other documentation required by local regulations

The following documents must be provided to the sponsor before enrollment of the first subject:

- Completed investigator financial disclosure forms from all subinvestigators
- Documentation of subinvestigator qualifications (eg, curriculum vitae)
- Name and address of any local laboratory conducting tests for the study, and a dated copy of current laboratory normal ranges for these tests, if applicable
- Local laboratory documentation demonstrating competence and test reliability (eg, accreditation/license), if applicable

17.3. Subject Identification, Enrollment, and Screening Logs

The investigator agrees to complete a subject identification and enrollment log to permit easy identification of each subject during and after the study. This document will be reviewed by the sponsor study-site contact for completeness.

The subject identification and enrollment log will be treated as confidential and will be filed by the investigator in the study file. To ensure subject confidentiality, no copy will be made. All reports and communications relating to the study will identify subjects by subject identification and date of birth. In cases where the subject is not randomized into the study, the date seen and date of birth will be used.

The investigator must also complete a subject screening log, which reports on all subjects who were seen to determine eligibility for inclusion in the study.

17.4. Source Documentation

At a minimum, source documents consistent in the type and level of detail with that commonly recorded at the study site as a basis for standard medical care, must be available for the following: subject identification, eligibility, and study identification; study discussion and date of signed informed consent; dates of visits; results of safety and immunogenicity parameters as required by the protocol; record of all adverse events and follow-up of adverse events; concomitant medication; study vaccine receipt/dispensing/return records; study vaccine administration information; and date of study completion and reason for early discontinuation of study vaccine or withdrawal from the study, if applicable. The author of an entry in the source documents should be identifiable.

Specific details required as source data for the study and source data collection methods will be reviewed with the investigator before the study and will be described in the monitoring guidelines (or other equivalent document).

The subject diary will be considered a source document. Information from the diary provided to subjects to record symptoms of solicited local and systemic adverse events until 7 days after each vaccination will be reviewed by the investigator to transcribe into the relevant parts of the CRF as described in the CRF Completion Guidelines.

17.5. Case Report Form Completion

Case report forms are prepared and provided by the sponsor for each subject in electronic format. All CRF entries, corrections, and alterations must be made by the investigator or authorized study-site personnel. The investigator must verify that all data entries in the CRF are accurate and correct.

The study data will be transcribed by study-site personnel from the source documents onto an electronic CRF, if applicable. Study-specific data will be transmitted in a secure manner to the sponsor. Data must be entered into CRF in English. The CRF must be completed as soon as possible after a subject visit, and the forms should be available for review at the next scheduled monitoring visit.

If necessary, queries will be generated in the eDC tool. If corrections to a CRF are needed after the initial entry into the CRF, this can be done in either of the following ways:

- Investigator and study site personnel can make corrections in the eDC tool at their own initiative or as a response to an auto query (generated by the eDC tool).
- Sponsor or sponsor delegate can generate a query for resolution by the investigator and study-site personnel.

17.6. Data Quality Assurance/Quality Control

Steps to be taken to ensure the accuracy and reliability of data include the selection of qualified investigators and appropriate study sites, review of protocol procedures with the investigator and study-site personnel before the study, periodic monitoring visits by the sponsor, and direct transmission of clinical laboratory data from a central laboratory into the sponsor's database. Written instructions will be provided for collection, handling, storage, and shipment of samples.

Guidelines for CRF completion will be provided and reviewed with study-site personnel before the start of the study. The sponsor will review CRF for accuracy and completeness during on-site monitoring visits and after transmission to the sponsor; any discrepancies will be resolved with the investigator or designee, as appropriate. After upload of the data into the study database they will be verified for accuracy and consistency with the data sources.

17.7. Record Retention

In compliance with the ICH/GCP guidelines, the investigator/institution will maintain all CRF and all source documents that support the data collected from each subject, as well as all study documents as specified in ICH/GCP Section 8, Essential Documents for the Conduct of a Clinical Trial, and all study documents as specified by the applicable regulatory requirement(s). The investigator/institution will take measures to prevent accidental or premature destruction of these documents.

Essential documents must be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal

discontinuation of clinical development of the investigational product. These documents will be retained for a longer period if required by the applicable regulatory requirements or by an agreement with the sponsor. It is the responsibility of the sponsor to inform the investigator/institution as to when these documents no longer need to be retained.

If the responsible investigator retires, relocates, or for other reasons withdraws from the responsibility of keeping the study records, custody must be transferred to a person who will accept the responsibility. The sponsor must be notified in writing of the name and address of the new custodian. Under no circumstance shall the investigator relocate or dispose of any study documents before having obtained written approval from the sponsor.

If it becomes necessary for the sponsor or the appropriate regulatory authority to review any documentation relating to this study, the investigator/institution must permit access to such reports.

17.8. Monitoring

The sponsor will perform on-site monitoring visits as frequently as necessary. The monitor will record dates of the visits in a study site visit log that will be kept at the study site. The first post-initiation visit will be made as soon as possible after enrollment has begun. At these visits, the monitor will compare the data entered into the CRF with the source documents (eg, hospital/clinic/physician's office medical records). The nature and location of all source documents will be identified to ensure that all sources of original data required to complete the CRF are known to the sponsor and study-site personnel and are accessible for verification by the sponsor study-site contact. If electronic records are maintained at the study site, the method of verification must be discussed with the study-site personnel.

Direct access to source documents (medical records) must be allowed for the purpose of verifying that the recorded data are consistent with the original source data. Findings from this review will be discussed with the study-site personnel. The sponsor expects that, during monitoring visits, the relevant study-site personnel will be available, the source documents will be accessible, and a suitable environment will be provided for review of study-related documents. The monitor will meet with the investigator on a regular basis during the study to provide feedback on the study conduct.

There will be independent monitoring of the pharmacy and preparation of study vaccines by an unblinded monitor (independent study vaccine monitor, see Definitions of Terms); regular monitors will be blinded.

In addition to on-site monitoring visits, remote contacts can occur. It is expected that during these remote contacts, study-site personnel will be available to provide an update on the progress of the study at the site.

17.9. Study Completion/Termination

17.9.1. Study Completion/End of Study

The study is considered completed with the last visit for the last subject participating in the study. The final data from the study site will be sent to the sponsor (or designee) after completion of the final subject visit at that study site, in the time frame specified in the Clinical Trial Agreement.

17.9.2. Study Termination

The sponsor reserves the right to close the study site or terminate the study at any time for any reason at the sole discretion of the sponsor. Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the sponsor or investigator may include but are not limited to:

- Failure of the investigator to comply with the protocol, the requirements of the IEC/IRB or local health authorities, the sponsor's procedures, or GCP guidelines
- Inadequate recruitment of subjects by the investigator
- Discontinuation of further study vaccine development

17.10. On-site Audits

Representatives of the sponsor's clinical quality assurance department may visit the study site at any time during or after completion of the study to conduct an audit of the study in compliance with regulatory guidelines and company policy. These audits will require access to all study records, including source documents, for inspection. Subject privacy must, however, be respected. The investigator and study-site personnel are responsible for being present and available for consultation during routinely scheduled study-site audit visits conducted by the sponsor or its designees.

Similar auditing procedures may also be conducted by agents of any regulatory body, either as part of a national GCP compliance program or to review the results of this study in support of a regulatory submission. The investigator should immediately notify the sponsor if he/she has been contacted by a regulatory agency concerning an upcoming inspection.

17.11. Use of Information and Publication

All information, including but not limited to information regarding Ad26.ZEBOV and MVA-BN-Filo or the sponsor's operations (eg, patent application, formulas, manufacturing processes, basic scientific data, prior clinical data, formulation information) supplied by the sponsor to the investigator and not previously published, and any data, including exploratory

research data, generated as a result of this study, are considered confidential and remain the sole property of the sponsor and may be apportioned between the consortium members, if contemplated and as detailed in the Consortium Agreement. The investigator agrees to maintain this information in confidence and use this information only to accomplish this study, and will not use it for other purposes without the sponsor's prior written consent.

The investigator understands that the information developed in the study will be used by the sponsor in connection with the continued development of Ad26.ZEBOV and MVA-BN-Filo, and thus may be disclosed as required to other clinical investigators or regulatory agencies. To permit the information derived from the clinical studies to be used, the investigator is obligated to provide the sponsor with all data obtained in the study.

The results of the study will be reported in a Clinical Study Report generated by the sponsor and will contain data from all study sites that participated in the study as per protocol. Recruitment performance or specific expertise related to the nature and the key assessment parameters of the study will be used to determine a coordinating investigator. Results of exploratory analyses performed after the Clinical Study Report has been issued will be reported in a separate report and will not require a revision of the Clinical Study Report. Study subject identifiers will not be used in publication of results. Any work created in connection with performance of the study and contained in the data that can benefit from copyright protection (except any publication by the investigator as provided for below) shall be the property of the sponsor as author and owner of copyright in such work. Further details regarding ownership and access rights by consortium members to the data and results of the study are detailed in the Consortium Agreement.

Consistent with Good Publication Practices and International Committee of Medical Journal Editors guidelines, the sponsor and consortium members shall have the right to publish such primary (multicenter) data and information as detailed in the Consortium Agreement without approval from the individual investigators. The individual investigators have the right to publish study site-specific data after the primary data are published. Further details regarding publications by consortium members and individual investigators are detailed in the Clinical Trial Agreement and Consortium Agreement. The relevant publication sections of the Consortium Agreement will be shared with the investigator once executed by all the consortium partners. For multicenter study designs and substudy approaches, secondary results generally should not be published before the primary endpoints of a study have been published. Similarly, consortium members and investigators will recognize the integrity of a multicenter study by not submitting for publication data derived from the individual study site until the combined results from the completed study have been submitted for publication, within 12 months of the availability of the final data (tables, listings, graphs), or the sponsor confirms there will be no multicenter study publication. Authorship of publications resulting from this study will be based on the guidelines on authorship, such as those described in the Consortium Agreement and the Uniform Requirements for Manuscripts Submitted to Biomedical Journals, which state that the named authors must have made a significant contribution to the design of the study or analysis and interpretation of the data, provided critical review of the paper, and given final approval of the final version.

Registration of Clinical Studies and Disclosure of Results

The sponsor will register and disclose the existence of and the results of clinical studies as required by law.

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ATTACHMENTS

Attachment 1: Toxicity Tables for Use in Trials Enrolling Healthy Adults

The abbreviations used in the following tables are: ALT: alanine aminotransferase; aPTT: activated partial thromboplastin time; AST: aspartate aminotransferase; AV block: atrioventricular block; bpm: beats per minute; CK: creatine kinase; FEV_1: forced expiratory volume in 1 second; g: gram; HI: high; HPF: high power field; INR: international normalized ratio; IV: intravenous; LO: low; mEq: milliequivalent; mm Hg: millimeter of mercury; ms: millisecond; N: not graded; PT: prothrombin time; PTT: partial thromboplastin time; QTc: QT-interval corrected for heart rate; QTcB: Bazett's corrected QT interval; QTcF: Fridericia's corrected QT interval; RBC: red blood cell; Rx: therapy; s: second; U: unit; ULN: upper limit of normal

CLINICAL ADVERSE EVENTS

Grading scale used for clinical adverse events is adapted from the DMID Toxicity Tables (2014). For adverse events not included in the tables below, refer to the severity criteria guidelines in Section 12.1.3.

Cardiovascular	**Grade 1**	**Grade 2**	**Grade 3**
Arrhythmia		Asymptomatic, transient signs, no Rx required	Recurrent/persistent; symptomatic Rx required
Hemorrhage, blood loss	Estimated blood loss ≤100 mL	Estimated blood loss >100 mL, no transfusion required	Transfusion required
QTcF (Fridericia's correction) [a] or QTcB (Bazett's correction)	Asymptomatic, QTc interval 450-479 ms, *OR* Increase in interval<30 ms above baseline	Asymptomatic, QTc interval 480-499 ms, *OR* Increase in interval 30-60 ms above baseline[b]	Asymptomatic, QTc interval ≥500 ms, *OR* Increase in interval ≥60 ms above baseline
PR interval (prolonged)	PR interval 0.21-0.25 s	PR interval >0.25 s	Type II 2nd degree AV block *OR* Ventricular pause >3.0 s
Respiratory	**Grade 1**	**Grade 2**	**Grade 3**
Cough	Transient-no treatment	Persistent cough	Interferes with daily activities
Bronchospasm, acute	Transient; no treatment; FEV_1 71%-80% of peak flow	Requires treatment; normalizes with bronchodilator; FEV_1 60%-70% (of peak flow)	No normalization with bronchodilator; FEV_1 <60% of peak flow
Dyspnea	Does not interfere with usual and social activities	Interferes with usual and social activities, no treatment	Prevents daily and usual social activity or requires treatment

[a] Inclusion dependent upon protocol requirements.
[b] The Grade 2 increase in interval is changed from 30-50 ms to 30-60 ms since the original DMID Toxicity Tables (2014) did not cover the increase in interval between 50 and 60 ms.

Gastrointestinal	**Grade 1**	**Grade 2**	**Grade 3**
Nausea/vomiting	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities
Diarrhea	2-3 loose or watery stools or <400 g/24 hours	4-5 loose or watery stools or 400-800 g/24 hours	6 or more loose or watery stools or >800 g/24 hours or requires IV hydration
Reactogenicity	**Grade 1**	**Grade 2**	**Grade 3**
Local reactions			
Pain/tenderness at injection site	Aware of symptoms but easily tolerated; does not interfere with activity; discomfort only to touch	Notable symptoms; required modification in activity or use of medications; discomfort with movement	Incapacitating symptoms; inability to do work or usual activities; significant discomfort at rest
Erythema/redness[a]	2.5-5 cm	5.1-10 cm	>10 cm
Induration/swelling[b]	2.5-5 cm and does not interfere with activity	5.1-10 cm or interferes with activity	>10 cm or prevents daily activity
Itching at the injection site	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities
Systemic reactions			
Allergic reaction	Pruritus without rash	Localized urticaria	Generalized urticaria; angioedema or anaphylaxis
Headache	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities
Fatigue/malaise	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities
Myalgia	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities

[a] In addition to grading the measured local reaction at the greatest single diameter, the measurement should be recorded as a continuous variable.
[b] Induration/swelling should be evaluated and graded using the functional scale as well as the actual measurement.

Arthralgia	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities
Chills	Minimal symptoms; caused minimal or no interference with work, school or self-care activities	Notable symptoms; required modification in activity or use of medications; did not result in loss of work or cancellation of social activities	Incapacitating symptoms; required bed rest and/or resulted in loss of work or cancellation of social activities

LABORATORY TOXICITY GRADING

Grading scale used for lab assessments is based on 'FDA's Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials', but grade 3 and 4 are pooled below, consistent with the 3 scale toxicity grading used throughout the protocol. **If a laboratory value falls within the grading as specified below but also within the laboratory normal limits, the value is considered as normal.** For hemoglobin only the change from reference is used for the grading. The FDA table does not include toxicity grading for hematocrit, red blood cell counts or INR.

Blood, Serum, or Plasma Chemistries [a,b]	LO/HI/N [c]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Sodium (mEq/L or mmol/L)	LO	132-134	130-131	≤129
	HI	144-145	146-147	≥148
Potassium (mEq/L or mmol/L)	LO	3.5-3.6	3.3-3.4	≤3.2
	HI	5.1-5.2	5.3-5.4	≥5.5
Glucose (mg/dL)	LO	65-69	55-64	≤54
	HI [d]	100-110	111-125	>125
	HI [e]	110-125	126-200	>200
Blood urea nitrogen	HI	23-26 (mg/dL) or 8.3-9.4 (mmol/L)	27-31 (mg/dL) or 9.5- 11.2 (mmol/L)	>31 (mg/dL) or >11.2 (mmol/L)
Creatinine	N	1.5-1.7 (mg/dL) or 133-151 (µmol/L)	1.8-2.0 (mg/dL) or 152-177 (µmol/L)	>2.0 (mg/dL) or >177 (µmol/L)
Calcium (mg/dL)	LO	8.0-8.4	7.5-7.9	<7.5
	HI	10.5-11.0	11.1-11.5	>11.5
Magnesium (mg/dL)	LO	1.3-1.5	1.1-1.2	<1.1
Phosphorus (mg/dL)	LO	2.3-2.5	2.0-2.2	<2.0
Creatine kinase (CK) (mg/dL)	N	1.25-1.5xULN	1.6-3.0xULN	≥3.1xULN
Albumin (g/dL)	LO	2.8-3.1	2.5-2.7	<2.5
Total protein (g/dL)	LO	5.5-6.0	5.0-5.4	<5.0
Alkaline phosphatase (U/L)	N	1.1-2xULN	2.1-3xULN	>3xULN
AST (U/L)	HI	1.1-2.5xULN	2.6-5xULN	>5xULN
ALT (U/L)	HI	1.1-2.5xULN	2.6-5xULN	>5xULN
Bilirubin, serum total (mg/dL) – when accompanied by any increase in Liver Function Test		1.1-1.25xULN	1.26-1.5xULN	>1.5xULN
Bilirubin, serum total (mg/dL) – when Liver Function Test is normal		1.1-1.5xULN	1.6-2.0xULN	>2.0xULN
Amylase (U/L)	N	1.1x1.5ULN	1.6-2.0xULN	>2.0xULN
Lipase (U/L)	N	1.1x1.5ULN	1.6-2.0xULN	>2.0xULN

[a] Depending upon the laboratory used, references ranges, eligibility ranges and grading may be split out by sex and/or age.
[b] Cardiac troponin I increase by factor: >ULN-<2.0xULN; ≥2.0-<5.0xULN; ≥5.0xULN. (This footnote is added by the sponsor).
[c] Low, High, Not Graded.
[d] Fasting.
[e] Non-fasting.

Hematology	LO/HI/N [a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Hemoglobin (women) change from baseline (g/dL)	LO	Any decrease-1.5	1.6-2.0	>2.0
Hemoglobin (men) change from baseline (g/dL)	LO	Any decrease-1.5	1.6-2.0	>2.0
White blood cell count (cell/mm^3)	HI	10,800-15,000	15,001-20,000	>20,000
	LO	2,500-3,500	1,500-2,499	<1,500
Lymphocytes (cell/mm^3)	LO	750-1,000	500-749	< 500
Neutrophils (cell/mm^3)	LO	1,500-2,000	1,000-1,499	< 1000
Eosinophils (cell/mm^3)	HI	650-1500	1501-5000	> 5000
Platelets (cell/mm^3)	LO	125,000-140,000	100,000-124,000	<100,000
Coagulation				
Prothrombin time (PT, seconds)	HI	1.0-1.10xULN	1.11-1.20xULN	>1.20xULN
International normalized ratio (INR)[b]	HI	1.1-1.5xULN	1.6-2.0xULN	>2.0xULN
Partial thromboplastin time (PTT or aPTT, seconds)	HI	1.0-1.2xULN	1.21-1.4xULN	>1.4xULN
Fibrinogen (mg/dL)	HI	400-500	501-600	>600
	LO	150-200	125-149	<125
Urine				
Protein (dipstick)	HI	Trace	1+	2+
Glucose (dipstick)	HI	Trace	1+	2+
Blood (microscopic) - red blood cells per high power field (RBC/HPF)	HI	1-10	11-50	>50 and/or gross blood

[a] Low, High, Not Graded.
[b] For INR, the values in the table are based on the Division of AIDS Table for Grading the Severity of Adult and Pediatric Adverse Events, 2009.

VITAL SIGNS TOXICITY GRADING

Grading scale used for vital signs is according to DMID Toxicity Tables (2014)

Vital Signs	LO/HI/N [a]	Mild (Grade 1) [b]	Moderate (Grade 2)	Severe (Grade 3)
Fever (°C)[c]	HI	38.0-38.4	38.5-38.9	>38.9
Fever (°F)	HI	100.4-101.1	101.2-102.0	>102.0
Tachycardia - beats per minute	HI	101-115	116-130	>130 or ventricular dysrhythmias
Bradycardia - beats per minute	LO	50-54 or 45-50 bpm if baseline <60 bpm	45-49 or 40-44 bpm if baseline <60 bpm	<45 or <40 bpm if baseline <60 bpm
Hypertension (systolic) - mm Hg[d]	HI	141-150	151-160	>160
Hypertension (diastolic) - mm Hg	HI	91-95	96-100	>100
Hypotension (systolic) - mm Hg	LO	85-89	80-84	<80
Tachypnea - breaths per minute	HI	23-25	26-30	>30

[a] Low, High, Not Graded.
[b] If initial bound of grade 1 has gap from reference range or eligibility range, calculations based on the New England Journal of Medicine (NEJM) reference ranges.
[c] Oral temperature; no recent hot or cold beverages or smoking. A protocol should select either °C or °F for inclusion.
[d] Assuming subject is awake, resting, and supine; for adverse events, 3 measurements on the same arm with concordant results.

INVESTIGATOR AGREEMENT

I have read this protocol and agree that it contains all necessary details for carrying out this study. I will conduct the study as outlined herein and will complete the study within the time designated.

I will provide copies of the protocol and all pertinent information to all individuals responsible to me who assist in the conduct of this study. I will discuss this material with them to ensure that they are fully informed regarding the study vaccine, the conduct of the study, and the obligations of confidentiality.

Coordinating Investigator (where required):

Name (typed or printed): _____

Institution and Address: _____

_____

_____

_____

Signature: _____ Date: _____

(Day Month Year)

Principal (Site) Investigator:

Name (typed or printed): _____

Institution and Address: _____

_____

_____

_____

Telephone Number: _____

Signature: _____ Date: _____

(Day Month Year)

Sponsor's Responsible Medical Officer:

Name (typed or printed): Cynthia Robinson, MD

Institution: Janssen Vaccines & Prevention B.V.

Signature: [electronic signature appended at the end of the protocol] Date: _____

(Day Month Year)

Note: If the address or telephone number of the investigator changes during the course of the study, written notification will be provided by the investigator to the sponsor, and a protocol amendment will not be required.

LAST PAGE

SIGNATURES

Signed by	Date	Justification
Cynthia Robinson	25Apr2017, 12:58:43 PM, UTC	Document Approval