CLINICAL STUDY PROTOCOL

Randomized, Double-Blinded, Phase 2 Study to Assess Safety and Immunogenicity of Panblok H7 Vaccine at Three Antigen Dose Levels Adjuvanted with AS03® or MF59®

BARDA BP-I-17-002

Sponsor: Biomedical Advanced Research and Development Authority (BARDA)

Sponsor Contact:

Project Manager:

Medical Monitor:

Version of Protocol: 1.0

Date of Protocol: 03 July 2017

Previous Date and Version: N/A

CONFIDENTIAL

The study will be conducted according to the International Conference on Harmonisation (ICH) harmonised tripartite guideline E6 (R2): Good Clinical Practice (GCP).

Protocol Approval- Sponsor Signatory

Study Title	Randomized, Double-Blinded, Phase 2 Study to Assess Safety and Immunogenicity of Panblok H7 Vaccine at Three Antigen Dose Levels Adjuvanted with AS03® or MF59®
Protocol Number	BARDA BP-I-17-002
Protocol Version	Version 1.0
Protocol Date	03 July 2017

Protocol accepted and approved by:

[redacted]

Signature

July 5, 2017

Date

Investigator's Agreement

Study Title	Randomized, Double-Blinded, Phase 2 Study to Assess Safety and Immunogenicity of Panblok H7 Vaccine at Three Antigen Dose Levels Adjuvanted with AS03® or MF59®
Protocol Number	BARDA BP-I-17-002
Protocol Version	Version 1.0
Protocol Date	03 July 2017

I have read and understood all sections of the above referenced protocol and the current investigator's brochures for Panblok H7 vaccine, AS03 adjuvant, and MF59 adjuvant.

I agree to supervise all aspects of the protocol at my clinical research site and to conduct the clinical investigation in accordance with the Protocol and the ICH regulations. I will not make changes to the protocol before consulting with BARDA or implement protocol changes without institutional review board approval except to eliminate an immediate risk to subjects. I agree to administer study treatment only to subjects under my personal supervision or the supervision of a subinvestigator.

I will not supply the investigational product to any person not authorized to receive it. Confidentiality will be protected. Subject identity will not be disclosed to third parties or appear in any study reports or publications.

I will not disclose information regarding this clinical investigation or publish results of the investigation without authorization from BARDA.

_____

Printed Name of Investigator

_____

Signature of Investigator

_____

Date

Procedures in Case of Emergency

Table 1: Emergency Contact Information

Role in Study	Name	Address and Telephone Number
Sponsor Contact:	[redacted]	[redacted]
Medical Monitor	[redacted]	[redacted]

1. SYNOPSIS

Name of Sponsor/Company: Biomedical Advanced Research and Development Authority (BARDA)	
Name of Investigational Product: Panblok H7 influenza vaccine antigen prepared with AS03 or MF59 adjuvant	
Name of Active Ingredients: Antigen: Panblok H7, derived from A/Guangdong/17SF003/2016 (H7N9) influenza virus strain Adjuvants: AS03 and MF59	
Title of Study: Randomized, Double-Blinded, Phase 2 Study to Assess Safety and Immunogenicity of Panblok H7 Vaccine at Three Antigen Dose Levels Adjuvanted with AS03® or MF59®	
Protocol Number: BP-I-17-002	
Version Number: 1.0	
Study center(s): Approximately four clinical sites in the United States (US)	
Study period (years): Estimated date first subject enrolled: August 2017 Estimated date last subject completed: October 2018	**Phase of development:** 2

Study Objectives:

Primary Objectives

Safety

- To assess the safety and reactogenicity for 8 days postvaccination, inclusive of the vaccination day, (Day 1 through Day 8 and Day 29 through Day 36) of 3 different antigen dosages of Panblok H7 vaccine given with AS03 or MF59 adjuvant (henceforth referred to as "study vaccines") administered intramuscularly (IM) on Days 1 and 29, as determined by solicited local and systemic reactogenicity symptoms.

Immunogenicity

- To assess the serum hemagglutination-inhibition (HAI) antibody seroprotection rate on Day 50 of 3 different antigen dosages of study vaccines administered IM on Days 1 and 29.

Secondary Objectives

Safety

- To assess the occurrence of unsolicited AEs, SAEs and medically attended adverse events (MAAEs) including a subset of specific potentially immune-mediated medical conditions (PIMMCs) in the 6 treatment groups for 13 months after the first dose of study vaccine.

Immunogenicity

- To assess the serum HAI antibody titers, seroprotection rates, and seroconversion rates of 3 different antigen dosages of the study vaccines through Day 212.
- To assess the serum microneutralization (MN) antibody titers and seroconversion rates of

3 different antigen dosages of the study vaccines through Day 212.

Methodology:

This is a randomized, double-blinded, phase 2 study to assess safety and immunogenicity of Panblok H7 vaccine at three antigen dose levels (3.75, 7.5, and 15 μg) adjuvanted with AS03 or MF59. The main purpose of this study is to assess the safety and ability of the recombinant Panblok H7 influenza vaccine adjuvanted with AS03 or MF59, to generate an immune response after 2 doses separated by 28 days in healthy males and nonpregnant females, aged 18 to 49 years, inclusive.

Number of subjects (planned): Approximately 360 healthy subjects (approximately 60 subjects per treatment group). There will be a total of 6 treatment groups.

Diagnosis and main criteria for inclusion:

Subjects will be randomized to study treatment only if they meet all of the inclusion criteria and none of the exclusion criteria. In addition, in order to receive the second vaccination, subjects must again have all of the inclusion and exclusion criteria assessed; if the subject no longer meets eligibility criteria, the investigator, in consultation with the medical monitor in cases of uncertainty, must determine if the subject should receive the second vaccination or be terminated early from study vaccination. Subjects who do not receive the second vaccination will be followed as defined in Section 6.4.1.

Deviations from the inclusion and exclusion criteria are not allowed because they can potentially jeopardize the scientific integrity of the study, regulatory acceptability, or subject safety. Therefore, adherence to the criteria as specified in the protocol is essential.

Subject Inclusion Criteria

1. Male or nonpregnant female 18 to 49 years of age, inclusive, at the time of the first study vaccination.
2. Provide written informed consent prior to the initiation of any study-related procedures.
3. Are able to understand and comply with planned study procedures.
4. Have a stable health status based on site investigator's clinical judgment, as established by physical examination, vital signs, and medical history.
5. Have access to a consistent and reliable means of telephone contact, which may be in the home, workplace, or by personal mobile electronic device.
6. Agree to stay in contact with the study site for the duration of the study, have no current plans to move from the study area, and agree to provide updated contact information as necessary.

Subject Exclusion Criteria

1. Have had a prior severe reaction to any influenza vaccine or have a known allergy to squalene-based adjuvants.
2. Women who are pregnant or breast feeding. Women of childbearing potential must have a negative urine pregnancy test at screening and within 24 hours prior to each vaccination.

   Women of childbearing potential are defined as postmenarcheal and premenopausal females capable of becoming pregnant. This does not include females who meet any of the following conditions: menopausal >12 months, tubal ligation >12 months, bilateral salpingo-

oophorectomy, or hysterectomy.

3. Women of childbearing potential who refuse to use an acceptable method of birth control from screening to Day 50 (Visit 7) or, if sexually active with a male partner, who have not used a reliable birth control method during the 2 months prior to screening.

   Adequate contraception is defined as a contraceptive method with a failure rate of less than 1% per year when used consistently and correctly and when applicable, in accordance with the product label, for example: abstinence from penile-vaginal intercourse; oral contraceptives, either combined or progestogen alone; injectable progestogen; implants of etonogestrel or levonorgestrel; estrogenic vaginal ring; percutaneous contraceptive patches; intrauterine device or intrauterine system; male partner sterilization at least 6 months prior to the female subject's Screening Visit, and this male is the sole partner for that subject (the information on the male partner's sterility can come from the site personnel's review of the subject's medical records or interview with the subject on her medical history); male condom combined with a vaginal spermicide (foam, gel, film, cream, or suppository); male condom combined with a female diaphragm, either with or without a vaginal spermicide (foam, gel, film, cream, or suppository).

4. Have immunosuppression as a result of an underlying illness or treatment, or use of anticancer chemotherapy or radiation therapy (cytotoxic) within the preceding 36 months, or plans to receive immunosuppressive therapy/cytotoxic treatment during study participation.

5. Have an active neoplastic disease or a history of any hematologic malignancy. However, subjects with superficial skin cancer who do not require intervention other than local excision are not excluded.

6. Have long-term use (≥14 consecutive days) of glucocorticoids including oral or parenteral prednisone or prednisone equivalent (>20 mg total dose per day) or high-dose inhaled steroids (>800 µg/day of beclomethasone dipropionate or equivalent) within 1 month prior to screening in this study. However, subjects on low-dose inhaled steroids (≤800 µg/day of beclomethasone dipropionate or equivalent) or topical steroids are not excluded.

7. History of schizophrenia, bipolar disease, psychosis, or severe personality disorder.

8. History of hospitalization for psychiatric illness, attempted suicide, or having been deemed a danger to self or others within the past 10 years.

9. Have received immunoglobulin or other blood product (with the exception of Rho[D] immune globulin) within the 3 months prior to screening in this study.

10. Have received any live vaccines within 4 weeks or inactivated or recombinant protein vaccines within 2 weeks prior to screening in this study or plan to receive such vaccines (including seasonal influenza vaccines) from screening through 21 days following the second dose of the study vaccine (Screening Visit through Day 50).

11. Have an acute or chronic medical condition that, in the opinion of the site investigator, would render vaccination unsafe or would interfere with the evaluation of responses. This includes all PIMMCs such as Guillain-Barré syndrome, narcolepsy, and current or history of autoimmune or chronic inflammatory disease (as listed in Appendix 3).

12. Have an acute illness, including body temperature greater than 100.4°F, at screening, immediately prior to each vaccination or, per subject report, within 3 days prior to each

vaccination in this study.

13. Received an experimental agent (vaccine, drug, biologic, device, blood product, or medication) within 1 month prior to screening in this study or expect to receive an experimental agent during the study period.
14. Are participating or plan to participate in another interventional clinical trial (either active or follow-up phase) during the study period.
15. Participated in an A(H7) influenza vaccine study in the past or have a history of A(H7) influenza infection prior to vaccination in this study.
16. Have known human immunodeficiency virus, hepatitis B, or hepatitis C infection (based on medical history).
17. Have a history of alcohol or drug abuse in the last 5 years.
18. Have a body mass index >35 kg/m^2.
19. Have a first degree relative with narcolepsy.
20. Have any laboratory test result or clinical findings (including vital signs) that singly or in combination are likely to unfavorably alter the risk-benefit of subject participation or to confound study safety or immunogenicity results. Subjects cannot be rescreened based on abnormal laboratory test results.
21. Alanine aminotransferase or aspartate aminotransferase (AST) >2 times the upper limit of normal (ULN), or bilirubin >1.5 times the ULN unless isolated Gilbert's syndrome. Subjects cannot be rescreened based on abnormal laboratory test results.

Investigational product, dosage and mode of administration:

The Panblok H7 vaccine will be administered at three antigen dose levels (3.75, 7.5, and 15 μg) adjuvanted with AS03 or MF59. Two doses of adjuvanted vaccine separated by 28 days will be administered to approximately 360 subjects who will be randomly assigned in a 1:1:1:1:1:1 ratio to 1 of 6 treatment groups.

The study vaccines will be prepared by mixing recombinant Panblok H7 influenza vaccine antigen 1:1 with either MF59 or AS03 adjuvant prior to administration. The study vaccine (0.5 mL) should be administered by IM injection in the deltoid muscle of the arm within 30 minutes of mixing. Each vaccination will be given in a different arm.

Duration of treatment:

The expected study duration is approximately 13.5 months per subject.

Reference therapy, dosage and mode of administration: Not applicable

Criteria for evaluation:

Immunogenicity

- Serum hemagglutination-inhibition (HAI) antibody titers
- Seroprotection based on serum HAI antibody titers, defined as an HAI antibody titer ≥1:40
- Seroconversion based on serum HAI antibody titers, defined as either a prevaccination HAI

titer <1:10 and a post-vaccination HAI titer ≥1:40, or a prevaccination HAI titer ≥1:10 and a minimum 4-fold rise in postvaccination HAI titer.

- Serum microneutralization (MN) antibody titers
- Seroconversion based on serum MN antibody titers, defined as either a prevaccination MN titer <1:10 and a postvaccination MN titer ≥1:40, or a prevaccination MN titer ≥1:10 and a minimum 4-fold rise in postvaccination MN titer.

Safety

Safety will be evaluated by assessing frequency and severity of solicited local and systemic reactogenicity symptoms occurring within 8 days of each vaccination and treatment-emergent SAEs, MAAEs, PIMMCs, and unsolicited AEs occurring during study participation. Safety assessments will also include physical examination, vital signs, and clinical laboratory tests (hematology and chemistry).

Study Endpoints:

Primary Endpoints

There are two primary endpoints for this study.

- All solicited local and systemic reactogenicity symptoms occurring within 8 days of each vaccination, inclusive of the vaccination day.
- Seroprotection at Day 50 based on serum HAI antibody titers.

Secondary Endpoints

Secondary Safety Endpoints

- All treatment-emergent SAEs occurring during study participation.
- All treatment-emergent MAAEs occurring during study participation.
- All treatment-emergent PIMMCs occurring during study participation.
- All treatment-emergent unsolicited AEs occurring during study participation.

Secondary Immunogenicity Endpoints

- Serum HAI antibody titers at Screening and Days 29, 50, 121, and 212.
- Serum MN antibody titers at Screening and Days 29, 50, 121, and 212.
- Seroprotection at Screening and Days 29, 121, and 212 based on serum HAI antibody titers.
- Seroconversion at Days 29, 50, 121, and 212 based on serum HAI antibody titers.
- Seroconversion at Days 29, 50, 121, and 212 based on serum MN antibody titers.

Statistical methods:

Analysis Plan

Due to the exploratory nature of this study, no inferential analyses are planned. Descriptive statistics (such as medians, quartiles, and ranges for continuous data and percentages for categorical data) will be used to summarize subject characteristics, safety, and immunogenicity parameters. These summaries will be presented overall and separately for the subjects in the different treatment groups, as well as

pooled groups by antigen dose level and by adjuvant. Details of the statistical analyses, methods, and data conventions will be described in the Statistical Analysis Plan (SAP). Details of the primary and secondary analyses for safety and immunogenicity assessments are described in Section 12.1.4.2 and Section 12.1.4.3.

Interim Analysis

An interim analysis will be performed based on cumulative immunogenicity and safety data through Day 50 for all subjects. At the interim analysis, the study database (all data through Day 50) will be monitored and cleaned per the Data Management Plan. Data for the interim analysis will be unblinded solely at the group level, thus, the blinding at the subject level will be maintained until the study unblinding of all data through Day 212 for the clinical study report. All primary and secondary endpoint analyses will be performed for the interim analysis as specified in Section 12.1.4.2 and Section 12.1.4.3.

Ad Hoc Safety Monitoring Committee (SMC) Meeting

There are no formal or planned SMC meetings. A SMC review will only occur when a stopping rule is met and for any immediate safety concerns observed, as defined in Section 6.4.2. Data displays for any SMC review will be generated by an unblinded statistician. Safety analyses for this study will be descriptive rather than inferential. Detailed listings and summary tabulations will be generated as specified in the SMC charter. The safety analyses will be completed using the safety population.

Final Analyses

A clinical study report will be written to include all safety and immunogenicity data through Day 212. For this Day 212 analysis, the study database (all data through Day 212) will be monitored and cleaned, per the Data Management Plan. Study data will be unblinded to prepare the study report. Safety data collected after Day 212 will be provided as a supplemental report to the clinical study report. Cumulative summaries of all SAEs, MAAEs, and PIMMCs occurring from Day 1 through the end of study will be included in the supplemental report.

Analysis Populations

Safety Population

The safety population will include all subjects who are randomized and receive at least one vaccination. Each subject will be analyzed as part of the treatment group corresponding to the actual treatment received. The safety population will be used for all safety analyses.

Immunogenicity Full Analysis Population

The immunogenicity full analysis population will include all subjects who are randomized, receive at least one vaccination, and have determinate assay results at any postvaccination visit. Each subject will be analyzed as part of the treatment group assigned by randomization, regardless of the treatment actually received. All immunogenicity analyses will be performed on the immunogenicity full analysis population.

Immunogenicity Per Protocol Population

The immunogenicity per protocol population will include all subjects who meet the following criteria:

- Are in the immunogenicity full analysis population.

- Received a full dose of vaccine at Day 1 and a full dose of vaccine at Day 29.
- Received the correct treatment as assigned by randomization.
- Have no other major protocol deviations that may have an impact on immunogenicity assessments.
- Have determinate assay results at the Day 50 visit.

All immunogenicity analyses will also be performed on the immunogenicity per protocol population by the treatment group actually received.

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3. LIST OF ABBREVIATIONS

Abbreviation or Specialist Term	Explanation
AE	adverse event
ASPR	Assistant Secretary for Preparedness and Response
AST	aspartate aminotransferase
BARDA	Biomedical Advanced Research and Development Authority
CBC	complete blood count
CFR	Code of Federal Regulations
CI	confidence interval
CVV	candidate vaccine virus
eCRF	electronic case report form
ET	early termination
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GMT	geometric mean titer
HA	hemagglutinin
HAI	hemagglutination-inhibition
HHS	Health and Human Services
ICF	informed consent form
ICH	International Conference on Harmonisation
IM	intramuscular
IRB	institutional review board
IWRS	interactive web response system
MAAE	medically attended adverse event
MedDRA	Medical Dictionary for Regulatory Activities
MN	microneutralization
MOP	manual of procedures
NPIVS	National Pre-Pandemic Influenza Vaccine Stockpile
OTC	over-the-counter
PIMMC	potentially immune-mediated medical condition
rHA	recombinant hemagglutinin
SAE	serious adverse event

Abbreviation or Specialist Term	Explanation
SAP	Statistical Analysis Plan
SAR	suspected adverse reaction
SE	stable oil-in-water emulsion
SMC	safety monitoring committee
SUSAR	serious and unexpected suspected adverse reaction
ULN	upper limit of normal
US	United States
WHO	World Health Organization

4. INTRODUCTION

4.1. Background

In March 2013, zoonotic infections with novel avian-origin influenza A(H7N9) virus were reported in China, causing severe lower respiratory tract disease in humans[1,2]. Since then, influenza A(H7N9) virus was found to be circulating in poultry in China, and sporadic human infections have been reported annually between fall and early spring[3]. During the 5th epidemic in 2016-2017, an unprecedented number of influenza A(H7N9) cases have been identified, and genetic and antigenic analyses indicate that more than 90% of the H7N9 viruses have evolved into an antigenically distinct group designated Yangtze River Delta with reduced cross-reactivity with existing candidate vaccine viruses (CVVs) made in 2013, prompting the World Health Organization (WHO) to update CVV recommendations[4]. Per the Hong Kong Centre of Health Protection, since the 5th wave started on October 1, 2016, through June 5, 2017, a total of 706 new cases of the H7N9 illness occurred in mainland China, bringing the total number of human cases of H7N9 illness to 1512.

While there is no evidence of sustained transmission, limited human-to-human spread of A(H7N9) virus has been identified. Although since 2013, the influenza A(H7N9) viruses have been characterized as having low pathogenicity, viruses highly pathogenic for poultry emerged in February 2017 and caused several human infections[3]. In fact, of the influenza viruses that are of special concern to public health, H7N9 is rated by the Influenza Risk Assessment Tool as having the greatest potential to cause a pandemic, as well as potentially posing the greatest risk to severely impact public health[5, 6].

4.2. Study Rationale

The Department of Health and Human Services (HHS) continuously monitors pandemic risk and prepares to respond to the threat of novel influenza outbreaks in the United States (US). To this end, the Biomedical Advanced Research and Development Authority (BARDA), within the Office of the Assistant Secretary for Preparedness and Response (ASPR), has established and maintains the National Pre-Pandemic Influenza Vaccine Stockpile (NPIVS) comprised of AS03 and MF59 adjuvants and pre-pandemic H5N1 and H7N9 bulk antigens. To shorten timelines to make vaccines available to immunize the US population, HHS has supported expansion of domestic vaccine manufacturing capacity, including the use of adjuvanted vaccines, and the licensure of seasonal cell-based and recombinant influenza vaccines. While adjuvanted egg- and cell-based vaccine antigens have been evaluated with adjuvants from the NPIVS (AS03 and MF59) to support their deployment for pandemic mitigation under Emergency Use Authorization by the FDA, safety and dose-sparing evidence of these adjuvants with recombinant H7 hemagglutinin (HA) antigens is lacking. For these reasons, BARDA seeks to clinically evaluate the safety and immunogenicity of adjuvanted influenza H7 vaccine utilizing recombinant protein technology that can be used to respond quickly to a new emerging strain. By pairing the recombinant protein vaccine with adjuvants in the NPIVS, the study will provide critical insights for the US government to develop a response strategy for a pandemic emergency.

This is a randomized, double-blinded, phase 2 study to assess safety and immunogenicity of Panblok H7 vaccine at three antigen dose levels (3.75, 7.5, and 15 µg) adjuvanted with AS03 or MF59. The main purpose of this study is to assess the safety and ability of the recombinant Panblok H7 influenza vaccine adjuvanted with AS03 or MF59 to generate an immune response after 2 doses separated by 28 days in healthy males and nonpregnant females, aged 18 to 49 years, inclusive.

The dosing interval for this study (28 days) is expanded from the traditional pre-pandemic studies that utilized a 21-day interval. Published adjuvanted H5N1 clinical vaccine studies indicate that expanding the dosing interval may result in improved immune responses[7,10]. Additionally, a 28-day or longer dosing interval harmonizes with standard clinical practice of many licensed vaccines[8,9].

4.3. Previous Clinical Studies

4.3.1. Antigen

No clinical data are available for Panblok H7, derived from A/Guangdong/17SF003/2016 (H7N9) influenza virus strain. Pandemic A/H5 recombinant HA (rHA) and A/H7 rHA clinical information is provided from previous studies PSC25[11] and PSC26[12].

Study PSC25 evaluated recombinant Panblok (A/H5 rHA) in subjects 18-49 years of age. Two doses of unadjuvanted 7.5 µg vaccine administered at Days 0 and 21 were poorly immunogenic at Day 42. Two doses of H5 rHA antigen at 3.75, 7.5 and 15 µg adjuvanted with 2% stable oil-in-water emulsion (SE) administered at Days 0 and 21 were immunogenic and elicited seroconversion rates of 70%, 76%, and 83%, respectively, and hemagglutination-inhibition (HAI) geometric mean titers (GMTs) of 76.1, 77.5, and 121.7, respectively, at Day 42. The Center for Biologics Evaluation and Research criterion for postimmunization titer ≥1:40 (i.e., the lower bound of the two-sided 95% confidence interval [CI] for the percentage of subjects achieving an HAI antibody titer ≥1:40 should be ≥70%) was only met by the 15 µg adjuvanted antigen dosage group. Study PSC25 revealed no product-related serious adverse events (SAEs) or potentially immune-mediated medical conditions (PIMMCs). Elicited adverse events (AEs) following adjuvanted (2% SE) or unadjuvanted H5 rHA vaccine were generally mild to moderate with 0 to 3% experiencing severe events [13].

In study PSC26, the safety and immunogenicity of Panblok H7 derived from H7 A/Anhui/1/2013 was evaluated at 3 dose levels administered on Days 0 and 21. This study was designed as a two-stage, adaptive design Phase 1/2 randomized, observer-blind, multi-center clinical trial intended to enroll ~1100 healthy adults 18 years of age and older. Subjects in Stage 1 were stratified to age categories 18 – 64 and ≥65 years of age and randomized in a 1:1:1:1 ratio (N≈100 subjects/group) to receive 7.5 µg + SE 2.0%, 15 µg + SE 2.0%, 30 µg + SE 2.0%, or 30 µg unadjuvanted. A planned interim analysis of all available safety and immunogenicity data in Stage 1 subjects was performed after completion of the HAI assays from Day 42. In this study, there were no safety concerns observed, but immunogenicity was insufficient to support further enrollment into Stage 2 of the study.

4.3.2. Adjuvants

No clinical data is available for A/H7 rHA vaccines adjuvanted with AS03 or MF59.

Both AS03 and MF59-adjuvanted vaccines have been licensed in the US (e.g., AS03 in the Influenza A (H5N1) Virus Monovalent Vaccine, Adjuvanted vaccine [14] and MF59 in the FLUAD™ vaccine[15]). There are no major safety concerns associated with the use of these adjuvants; however, there is epidemiological evidence of an association between influenza vaccine adjuvanted with AS03 and narcolepsy. In studies of influenza A(H1N1) vaccine adjuvanted with AS03, there was a small increased risk of narcolepsy in subjects who received adjuvanted vaccine; however, narcolepsy developed almost exclusively in the pediatric population vaccinated with influenza A(H1N1) adjuvanted with AS03 (Pandemrix, GSK, Wavre, Belgium, produced in Dresden, Germany) and primarily in Scandinavian countries[16,17]. A small study to assess the risk of narcolepsy following administration of a similar vaccine adjuvanted with AS03 (Arepanrix, GSK, Wavre, Belgium, produced in Quebec City, Canada) was performed in Quebec, Canada[18]. The Canadian study showed a small risk of developing narcolepsy after receiving the AS03 adjuvanted A(H1N1) pandemic vaccine manufactured in Quebec. This risk occurred primarily in people less than 20 years of age and was much less than that seen in some European countries.

There was also a study conducted by the Centers for Disease Control "SOMNIA study" that assessed the risk of narcolepsy following administration of adjuvanted 2009 H1N1 pandemic vaccines. Comparison of incidence rates of narcolepsy before, during, and after the use of adjuvanted 2009 pandemic influenza vaccines provided no evidence outside of the signaling country Sweden[19].

5. STUDY OBJECTIVES

5.1. Primary Objectives

5.1.1. Safety

- To assess the safety and reactogenicity for 8 days postvaccination, inclusive of the vaccination day, (Day 1 through Day 8 and Day 29 through Day 36) of 3 different antigen dosages of Panblok H7 vaccine given with AS03 or MF59 adjuvant (henceforth referred to as "study vaccines") administered intramuscularly (IM) on Days 1 and 29, as determined by solicited local and systemic reactogenicity symptoms.

5.1.2. Immunogenicity

- To assess the serum HAI antibody seroprotection rate on Day 50 of 3 different antigen dosages of study vaccines administered IM on Days 1 and 29.

5.2. Secondary Objectives

5.2.1. Safety

- To assess the occurrence of unsolicited AEs, SAEs and medically attended adverse events (MAAEs) including a subset of specific PIMMCs in the 6 treatment groups for 13 months after the first dose of study vaccine.

5.2.2. Immunogenicity

- To assess the serum HAI antibody titers, seroprotection rates, and seroconversion rates of 3 different antigen dosages of the study vaccines through Day 212.
- To assess the serum microneutralization (MN) antibody titers and seroconversion rates of 3 different antigen dosages of the study vaccines through Day 212.

6. INVESTIGATIONAL PLAN

6.1. Overall Study Design

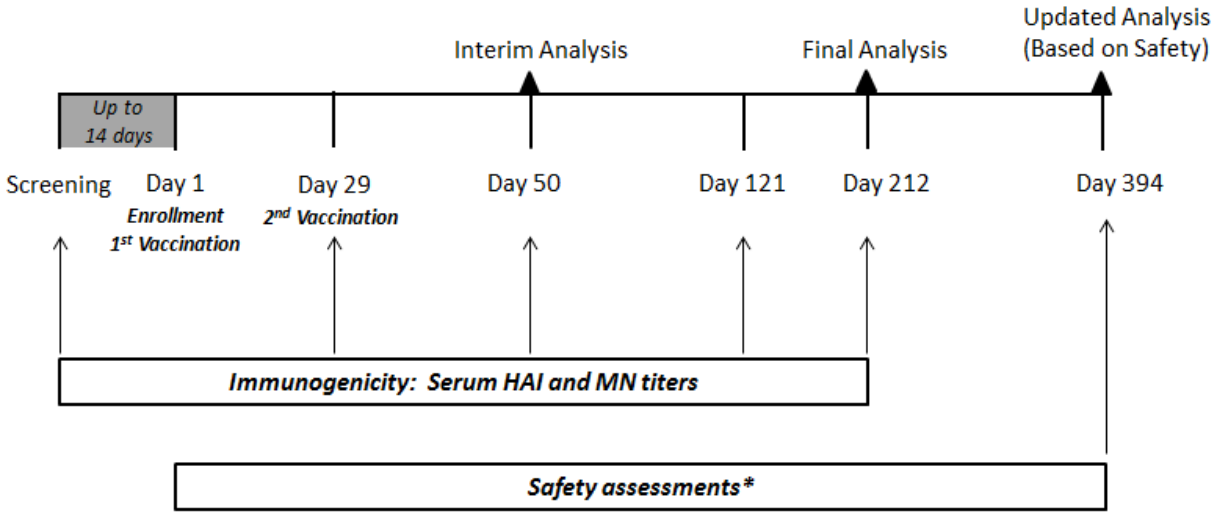
This phase 2 study will be conducted as a randomized, double-blinded, multi-site study to assess the safety and immunogenicity of Panblok H7 vaccine at 3 ascending antigen dose levels adjuvanted with AS03 or MF59. The main purpose of this study is to assess the safety and ability of a Panblok H7 influenza vaccine adjuvanted with AS03 or MF59 to generate an immune response after 2 doses separated by 28 days. Figure 1 presents a diagram of the overall study design.

The target population consists of healthy males and nonpregnant females, aged 18 to 49 years, inclusive (Section 7.1). The proposed enrollment for this study is approximately 360 healthy subjects divided into 6 treatment groups (approximately 60 subjects per group), at ascending dosages (3.75, 7.5, and 15 µg) of Panblok H7 vaccine adjuvanted with AS03 or MF59 and administered IM (Table 2).

The expected study duration is approximately 13.5 months per subject.

The Schedule of Assessments can be found in Appendix 1.

Figure 1: Overall Study Design



*AEs will be assessed from the time the subject receives the first vaccination through Day 50 as follows:

- Solicited AEs (i.e., solicited local and systemic reactogenicity symptoms) will be collected for 8 days after each vaccination, inclusive of the vaccination day (i.e., from Day 1 to Day 8 and from Day 29 to Day 36)
- Unsolicited AEs will be collected from the time the subject received the first vaccination (Day 1) until up to 21 days after the second vaccination (Day 50)

SAEs, MAEs and PIMMCs will be assessed from the time the subject receives the first vaccination (Day 1) until exit from the study (Day 394).

AE = adverse event; HAI = hemagglutination inhibition; MAAEs = medically attended adverse events; MN = microneutralization; PIMMCs = potentially immune-mediated medical conditions; SAEs = serious adverse events

6.2. Number of Subjects

The proposed enrollment for this study is approximately 360 healthy subjects (approximately 60 subjects per treatment group). There will be a total of 6 treatment groups (Table 2).

6.3. Treatment Assignment

Two doses of adjuvanted vaccine separated by 28 days will be administered IM to approximately 360 subjects who will be randomly assigned in a 1:1:1:1:1:1 ratio to 1 of 6 treatment groups as presented in Table 2 below.

Table 2: Treatment Groups

Treatment Group	Approximate Number of Subjects	Hemagglutinin Antigen (µg)	Adjuvant	Injection Volume (mL)
A	60	3.75	AS03	0.5
B	60	7.5	AS03	0.5
C	60	15	AS03	0.5
D	60	3.75	MF59	0.5
E	60	7.5	MF59	0.5
F	60	15	MF59	0.5

6.4. Individual Subject Dosing and Study Stopping Rules

6.4.1. Early Termination of Dosing for an Individual Subject

Vaccinations in a subject are to be stopped if the subject experiences any of the following. In situations of ambiguity or uncertainty, the investigator is encouraged to discuss the situation and any questions with the medical monitor before making a decision on subject disposition.

1. ≥3 occurrences of Grade 3 or higher AEs assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation (Section 11.3.3, Section 11.4, and Appendix 2).
2. Any Grade 3 or higher MAAE assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation (Section 11.3.3, Section 11.4, and Appendix 2).
3. Any SAE or PIMMC assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation (Section 11.4 and Appendix 3).
4. Any Grade 3 or higher abnormal laboratory test result assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation (Section 11.3.3, Section 11.4, and Appendix 2).
5. Anaphylaxis.
6. Grade 3 or higher allergic reaction, as judged by the investigator (such as, but not limited to, difficulty breathing, rapid increase in heart rate, dizziness, nausea, evidence of urticaria, or periorbital swelling) assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation.

7. Intolerable AE.
8. Pregnancy.
9. Subject no longer meets eligibility criteria, such that the safety of the subject may be compromised by continued participation or interpretation of the subject's subsequent study data are likely to be significantly compromised.
10. Any other condition that, in the judgment of the investigator, would make further vaccination unsafe or render the subject unable to comply with protocol-mandated safety follow-up.

Subjects who do not receive the second vaccination will continue to be followed for immunogenicity through the Day 212 Visit and for safety through the Day 394 visit. Participants who become pregnant during the study will only be followed for safety and will not provide blood for the immunogenicity assays. Additional clinical laboratory assessments will only be completed during the safety follow-up when determined to be clinically necessary by the investigator in consultation with the medical monitor.

6.4.2. Study Suspension and Safety Monitoring Committee Responsibility

The blinded medical monitor will assess cumulative safety information for this study per the Medical Monitoring Plan and advise BARDA should he become aware that any of the below safety findings have occurred. An unblinded statistician or designee will assist with determining whether stopping rule #7 was met. The occurrence of 1 or more of these findings will result in immediate suspension of further enrollment and study vaccine administration (as applicable per the study timeline) pending urgent review (within 1 week) of the safety data by the Safety Monitoring Committee (SMC).

A safety review will be triggered if any of the below occur:

1. Five or more subjects have generalized urticaria.
2. One or more subjects have injection site ulceration, abscess, or necrosis.
3. One or more subjects have Grade 3 or worse laryngospasm, bronchospasm, or anaphylactic shock within 24 hours of vaccine administration.
4. One or more subjects are withdrawn from further vaccination due to meeting vaccination termination criteria #1-4 in Section 6.4.1.
5. One or more subjects are withdrawn from further vaccination or from the study by the investigator due to a Grade 3 or higher AE assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation.
6. One or more subjects experience an SAE or PIMMC assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation.
7. Any Grade 3 or higher abnormality in the same pre-specified laboratory parameter (Section 11.1.5) occurring in ≥2 subjects within an individual treatment group or ≥4 subjects across treatment groups assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation.

8. Five or more subjects across treatment groups experience a Grade 3 or higher AE or MAAE of the same type (as categorized by Medical Dictionary for Regulatory Activities [MedDRA] preferred term) assessed as possibly related, probably related, or related to study vaccine without a plausible alternative explanation.
9. A pattern of significant symptoms, physical findings, or laboratory abnormalities that, although individually minor, collectively represent a safety concern in the opinion of the investigator, medical monitor, or BARDA.

Following their unblinded review of available safety data, the SMC will make 1 or more of the following recommendations:

- All vaccinations should resume.
- Specific treatment groups should be discontinued while other treatment groups should resume vaccinations.
- No vaccinations should resume.
- Modifications to study conduct (e.g. additional safety laboratory assessments).

BARDA retains the right to suspend or end the study or to discontinue specific treatment groups at any time. In case of premature termination or suspension and safety review of the study, BARDA will promptly inform the investigators and regulatory authorities as appropriate of the termination or suspension and the reason for termination/suspension.

7. SELECTION AND WITHDRAWAL OF SUBJECTS

7.1. Selection of Study Population

Subjects will be randomized to study treatment only if they meet all of the inclusion criteria and none of the exclusion criteria. In addition, in order to receive the second vaccination, subjects must again have all of the inclusion and exclusion criteria assessed; if the subject no longer meets eligibility criteria, the investigator, in consultation with the medical monitor in cases of uncertainty, must determine if the subject should receive the second vaccination or be terminated early from study vaccination. Subjects who do not receive the second vaccination will be followed as defined in Section 6.4.1.

Deviations from the inclusion and exclusion criteria are not allowed because they can potentially jeopardize the scientific integrity of the study, regulatory acceptability, or subject safety. Therefore, adherence to the criteria as specified in the protocol is essential.

7.1.1. Subject Inclusion Criteria

1. Male or nonpregnant female 18 to 49 years of age, inclusive, at the time of the first study vaccination.
2. Provide written informed consent prior to the initiation of any study-related procedures.
3. Are able to understand and comply with planned study procedures.
4. Have a stable health status based on site investigator's clinical judgment, as established by physical examination, vital signs, and medical history.
5. Have access to a consistent and reliable means of telephone contact, which may be in the home, workplace, or by personal mobile electronic device.
6. Agree to stay in contact with the study site for the duration of the study, have no current plans to move from the study area, and agree to provide updated contact information as necessary.

7.1.2. Subject Exclusion Criteria

1. Have had a prior severe reaction to any influenza vaccine or have a known allergy to squalene-based adjuvants.
2. Women who are pregnant or breast feeding. Women of childbearing potential must have a negative urine pregnancy test at screening and within 24 hours prior to each vaccination.

   Women of childbearing potential are defined as postmenarcheal and premenopausal females capable of becoming pregnant. This does not include females who meet any of the following conditions: menopausal >12 months, tubal ligation >12 months, bilateral salpingo-oophorectomy, or hysterectomy.

3. Women of childbearing potential who refuse to use an acceptable method of birth control from screening to Day 50 (Visit 7) or, if sexually active with a male partner, who have not used a reliable birth control method during the 2 months prior to screening.

   Adequate contraception is defined as a contraceptive method with a failure rate of less than 1% per year when used consistently and correctly and when applicable, in accordance with the product label, for example: abstinence from penile-vaginal intercourse; oral contraceptives, either combined or progestogen alone; injectable progestogen; implants of etonogestrel or levonorgestrel; estrogenic vaginal ring; percutaneous contraceptive patches; intrauterine device or intrauterine system; male partner sterilization at least 6 months prior to the female subject's Screening Visit, and this male is the sole partner for that subject (the information on the male partner's sterility can come from the site personnel's review of the subject's medical records or interview with the subject on her medical history); male condom combined with a vaginal spermicide (foam, gel, film, cream, or suppository); male condom combined with a female diaphragm, either with or without a vaginal spermicide (foam, gel, film, cream, or suppository).

4. Have immunosuppression as a result of an underlying illness or treatment, or use of anticancer chemotherapy or radiation therapy (cytotoxic) within the preceding 36 months, or plans to receive immunosuppressive therapy/cytotoxic treatment during study participation.

5. Have an active neoplastic disease or a history of any hematologic malignancy. However, subjects with superficial skin cancer who do not require intervention other than local excision are not excluded.

6. Have long-term use (≥14 consecutive days) of glucocorticoids including oral or parenteral prednisone or prednisone equivalent (>20 mg total dose per day) or high-dose inhaled steroids (>800 µg/day of beclomethasone dipropionate or equivalent) within 1 month prior to screening in this study. However, subjects on low-dose inhaled steroids (≤800 µg/day of beclomethasone dipropionate or equivalent) or topical steroids are not excluded.

7. History of schizophrenia, bipolar disease, psychosis, or severe personality disorder.

8. History of hospitalization for psychiatric illness, attempted suicide, or having been deemed a danger to self or others within the past 10 years.

9. Have received immunoglobulin or other blood product (with the exception of Rho[D] immune globulin) within the 3 months prior to screening in this study.

10. Have received any live vaccines within 4 weeks or inactivated or recombinant protein vaccines within 2 weeks prior to screening in this study or plan to receive such vaccines (including seasonal influenza vaccines) from screening through 21 days following the second dose of the study vaccine (Screening Visit through Day 50).

11. Have an acute or chronic medical condition that, in the opinion of the site investigator, would render vaccination unsafe or would interfere with the evaluation of responses. This includes all PIMMCs such as Guillain-Barré syndrome, narcolepsy, and current or history of autoimmune or chronic inflammatory disease (as listed in Appendix 3).

12. Have an acute illness, including body temperature greater than 100.4°F, at screening, immediately prior to each vaccination or, per subject report, within 3 days prior to each vaccination in this study.
13. Received an experimental agent (vaccine, drug, biologic, device, blood product, or medication) within 1 month prior to screening in this study or expect to receive an experimental agent during the study period.
14. Are participating or plan to participate in another interventional clinical trial (either active or follow-up phase) during the study period.
15. Participated in an A(H7) influenza vaccine study in the past or have a history of A(H7) influenza infection prior to vaccination in this study.
16. Have known human immunodeficiency virus, hepatitis B, or hepatitis C infection (based on medical history).
17. Have a history of alcohol or drug abuse in the last 5 years.
18. Have a body mass index >35 kg/m^2.
19. Have a first degree relative with narcolepsy.
20. Have any laboratory test result or clinical findings (including vital signs) that singly or in combination are likely to unfavorably alter the risk-benefit of subject participation or to confound study safety or immunogenicity results. Subjects cannot be rescreened based on abnormal laboratory test results.
21. Alanine aminotransferase or AST >2 times the upper limit of normal (ULN), or bilirubin >1.5 times the ULN unless isolated Gilbert's syndrome. Subjects cannot be rescreened based on abnormal laboratory test results.

7.2. Subject Withdrawal Criteria

Every subject has the right to refuse participation in the study (i.e. withdraw consent) at any time without providing any reason for withdrawal. A subject's participation must be terminated immediately upon his/her request, and the reason(s) for discontinuation documented accordingly in the corresponding electronic case report form (eCRF).

Any subject who meets one or more of the following criteria will be removed from the study without prejudice:

- Subject request.
- Subject noncompliance, defined as refusal or inability to adhere to the study protocol or any other instances determined by the investigator or BARDA.
- Subject lost to follow-up.
- Investigator no longer believes participation is in the best interest of the subject.
- At request of BARDA, regulatory agencies, or the institutional review board (IRB).

For any subject who meets the above criteria, the early termination (ET) visit assessments should be performed as outlined in Appendix 1, when possible. If not possible to conduct a visit, site

staff should make every effort to obtain updated safety information (AE assessment including pregnancy information) by phone.

7.2.1. Replacements

If a subject is withdrawn from the clinical study after receipt of the first dose of vaccine, the subject will not be replaced.

If a subject is randomized but does not receive the first dose of vaccine, the subject will be withdrawn and will not be counted toward the total enrollment goal. Additional subjects will be randomized to achieve enrollment goals.

8. TREATMENT OF SUBJECTS

8.1. Description of Study Vaccines

A summary of the study vaccines to be evaluated in this study is presented in Table 3. The study vaccines will be prepared by mixing recombinant Panblok H7 influenza vaccine antigen 1:1 with either MF59 or AS03 adjuvant prior to administration. Three concentrations of antigen will be provided to prepare the 3 required dosages of the study vaccines being examined.

Table 3: Summary of Study Vaccines

Product	Manufacturer	Concentration	Fill volume (mL)	Vial Size (mL)
Influenza Vaccine Antigen Panblok H7				
A/Guangdong/ 17SF003/2016	PSC	60 µg/mL	0.65	2.0
A/Guangdong/ 17SF003/2016	PSC	30 µg/mL	0.65	2.0
A/Guangdong/ 17SF003/2016	PSC	15 µg/mL	0.65	2.0
Adjuvant				
MF59	Seqirus (formerly Novartis)	39 mg squalene/mL	3.2	5.0
AS03	GSK	42.4 mg squalene/mL	3.0	3.0

GSK = GlaxoSmithKline; PSC = Protein Sciences Corporation

8.1.1. Description of Influenza Vaccine Antigen

Panblok H7 consists of full length rHA derived from the A/Guangdong/17SF003/2016 (H7N9) influenza strain. This protein is produced in Protein Sciences Corporation's patented *expres*SF+® (Lepidopteran) insect cells, [redacted] and the rHA is formulated in phosphate-buffered saline without preservatives, antibiotics, or adjuvants. The vaccine is a sterile liquid with no added preservatives, for IM injection, supplied in single dose vials containing 0.65 mL. The vaccine antigen is clear and slightly opalescent in appearance.

[redacted]

[redacted]

[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]

8.1.2. Description of Adjuvants

8.1.2.1. MF59

The MF59 adjuvant is an oil-in-water emulsion composed of squalene, stabilized by the addition of two emulsifiers, polysorbate 80 and sorbitan trioleate (Span 51), and a low ionic strength buffer. The MF59 adjuvant is milky white in appearance. [redacted]

[redacted]

[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]

8.1.2.2. AS03

The AS03 adjuvant is an oil-in-water emulsion containing DL-α-tocopherol, squalene, polysorbate 80, and a buffer. The AS03 adjuvant is milky white in appearance. [redacted]

[redacted]

[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]
[redacted]	[redacted]

8.2. Concomitant Medications

Any treatment including all prescription drugs, herbal products, vitamins, minerals, and over-the-counter (OTC) medications or therapies administered from the time of consent through the end of study participation is considered a concomitant medication. Concomitant medication use will be recorded in the eCRF and will include the medication name, dose, frequency, route of administration, and the dates of administration. Any changes, additions, and/or deletions in concomitant medications will be recorded in the subject's eCRF throughout the course of the subject's participation in the study.

If it is discovered that a subject is using a prohibited concomitant medication after he or she is enrolled in the study, the investigator, in consultation with the medical monitor in cases of uncertainty, should determine the impact on the subject's participation. All instances of use of prohibited concomitant medications must be documented on the appropriate eCRFs.

In addition, any vaccines received 3 months prior to the Screening Visit will be recorded in the subject's eCRF.

8.2.1. Prohibited Concomitant Medications and Therapies

The following concomitant medications and therapies are prohibited for the duration of the study unless otherwise specified.

- Anticancer chemotherapy or radiation therapy (cytotoxic).
- Long-term use (≥14 consecutive days) of glucocorticoids including oral or parenteral prednisone or prednisone equivalent (>20 mg total dose per day) or high-dose inhaled steroids (>800 μg/day of beclomethasone dipropionate or equivalent).
- Treatment with immunoglobulin or other blood products (with the exception of Rho[D] immune globulin).
- Concomitant vaccines (including seasonal influenza vaccines) from screening through 21 days following the second dose of the study vaccine (Screening Visit through Day 50).
- Experimental agents (vaccine, drug, biologic, device, blood product, or medication).

8.2.2. Permitted Concomitant Medications

Other than the prohibited medications and therapies listed in Section 8.2.1, treatment with concomitant medications and therapies is permitted during the study. Concomitant medication deemed necessary for the welfare of the subject during the study may be given at the discretion of the investigator. However, it is the responsibility of the investigator to ensure that details regarding the medication will be recorded in full in the eCRF.

8.3. Treatment Compliance

The study vaccines will be administered by an unblinded study staff member and thus is an observed compliance. Subject compliance will be determined by the number and percentage of subjects who receive study vaccine on Day 1 and Day 29 by treatment group. Any deviations from the dosing schedule outside the defined visit windows (Appendix 1) will be recorded on the appropriate eCRF.

9. STUDY VACCINE MATERIALS AND MANAGEMENT

9.1. Study Vaccines

Panblok H7 vaccine, adjuvanted, will be supplied as 2 components that must be mixed prior to administration: rHA H7 antigen and adjuvant (AS03 or MF59). Refer to Section 8.1 for detailed information regarding the vaccine components.

9.2. Study Vaccine Packaging and Labeling

Study vaccines will be packaged and labeled according to applicable local and regulatory requirements. Study vaccine will be provided in single-dose kits containing 1 vial of antigen and 1 vial of adjuvant. Vials of antigen and adjuvant will be labeled with a single unblinded panel label. Each kit will be labeled with a three panel blinded label: one part that stays on the kit, one tear-off part that can be affixed to the site's study vaccine accountability documentation, and one part to be affixed to the syringe used for vaccine administration.

The vials and kits will be labeled in English. The vial and kit labels will include the Sponsor Name (BARDA), protocol number, and a note that specifies the following "Caution: New Drug - Limited by Federal Law to Investigational Use Only", as well as storage specifications. In addition, the vial labels will contain the antigen or adjuvant lot number. Each kit label will include a unique and blinded kit number (kit ID).

9.3. Study Vaccine Storage

Study vaccine kits containing antigen and adjuvant must be stored in a secure area (e.g., a locked room or locked refrigerator), protected from light and moisture, and kept at a controlled standard refrigeration temperature between 2°C to 8°C (36° to 46°F), inclusive. The temperature of the storage unit must be monitored and documentation of proper storage must be maintained. Vaccine components should be removed from the refrigerated storage unit and allowed to warm to room temperature for approximately 15 minutes before mixing. Vaccine must be administered within 30 minutes of mixing.

9.4. Study Vaccine Preparation and Administration

Antigen (0.65 mL) and adjuvant (0.65 mL) will be mixed prior to administration according to the details outlined in the manuals of procedures (MOP). The vaccine once mixed should have a milky white appearance. The vaccine should be visually inspected for particulate matter and discoloration prior to administration; it should not be administered if either condition exists.

The vaccine (0.5 mL) should be administered by IM injection in the deltoid muscle of the arm within 30 minutes of mixing. Each vaccination will be given in a different arm. The vaccine administrator will record what arm received the deltoid injection.

9.5. Study Vaccine Accountability

The investigator is required to maintain adequate records of the disposition of the study vaccines, including the date and quantity of vaccines received, to whom the vaccine was dispensed

(subject-by-subject accounting), and a detailed accounting of any vaccine accidentally or deliberately destroyed. Records for receipt, storage, use, and disposition will be maintained by the study site. A vaccine-dispensing log will be kept current and will include kit ID, identification of each subject, and the date and quantity of vaccine dispensed. All records regarding the disposition of the study product will be available for inspection by the unblinded study monitor.

9.6. Study Vaccine Handling and Disposal

At the completion of the study, to satisfy regulatory requirements regarding accountability, all study vaccines will be reconciled and returned to the study vaccine distributor according to applicable regulations. No study vaccines are to be destroyed until authorized in writing by BARDA.

9.7. Randomization and Blinding

9.7.1. Randomization

Subjects will be randomly assigned to 1 of the 6 treatment groups in a 1:1:1:1:1:1 ratio. An interactive web response system (IWRS) will be used to centrally administer the randomization schedule. The randomization schedule will be generated using SAS software Version 9.3 or later (SAS Institute Inc., Cary, North Carolina). Randomization will take place according to a fixed schedule using a permuted block design stratified by clinical site. The IWRS will assign subjects to treatment groups based on the predefined randomization list. A kit ID corresponding to the assigned treatment will be assigned by the IWRS from the inventory available at the site. The randomization schedule will be generated by the unblinded randomization team and will be kept strictly confidential, accessible only to authorized unblinded persons (Section 9.7.2).

9.7.2. Blinding

This is a double-blinded study. All study vaccines will be prepared and administered by an unblinded study staff member. Study vaccine accountability will be monitored by a separate unblinded study monitor. The subject and all other study staff involved in observing the subject after vaccination will be blinded to group assignment. Laboratory staff performing the safety laboratory assessments and immunogenicity assays will be blinded to treatment group.

The unblinded Rho study team will include a statistician, statistical programmer, project manager, study monitors, and the Client Support Services and IWRS teams. A BARDA representative may also be unblinded to assist only with oversight of study vaccine management.

If an SMC meeting is needed (Section 6.4.2), the members of the SMC will review safety data fully unblinded at the treatment group and subject level. BARDA will review unblinded data by treatment group for the interim analysis (Section 12.1.4.4).

9.7.3. Breaking the Blind

Unblinding of individual subjects should occur only when knowledge of the treatment assignment will have a direct bearing on the medical treatment or evaluation of a subject. Whenever possible, the need to unblind should be discussed with BARDA and the medical monitor prior to unblinding. In the event of an emergency, the investigator or designated

qualified individual may obtain the subject's blinded treatment via IWRS. If an individual designated to perform emergency unblinding does not have access to the IWRS, Rho Client Support Services may be contacted and can perform the unblinding for that individual. Upon performance of a blind break, the IWRS system will send out a blinded notification to alert Rho and BARDA staff.

A full account of the unblinding event will be recorded in the subject's source document and eCRF, including the date and time of the unblinding, the reason for the decision to unblind, the extent of unblinding, and the name and signature of the individual who made the decision to unblind. The treatment assignment should not be included in either the source document or the eCRF.

The need for emergency unblinding is anticipated to be low since AS03 and MF59 are both squalene-based adjuvants with similar safety profiles.

In addition, as needed to meet regulatory reporting requirements, designated Rho pharmacovigilance personnel may be unblinded to the treatment status of individual subjects. In this circumstance, and if there are no other concerns, neither BARDA nor the study staff will be further unblinded to treatment status.

The study data through Day 212 will be unblinded to prepare the clinical study report. The blinded site and laboratory staff will be kept blinded at the subject level through the end of study.

10. ASSESSMENT OF IMMUNOGENICITY

Venous blood samples will be collected for immunogenicity assays at the Screening Visit, Visit 4 (Day 29) prior to vaccination, Visit 7 (Day 50), Visit 8 (Day 121), and Visit 9 (Day 212). Serum will be shipped to the central laboratory for storage and subsequently shipped to the immunogenicity laboratory for analysis. Good Laboratory Practice-compliant HAI and MN assays will be conducted to determine titers of HAI and MN antibodies, respectively.

The details for sample handling, processing, and shipping will be provided in the MOP.

11. ASSESSMENT OF SAFETY

11.1. Safety Parameters

Safety will be evaluated utilizing the assessments defined in this section. If deemed clinically necessary, additional safety assessments not currently specified in the protocol may be performed at the discretion of the investigator in consultation with the medical monitor and BARDA.

11.1.1. Demographic/Medical History

In order to define a baseline for potential AEs, the demographics and medical history of each subject will be collected at the Screening Visit and recorded on the appropriate eCRFs.

11.1.2. Vital Signs

Vital sign measurements, including oral temperature, pulse rate, respiratory rate, and diastolic and systolic blood pressure (after the subject is seated for at least 5 minutes), will be collected at the Screening Visit, Visit 1 (Day 1), Visit 3 (Day 8), Visit 4 (Day 29), Visit 6 (Day 36), Visit 7 (Day 50), ET Visit, and Unscheduled Visit (if clinically indicated) and will be recorded on the appropriate eCRF. Respiratory rate, if regular, may be assessed over 30 seconds and doubled, but in no case should it be assessed over a period of less than 30 seconds. Pulse rate, if performed manually, must be assessed over at least 15 seconds.

As the subject will self-assess oral temperature between clinic visits when solicited AEs are being collected (for 8 days following each vaccination, inclusive of the vaccination day) (see Section 11.2.1.2), sites will train each subject at Visit 1 (subjects will take their own temperature in front of site staff to confirm correct measurement before leaving the clinic). Diary cards (Section 11.1.7) will include instructive reminders, and sites will reinforce proper assessment at each applicable visit to continually assess subject competency.

11.1.3. Height and Weight

Subject's height and weight will be taken and BMI calculated at the Screening Visit and recorded on the appropriate eCRF.

11.1.4. Physical Examination

A physical examination will be performed at the Screening Visit and during Visit 4 (Day 29) prior to vaccination to assess and confirm eligibility. A physical examination will also be conducted at Visit 7 (Day 50). The examination will include a general assessment of the skin, head, ears, eyes, nose, throat, neck, thyroid, lungs, heart, abdomen, lymph nodes, and musculoskeletal system/extremities, and the results will be recorded on the appropriate eCRF.

A targeted physical examination will be performed at Visit 1 (Day 1), Visit 3 (Day 8), Visit 6 (Day 36), and at ET and Unscheduled Visits, at the discretion of the investigator to evaluate AEs or clinical laboratory abnormalities. Results will be recorded on the appropriate eCRF.

11.1.5. Clinical Laboratory Assessments

Venous blood samples will be collected for routine clinical laboratory safety evaluations at the Screening Visit, Visit 3 (Day 8), Visit 4 (Day 29) prior to vaccination, Visit 6 (Day 36), and at ET and Unscheduled Visits (if deemed clinically indicated by the investigator, in consultation with the medical monitor), and will be shipped to the central laboratory for analysis. The details for sample handling, processing, and shipping will be provided in the MOP.

Individual results will be sent to the applicable site, and the investigator will perform a clinical assessment of all laboratory safety data to assess eligibility and identify and document AEs as applicable. All results will be transferred electronically directly from the central laboratory to Rho using standard secure data transfer procedures.

The clinical laboratory assessments planned for this study include chemistry and hematology assessments as follows.

11.1.5.1. Chemistry

- Alanine aminotransferase
- Albumin
- Alkaline phosphatase
- Aspartate aminotransferase
- Bicarbonate
- Blood urea nitrogen
- Calcium
- Chloride
- Creatine kinase
- Creatinine
- Glucose, random, serum
- Potassium
- Sodium
- Total bilirubin (fractionated for values > ULN)
- Total protein

11.1.5.2. Hematology

- Coagulation: partial thromboplastin time and prothrombin time (international normalized ratio)
- Complete blood count (CBC) with differential; Table 7 lists the parameters of the CBC with differential.

Table 7: CBC with Differential

Red blood cells	**White blood cells**
Hematocrit	Basophils (% and absolute count)
Hemoglobin	Eosinophils (% and absolute count)
Mean corpuscular hemoglobin	Lymphocytes (% and absolute count)
Mean corpuscular hemoglobin concentration	Monocytes (% and absolute count)
Mean corpuscular volume	Neutrophils (% and absolute count)
Red blood cell count	White blood cell count
Platelets	
Platelet count	

11.1.6. Pregnancy Screen

At the Screening Visit, Visit 1 (Day 1), and Visit 4 (Day 29), a urine dipstick pregnancy test will be performed onsite for female subjects of childbearing potential. For Visits 1 and 4, the test must be performed within 24 hours prior to vaccination, and results must be final and negative prior to vaccination.

Additionally, a urine dipstick pregnancy test should be performed at any time during study participation if pregnancy is suspected.

11.1.7. Diary Cards

All subjects will be provided with diary cards (see Table 8) to aid in their recording of solicited local and systemic reactions to the study vaccine and other unsolicited symptoms/complaints, including start and stop dates (see Section 11.2.1.2). In addition, the diary cards will include training information on temperature measurements (Section 11.1.2) and injection site reaction assessment.

The investigator or his/her designee will review the information from the diary card or from memory with the subject at each applicable visit and assess whether any AE criteria are met based on standard toxicity grading scales for clinical and laboratory abnormalities (Section 11.3.3 and Appendix 2). Once confirmed to be an AE per the toxicity grading scales (Section 11.3.3 and Appendix 2), the investigator or designee will enter the information into the appropriate eCRF. In addition, any changes in concomitant medications will be documented on the appropriate eCRF.

The diary cards will be collected from the subjects and stored as source documents.

Table 8: Schedule of Diary Cards

Diary Card	Type of Event	Vaccination	Collection Period: Days After Dosing	Collection Period: Study
Diary Card I	Solicited, Unsolicited	Dose 1	Days 1-8 Postvaccination, inclusive of vaccination day	Days 1-8
Diary Card II	Unsolicited	Dose 1	Days 8-29 Postvaccination, inclusive of vaccination day	Days 8-29
Diary Card III	Solicited, Unsolicited	Dose 2	Days 1-8 Postvaccination, inclusive of vaccination day	Days 29-36
Diary Card IV	Unsolicited	Dose 2	Days 8-21 Postvaccination	Days 36-50

11.1.8. Postvaccination Evaluation

At Visit 1 (Day 1) and Visit 4 (Day 29), subjects will be monitored for at least 30 minutes after vaccination; any safety events and concomitant medications will be recorded on the appropriate eCRFs. Details regarding collection and reporting of solicited local and systemic reactogenicity symptoms are presented in Section 11.1.7 and Section 11.2.1.2.

After the subject has been monitored for 30 minutes (± 5 minutes) following vaccination, the following evaluations will be performed:

1. Obtain vital sign measurements, as described in Section 11.1.2,
2. Complete an injection site examination for erythema/redness and induration/swelling and monitor for pain. If a visible injection site abnormality is observed,
    a. document with a photograph if the subject consented.
    b. measure and document the area (largest diameter) of erythema/redness and induration/swelling.

Photographs will be taken by study staff only in the event of abnormal findings (not expected with usual vaccine administration) for accurate description and evaluation of the findings and safety reviews and will be retained with subject source documents. There are no digital format requirements; the photographs will not be stored in the study database and will not be used as part of the safety analysis. They may, however, be shared with the medical monitor, BARDA, and the SMC in a secure and blinded manner without exposing personal identification information.

11.1.9. AE Assessment

Every time the subject is in contact with site personnel after the first vaccination, site staff will ask nonleading questions regarding the subject's health status (Section 11.3.1) and document any new or changed AEs on the appropriate eCRF.

11.1.10. Concomitant Medication Assessment

Every time the subject is in contact with site personnel, site staff will review concomitant medications (Section 8.2) and document any new or changed medications on the appropriate eCRF.

11.2. Adverse and Serious Adverse Events

11.2.1. Definition of Adverse Events

11.2.1.1. Adverse Events

An AE is any untoward medical occurrence associated with the use of a drug in humans, whether or not it is considered drug-related [21 Code of Federal Regulations [CFR] 312.32(a)]. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product (International Conference on Harmonisation [ICH] E6 (R1) Guideline for Good Clinical Practice, June 1996).

Laboratory results and vital sign excursions of any magnitude will be defined as AEs if they are considered clinically significant by the investigator. Additionally, any laboratory or vital sign excursion to a clinical Grade 3 or higher severity will be considered an AE.

Suspected adverse reaction (SAR) means any adverse event for which there is a reasonable possibility that the drug caused the adverse event. For the purposes of safety reporting, 'reasonable possibility' means there is evidence to suggest a causal relationship between the drug and the adverse event. A SAR implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug [21 CFR 312.32(a)].

An AE or SAR is considered "unexpected" if it is not listed in the investigator brochures or is not listed at the specificity or severity that has been observed.

11.2.1.2. Solicited & Unsolicited Adverse Events

For the purposes of this study, the following specific local and systemic AEs will be solicited from the subject for 8 days postvaccination, inclusive of the vaccination day (Section 11.1.7).

- Solicited local reactions at the injection site will include erythema/redness, induration/swelling, and pain.
- Solicited systemic reactions will include fever, myalgia, arthralgia, fatigue, headache, nausea, vomiting, diarrhea, and chills.

All other AEs reported by the subject during the early part of the study (through Day 50, Section 11.1.7) will be defined as unsolicited AEs.

11.2.1.3. Serious Adverse Events

An adverse event or suspected adverse reaction is considered "serious" if, in the view of either the investigator or BARDA, it results in any of the following outcomes [21 CFR 312.32(a)]:

- Death.
- Life-threatening adverse event that in the view of the investigator or BARDA, places the subject at immediate risk of death. This does not, however, include an event that, had it occurred in a more severe form, might have caused death.
- Requires inpatient hospitalization or prolongs existing hospitalization. Planned hospitalizations will **not** be reported as SAEs unless categorized as medically

important. Emergency room visits and observational admissions of under 24 hours, in themselves, do not qualify as SAEs.

- Results in persistent or significant disability/incapacity or substantial disruption of the ability to conduct normal life functions.
- Congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered a serious adverse drug experience, when based on appropriate medical judgment, they may jeopardize the subject or the subject may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

11.2.1.4. Medically Attended Adverse Events (MAAEs)

Clinical trials of preventive vaccines with AS03 and MF59 adjuvants implement collection and analysis of data relating to MAAEs among subjects in all treatment groups through 13 months following the first study vaccination, due to the theoretical potential for induction of autoimmune or autoinflammatory diseases.

MAAEs are defined as AEs with medically attended visits including hospital, emergency room, urgent care clinic, or other visits to or from medical personnel (medical doctor) for any reason.

11.2.1.5. Potentially Immune-Mediated Medical Conditions (PIMMCs)

Clinical trials of preventive vaccines with AS03 and MF59 adjuvants implement collection and analysis of data relating to PIMMCs among subjects in all treatment groups through 13 months following the first study vaccination, due to the theoretical potential for induction of autoimmune or autoinflammatory diseases.

For this study, the occurrence of a PIMMC is considered to be unexpected and will be reported as a serious and unexpected suspected adverse reaction (SUSAR) per 21 CFR 312.32.

A list of PIMMCs defined for this study is presented in Appendix 3.

11.3. Collection, Recording, and Grading Severity of Adverse Events

11.3.1. Collection of Adverse Events

AEs of any type described above may be discovered through a variety of methods:

- Observing the subject
- Reviewing and discussing the diary cards with the subject (Section 11.1.7)
- Questioning the subject with standard nonleading questions to elicit any medically related changes in their well-being (Have you been hospitalized, had any accidents, used any new medications, or changed any medications [both prescription and OTC]?)
- Receiving an unsolicited complaint from the subject
- An abnormal value or result from a clinical (e.g. vital signs) or laboratory evaluation

11.3.2. Recording of Adverse Events

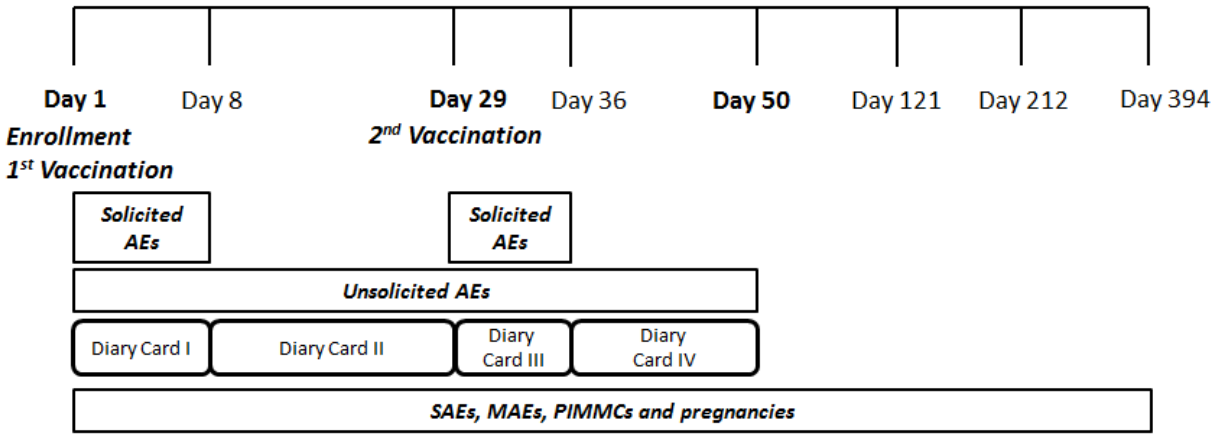
Throughout the study, the investigator will record AEs on the appropriate eCRF. Any clinically significant safety assessments that are associated with an underlying disease present at screening, unless judged by the investigator to be more severe than expected for the subject's condition, are **not** to be reported as AEs or SAEs.

AEs will be recorded during the periods defined below.

- Solicited AEs:
    - Those occurring within 8 days of each vaccination, inclusive of the vaccination day (Day 1 through Day 8 and Day 29 through Day 36).
    - Those judged related to study vaccine will be followed to resolution (with or without sequalae) or until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty).
    - Those judged unrelated to study vaccine will be followed to resolution (with or without sequalae) or, if not resolving, until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty), or until the end of the subject's study participation, whichever comes first.
- Unsolicited AEs:
    - Those occurring from the time the subject receives the first vaccination until Visit 7 (Day 50).
    - Those judged related to study vaccine will be followed to resolution (with or without sequalae) or until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty).
    - Those judged unrelated to study vaccine will be followed to resolution (with or without sequalae) or, if not resolving, until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty), or until the end of the subject's study participation, whichever comes first.
- SAEs, MAAEs, and PIMMCs:
    - Those occurring from the time the subject receives the first vaccination until exit from the study will be reported to Rho pharmacovigilance personnel within 1 business day of the site's awareness of the event.
    - Those judged related to study vaccine will be followed to resolution (with or without sequalae) or until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty).
    - Those judged unrelated to study vaccine will be followed to resolution (with or without sequalae) or, if not resolving, until considered stable by the investigator (in consultation with the medical monitor in situations of uncertainty), or until the end of the subject's study participation, whichever comes first.

Figure 2 provides the safety data reporting periods used in this study.

Figure 2: Safety Data Reporting Periods



AE= adverse event; MAAE = medically attended adverse event; PIMMC = potentially immune-mediated medical condition; SAE = serious adverse event

Note: Unsolicited AEs will be collected through the end of the study visit window for each AE collection period (i.e., 21 days after the second vaccination ± 3-day window means Day 50 + 3 days).

Note: The entire study period is from Day 1 from the time a subject receives the first study vaccination through Day 394.

Note: Pregnancies that occur during a subject's participation in the study will be followed until conclusion even if the pregnancy ends after Day 394.

11.3.3. Grading Severity of Adverse Events

The severity of AEs and SAEs will be graded as defined by the Food and Drug Administration's (FDA) Guidance for Industry *"Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials" (September 2007)* (Appendix 2).

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity, whereas seriousness is defined by the criteria in Section 11.2.1.3.

11.4. Relationship and Attribution to Study Vaccine

An investigator's causality assessment is the determination of whether there exists a reasonable possibility that the study vaccine caused or contributed to an AE and must be provided for all AEs (serious and non-serious). The investigator will assess the causality/relationship between the study vaccine and the AE and record that assessment in the appropriate eCRF.

BARDA's determination of attribution will be used for reporting to the appropriate health and regulatory authorities.

The relation and attribution of an AE to study treatment will be determined using the descriptors and definitions provided in Table 9.

Table 9: Attribution of Adverse Events

Unrelated Categories	
Not Related	The AE is clearly not related to vaccination.
Unlikely Related	The AE is unlikely related to vaccination.
Related categories	
Possibly Related	The AE has a reasonable possibility to be related to vaccination; there is evidence to suggest a causal relationship.
Probably Related	The AE is likely related to vaccination.
Related	The AE is clearly related to vaccination.

AE = adverse event

11.5. Reporting Safety Events to Regulatory Authorities

Once AEs, SAEs, MAAEs, and PIMMCs are recorded into the appropriate eCRF, Rho pharmacovigilance personnel will collaborate with BARDA and appropriate personnel to process and report events to the appropriate regulatory authorities within applicable regulatory timeframes. Procedures for AE processing and reporting are detailed in the Data Safety Monitoring Plan (DSMP).

The sponsor will report to the FDA any suspected adverse reaction that is both serious and unexpected (SUSAR). The sponsor will report an AE as a SAR only if there is evidence to suggest a causal relationship between the study vaccine and the adverse event, such as:

a) A single occurrence of an event that is uncommon and known to be strongly associated with vaccine exposure (e.g., angioedema, Stevens-Johnson Syndrome, Guillain-Barré syndrome);

b) One or more occurrences of an event that is not commonly associated with vaccine exposure, but is otherwise uncommon in the population exposed to the vaccine (e.g., narcolepsy);

c) An aggregate analysis of specific events observed in a clinical trial (such as known consequences of the underlying disease or condition under investigation or other events that commonly occur in the study population independent of treatment) that indicates those events occur more frequently in the treatment group than in a concurrent or historical control group (21CFR 312.32(a)).

d) PIMMCs (Section 11.2.1.5) will be submitted as SUSARs.

11.6. Pregnancy Reporting

This study includes pregnancy as safety data. Although pregnancy is not an SAE, information about any pregnancy in a female study subject should be reported promptly to Rho on the same timeline as an SAE for tracking purposes (Section 11.3.2).

Study vaccination will be discontinued for the pregnant subject. The investigator shall counsel the pregnant subject and discuss the risks of continuing with the pregnancy and the possible effects on the fetus. Monitoring of the pregnant subject shall continue until the conclusion of the pregnancy.

The pregnancy will be documented on the appropriate eCRF when identified and at its conclusion.

Should pregnancy result in a congenital abnormality, birth defect, miscarriage, or medically indicated abortion, an SAE must be submitted to Rho using the SAE reporting procedures described in Section 11.3.2.

11.7. Reporting Other Safety Information

Investigators should promptly notify Rho, BARDA, and the IRB when an unanticipated problem involving risks to subjects or others is identified, which is not otherwise reportable as an AE.

12. STATISTICS

12.1. Endpoints

12.1.1. Primary Endpoints

There are two primary endpoints for this study.

- All solicited local and systemic reactogenicity symptoms occurring within 8 days of each vaccination, inclusive of the vaccination day.
- Seroprotection at Day 50 based on serum HAI antibody titers, defined as an HAI antibody titer ≥1:40.

12.1.2. Secondary Endpoints

12.1.2.1. Secondary Safety Endpoints

- All treatment-emergent SAEs occurring during study participation.
- All treatment-emergent MAAEs occurring during study participation.
- All treatment-emergent PIMMCs occurring during study participation.
- All treatment-emergent unsolicited AEs occurring during study participation.

12.1.2.2. Secondary Immunogenicity Endpoints

- Serum HAI antibody titers at Screening and Days 29, 50, 121, and 212.
- Serum MN antibody titers at Screening and Days 29, 50, 121, and 212.
- Seroprotection at Screening and Days 29, 121, and 212 based on serum HAI antibody titers, defined as an HAI antibody titer ≥1:40.
- Seroconversion at Days 29, 50, 121, and 212 based on serum HAI antibody titers, defined as either a prevaccination HAI titer <1:10 and a post-vaccination HAI titer ≥1:40, or a prevaccination HAI titer ≥1:10 and a minimum 4-fold rise in postvaccination HAI titer.
- Seroconversion at Days 29, 50, 121, and 212 based on serum MN antibody titers, defined as either a prevaccination MN titer <1:10 and a postvaccination MN titer ≥1:40, or a prevaccination MN titer ≥1:10 and a minimum 4-fold rise in postvaccination MN titer.

12.1.3. Measures to Minimize Bias

Stratified block randomization will be performed centrally and will balance enrollment between treatment groups at each clinical site. Clinical staff observing the subject postvaccination, laboratory personnel analyzing samples, and study subjects will be blinded to treatment group. Site staff will be trained in procedures to ensure blinding is maintained for all blinded individuals.

Analyses will be prepared by a blinded team using dummy treatment groups. An unblinded team (Section 9.7.2) will generate, interpret, and report each analysis using appropriate treatment groups and will not modify any programs used to perform analyses.

12.1.4. Analysis Plan

Statistical analyses will be performed using SAS® software Version 9.3 or later.

Due to the exploratory nature of this study, no inferential analyses are planned.

Descriptive statistics (such as medians, quartiles, and ranges for continuous data and percentages for categorical data) will be used to summarize subject characteristics, safety, and immunogenicity parameters. These summaries will be presented overall and separately for the subjects in the different treatment groups, as well as pooled groups by antigen dose level and by adjuvant. Details of the statistical analyses, methods, and data conventions will be described in the Statistical Analysis Plan (SAP).

12.1.4.1. Analysis Populations

12.1.4.1.1. Safety Population

The safety population will include all subjects who are randomized and receive at least one vaccination. Each subject will be analyzed as part of the treatment group corresponding to the actual treatment received. The safety population will be used for all safety analyses.

12.1.4.1.2. Immunogenicity Full Analysis Population

The immunogenicity full analysis population will include all subjects who are randomized, receive at least one vaccination, and have determinate assay results at any postvaccination visit. Each subject will be analyzed as part of the treatment group assigned by randomization, regardless of the treatment actually received. All immunogenicity analyses will be performed on the immunogenicity full analysis population.

12.1.4.1.3. Immunogenicity Per Protocol Population

The immunogenicity per protocol population will include all subjects who meet the following criteria:

- Are in the immunogenicity full analysis population.
- Received a full dose of vaccine at Day 1 and a full dose of vaccine at Day 29.
- Received the correct treatment as assigned by randomization.
- Have no other major protocol deviations that may have an impact on immunogenicity assessments.
- Have determinate assay results at the Day 50 visit.

All immunogenicity analyses will also be performed on the immunogenicity per protocol population by the treatment group actually received.

12.1.4.2. Primary Analyses

12.1.4.2.1. Primary Safety Analyses

The frequency of solicited local and systemic reactogenicity symptoms, as defined in Section 11.2.1.2, will be summarized by system organ class and preferred term. The risk of the event in each treatment group will be described using event rates and corresponding 95% CIs. Events will only be included in the primary endpoint if they occur within 8 days of vaccination, inclusive of the vaccination day (i.e., Day 1 through Day 8 and Day 29 through Day 36).

12.1.4.2.2. Primary Immunogenicity Analyses

Proportions of subjects who achieve seroprotection based on serum HAI antibody titers at Day 50 will be reported for each treatment group with the corresponding 95% CI for that group.

12.1.4.3. Secondary Analyses

The four secondary safety endpoints (Section 12.1.2.1) will be summarized in the same manner as the primary safety endpoint. Additionally, both primary and secondary safety endpoints will be summarized by vaccination day, onset days post each vaccination, grade, and relationship to study vaccine.

Continuous HAI and MN antibody titers will be summarized as GMT by treatment group and visit with the corresponding 95% CIs about the GMT for each group.

Secondary immunogenicity endpoints related to seroprotection and seroconversion will be summarized in the same manner as the primary immunogenicity endpoint. Definitions of seroprotection and seroconversion can be found in Section 12.1.1 and Section 12.1.2.

Additional information regarding exploratory analyses, listings and tabular summaries of safety assessments and immunogenicity measures will be specified in the SAP.

12.1.4.4. Interim Analysis

An interim analysis will be performed based on cumulative immunogenicity and safety data through Day 50 for all subjects. At the interim analysis, the study database (all data through Day 50) will be monitored and cleaned per the Data Management Plan. Data for the interim analysis will be unblinded solely at the group level, thus, the blinding at the subject level will be maintained until the study unblinding of all data through Day 212 for the clinical study report.

All primary and secondary endpoint analyses will be performed for the interim analysis as specified in Section 12.1.4.2 and Section 12.1.4.3. Since all subjects will have completed Day 50 and no formal statistical comparisons are being made, there will be no penalty for an early look at the data.

Further details will be specified in the SAP.

12.1.4.5. Ad Hoc SMC Meeting

There are no formal or planned SMC meetings. A SMC review will only occur when a stopping rule is met and for any immediate safety concerns observed, as defined in Section 6.4.2. Data displays for any SMC review will be generated by an unblinded statistician.

Safety analyses for this study will be descriptive rather than inferential. Detailed listings and summary tabulations will be generated as specified in the SMC charter. The safety analyses will be completed using the safety population.

12.1.4.6. Final Analyses

A clinical study report will be written to include all safety and immunogenicity data through Day 212. For this Day 212 analysis, the study database (all data through Day 212) will be monitored and cleaned per the Data Management Plan. Study data will be unblinded to prepare the study report. Safety data collected after Day 212 will be provided as a supplemental report to the clinical study report. Cumulative summaries of all SAEs, MAAEs, and PIMMCs occurring from Day 1 through the end of study will be included in the supplemental report. Further details will be specified in the SAP.

12.1.4.7. Exploratory Analyses

Exploratory analyses will be defined in the SAP, if applicable.

12.2. Sample Size Considerations

No formal power analyses were conducted since the study objectives require no hypothesis testing. The sample size for this study is approximately 360 subjects, randomized 1:1:1:1:1:1 into 6 treatment groups of approximately 60 subjects each, defined in Table 2. This sample size is consistent with previous studies involving influenza vaccination[20,21].

Though no hypothesis testing will be performed as part of the primary analyses, Table 10 and Table 11 show the probability of observing a safety event and statistical power to detect seroprotection, respectively, under a hypothetical variety of scenarios. Within each table, separate calculations are performed for each pooled group of interest. Probabilities of detecting an event were calculated using the binomial distribution (Table 10) and power to detect a difference from a true seroprotection rate of 70% was calculated using an exact binomial test (Table 11), assuming a two-sided alpha level of 0.05.

Table 10: Probability of Observing at Least One Event of Interest in a Treatment Group or Pooled Grouping under Different True Event Rates

True Probability of an Event of Interest	Per Treatment Arm (N=60)	Per Antigen Dosage (N=120)	Per Adjuvant (N=180)	Overall (N=360)
0.005	0.260	0.452	0.594	0.835
0.01	0.453	0.701	0.836	0.973
0.02	0.702	0.911	0.974	0.999
0.05	0.954	0.998	>0.999	>0.999
0.1	0.998	>0.999	>0.999	>0.999

Table 11: Power for Detecting Observed Seroprotection Proportions Given a True Seroprotection Proportion of 70%

True Seroprotection Proportion (%)	Observed Seroprotection Proportion (%)	Per Treatment Arm (N=60) (%)	Per Antigen Dosage (N=120) (%)	Per Adjuvant (N=180) (%)	Overall (N=360) (%)
70	80	32.3	64.1	84.7	99.1
	85	71.6	96.8	99.8	>99.9
	90	96.6	>99.9	>99.9	>99.9

12.3. Statistical Considerations

12.3.1. Covariates

Since no statistical models will be used for the primary and secondary endpoints, adjustments for covariates will not be necessary.

12.3.2. Multi-center Studies

No by-site analyses are planned for this study.

12.3.3. Multiple Comparisons and Multiplicity

All safety and immunogenicity comparisons of treatment groups are considered exploratory in nature. As such, no adjustments will be required for multiple comparisons.

12.3.4. Subgroup Analyses

All exploratory analyses, including any subgroups that require additional consideration, will be discussed in the SAP, if necessary.

12.3.5. Missing Data

Standard procedures will be used to ensure that the data are as complete and accurate as possible. Due to the exploratory nature of this study, all descriptive summaries will be based upon all available data and no imputation will be done.

12.4. Procedure for Documenting Deviations from the Planned Analyses

The principal features of the design of this study and of the plan for statistical analysis of the data are outlined in this protocol. Additional details will be included in the SAP before initiating analyses. Any changes to that plan will be documented in the final clinical study report and will be approved by BARDA before being initiated.

13. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

13.1. Inspection of Records

BARDA, Rho, the IRB, or regulatory authorities will be allowed to conduct site visits to the investigational facilities for the purpose of monitoring, inspecting, or auditing any aspect of the study.

The investigator agrees to allow BARDA, Rho, the IRB, or regulatory authorities to inspect the clinical facilities, including the study vaccine storage area, and all documentation relating to the study, including but not limited to, all source documents, eCRFs, IRB submissions and approvals, study vaccine accountability logs, study vaccine temperature monitoring logs, regulatory documents, and correspondence.

The investigator also agrees to promptly notify BARDA and Rho of any audits scheduled by any regulatory authority and promptly forward copies of any audit reports received.

13.2. Institutional Review Board (IRB)

A copy of the protocol, informed consent forms (ICFs), subject diary, and any proposed advertising/recruitment materials will be submitted to the central IRB for written approval. Initial IRB approval and all materials approved by the IRB for this study, including the subject consent form and recruitment materials, must be maintained by the investigator and made available for inspection.

14. QUALITY CONTROL AND QUALITY ASSURANCE

Quality assurance includes all the planned and systematic actions that are established to ensure that the clinical study is performed and the data are generated, documented (recorded), and reported according to ICH GCP and local/regional regulatory standards.

A quality assurance representative from BARDA (or designee), who is independent of and separated from routine monitoring, may periodically arrange inspections/audits of the clinical study by reviewing the data obtained and procedural aspects. These inspections may include on-site inspections/audits and source data checks. Direct access to source documents is required for the purpose of these periodic inspections/audits.

14.1. Data Quality Assurance

Before enrolling any subjects in this study, BARDA personnel and the investigator will review the protocol, the investigator's brochures, the eCRFs and instructions for their completion, the procedure for obtaining informed consent, and the procedure for reporting AEs,SAEs, and PIMMCs.

As part of the responsibilities assumed by participating in the study, the investigator agrees to maintain adequate source documents for the subjects treated as part of the research under this protocol. In addition, the investigator agrees to provide access to those records to BARDA's qualified study monitor and auditor. Study monitors will verify information in the eCRFs against the source documents.

Data collected at the study site will be entered accurately and contemporaneously by study staff into Medidata RAVE, a 21 CFR 11-compliant, internet-based, remote data entry system, which is backed up nightly and those backup tapes are saved in a secure, off-site location. Data will be provided using the subject's unique identification number, not name or initials; Rho will not collect personally identifying information such as the subject's name or social security number. Subjects will provide demographic information such as race, ethnicity, and birth date. All elements of data entry (e.g., time, date, verbatim text, and the person performing the data entry) will be recorded within the RAVE system's audit trail to allow all data changes in the database to be monitored and maintained in accordance with federal regulations. Data collected by the laboratories will be transferred electronically directly from the laboratory to Rho using standard secure data transfer procedures. The analysis datasets will incorporate data from both sources. Data collected by Rho will be held in the strictest confidence, and are protected from access that could reveal personally identifying information about any subject in the study.

Clinical data management and data cleaning procedures (e.g., resolving errors and inconsistencies in the data) will be performed in accordance with applicable Rho and/or BARDA standards and validation plans to ensure the integrity of the data. Adverse events (including SAEs, MAAEs, and PIMMCs) and concomitant medication terms will be coded using the MedDRA and the WHO Drug dictionaries, respectively.

After study monitors verify the data and prior to the Day 212 analysis and the end of study, the investigator must sign each eCRF at the subject-level to confirm the integrity of the data recorded.

After the end of study database lock, each study site will receive an electronic copy of all of their site specific eCRF data as entered into Medidata RAVE for the study, including full discrepancy and audit history. Additionally, all of the study's analysis datasets will be sent to BARDA electronically for storage. Rho will maintain a duplicate copy for its records.

14.2. Study Monitoring

According to ICH GCP guidelines, the sponsor of the study is responsible for ensuring the proper conduct of the study with regard to protocol adherence and validity of data recorded on the eCRFs. This study will be monitored to ensure the maintenance of complete, accurate, legible, well-organized, and easily retrievable data. In addition, the study monitor will explain and interpret for the investigator all regulations applicable to the clinical evaluation of an investigational product as documented in ICH guidelines.

It is the study monitor's responsibility to inspect the eCRFs and source documentation directly throughout the study to protect the rights of the subjects; to verify adherence to the protocol; to verify completeness, accuracy, and consistency of the data; to perform study vaccine accountability; and to confirm adherence of study conduct to any local regulations. Details will be outlined in the Site Monitoring Plan.

14.3. Protocol Deviations

14.3.1. Protocol Deviation Definition

A protocol deviation is any noncompliance with the IRB approved study protocol, ICH GCP guidelines, or protocol specific MOPs. Any deviation that affects the rights or safety of the subject or the integrity of the data will be considered a major protocol deviation. Prospective permission to deviate from protocol requirements (i.e., "study waivers") will not be granted for this study.

14.3.2. Reporting and Managing Deviations

The investigator has the responsibility to identify, document, and report deviations. Protocol deviations may also be identified during site monitoring visits or during other forms of study conduct review. All deviations, regardless of the cause, must be documented on the appropriate eCRF, which will document at a minimum the date the deviation occurred, the date it was identified, a description of the deviation, whether the deviation resulted in an AE/SAE, and documentation of a corrective action plan. In addition, the investigator will report noncompliance to the IRB, as applicable.

Rho and/or BARDA may request discussion with the site investigator to determine the effect of any major protocol deviation on a study subject and his/her further study participation, the effect of the deviation on the overall study, and corrective actions.

15. ETHICS

15.1. Ethics Review

Before study initiation, the protocol, the informed consent documents, the subject diary, and any advertising materials will be reviewed and approved by the IRB centrally; in addition, each individual site will be reviewed and approved by the IRB prior to the shipment of study vaccine to and the activation of each individual site. Any amendments to those documents will be submitted to the IRB by Rho (following approval by BARDA) and must be approved by the IRB before they are implemented at the sites. Protocol documents must be re-approved by the IRB annually. Only institutions holding a current US Federal Wide Assurance issued by the Office for Human Research Protections at HHS may participate.

The investigator will promptly report all unanticipated problems involving risks to subjects to the IRB. The investigator will not make any changes to the research conduct without BARDA and IRB approval, except where necessary to eliminate apparent immediate hazards to the subjects.

The investigator will provide progress reports to the IRB as required by the IRB. The investigator will provide a final report to the IRB after completion of participation in the study.

15.2. Ethical Conduct of the Study

The investigator should conduct the study in accordance with this protocol, the Declaration of Helsinki, current ICH GCP guidelines, US 21 CFR Part 50 (Protection of Human Subjects) and Part 56 (Institutional Review Boards). The investigator and Rho (as BARDA's designee) will sign the protocol and study contract to confirm agreement.

15.3. Written Informed Consent

The informed consent document template will be approved by the IRB, which will create approved site-specific versions as sites are approved. The investigator is responsible for ensuring that the subject fully understands the nature and purpose of the study during the informed consent process. Information should be given in both oral and written form. No subject should be obliged to participate in the study. The information must make clear that refusal to participate in the study or withdrawal from the study at any stage is without any prejudice to the subject's subsequent care. Subjects must be allowed sufficient time to inquire about the details of the study and to decide whether they wish to participate. Written informed consent will be obtained before the subject undergoes any study procedures.

The subject must be made aware of and give consent to direct access to his/her source medical records by study monitors, auditors, the IRB, and regulatory authorities. The subject should be informed that such access will not violate subject confidentiality or any applicable regulations. The subject should also be informed that he/she is authorizing such access by signing the ICF.

The investigator will retain the original signed informed consent, and each subject will be given a signed copy to keep for his/her records.

Using the ICF, subjects who consent to participate in this study will also be asked to consent to the following:

- Permission to have a photograph of the rare and unanticipated event of a visible injection-site abnormality for the purpose of medical management and safety evaluations.
- Permission to be contacted with a request to participate in future clinical studies of subjects who have been vaccinated against influenza A/H7N9, such as longer interval prime-boost studies.
- Permission to store any leftover serum from research laboratories for future investigations, such as testing for immune response to heterologous influenza A/H7N9 or other strains or subtypes of influenza viruses.

16. DATA HANDLING AND RECORDKEEPING

16.1. Confidentiality

Each subject will be assigned a unique identification number and these numbers rather than names will be used to collect, store, and report subject information. All biological samples will be labeled with a random identification number. Site staff will not transmit documents containing personal health identifiers to BARDA or its representatives. Data reported in medical journals or scientific meetings will be presented in aggregate for subjects as a whole. No individual subject will be identified in any way.

16.2. Retention of Records

The investigator should retain all documentation relating to the study (including but not limited to ICFs, source documentation, study vaccine records, eCRFs, and essential documents) for a period of at least 2 years after the last marketing application approval or, if not approved, 2 years following the discontinuance of the test article for investigation.

If this requirement differs from any local regulations, the local regulations will take precedence unless the local retention policy is less than 2 years.

At study closure, the investigator must inform BARDA, or designee, of the long term storage location of the study's records. Following study closure, the investigator must inform BARDA if that location changes (e.g., the investigator leaves the institution where the study was conducted).

No study records will be destroyed without prior authorization from BARDA.

17. PUBLICATION POLICY

BARDA will be responsible for publication activities and will work with the investigators to define the manuscript/presentation development process, the number and order of authors, the publication/scientific meeting to which it will be submitted, and other related issues. BARDA has final approval authority over all such issues.

Data are the property of BARDA and cannot be published without prior authorization from BARDA, but data and publication thereof will not be unduly withheld.

The ASPR Public Access Plan[22] and the National Institutes of Health (NIH) Public Access Policy[23] will apply to this study. ASPR-funded investigators will be required to submit an electronic version of final, peer-reviewed manuscripts resulting from this study to the National Library of Medicine's PubMed Central upon acceptance for publication, to be made publicly available no later than 12 months after the official date of publication.

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APPENDIX 1. SCHEDULE OF ASSESSMENTS

Study Visit	Screen-ing	V1	V2[a]	V3	V4	V5[a]	V6	V7	V8	V9	V10[a] (EoS Visit)	ET	Unsch-eduled
Study Day and Visit Window	**D-14 to D-3**	**D1**	**D4 (±1 D)**	**D8 (±1 D)**	**D29 (±3 D)**	**D32 (±1 D)**	**D36 (±1 D)**	**D50 (±3 D)**	**D121 (±7 D)**	**D212 (±7 D)**	**D394 (±7 D)**	**N/A**	**N/A**
Procedure													
Obtain informed consent	X[b]												
Vital sign measurements[c]	X	X		X	X		X	X				X	X[d]
Height and weight	X												
Medical history	X												
Demographics	X												
Perform subject interview[f]			X	X	X[e]	X	X	X	X	X	X	X	X
Review diary card[g]			X	X	X[e]	X	X	X				X[h]	X[h]
Distribute diary card[g], thermometer, & measuring tool for injection site reactions and review instructions		X		X	X		X						X[h]
Collect diary card				X	X[e]		X	X				X[h]	X[h]
Record concomitant medications	X[i]	X[e]	X	X[j]	X[e,j]	X	X[j]	X[j]	X	X	X	X	X
Physical examination	X[k]				X[e]			X					
Targeted physical examination[l]		X[e]		X			X					X	X
Urine pregnancy test	X[m]	X[e,m]			X[e,m]								
Review inclusion and exclusion criteria	X	X[e]			X[e]								
Venous blood sample collection for clinical safety laboratory tests[n] (approximately 12 mL)	X			X	X[e]		X					X[d]	X[d]
Venous blood sample collection for immunogenicity assays (approximately 40 mL)	X				X[e]			X	X	X			
Randomization		X[e]											
Vaccination (per Rho-provided kit assignment)		X			X								
Monitor subject for 30 minutes following vaccination[o,p]		X			X								
Examine vaccination site		X[p]		X[q]	X[e,p,q]		X[q]					X[q r]	X[q r]
AE/SAE/MAAE/PIMMC assessment		X	X	X	X	X	X	X	X[s]	X[s]	X[s]	X[t]	X

AE = adverse event; D = day; eCRF = electronic case report form; EoS = end-of-study; ET = early termination; MAAE = medically attended adverse event; N/A = not applicable; PIMMC = potentially immune-mediated medical condition; SAE = serious adverse event; V = visit

[a] Telephone call assessment

[b] Prior to the completion of any study-related procedures

[c] Vital sign measurements include oral temperature, pulse rate, respiratory rate, and blood pressure. Vital signs will be measured before blood is collected. Blood pressure will be measured after subject is seated for at least 5 minutes. Vital signs will be taken both prevaccination and 30 ±5 minutes postvaccination on a vaccination day.

[d] If clinically indicated. The investigator in consultation with the medical monitor will determine whether safety laboratory assessments should be completed.

[e] Prior to vaccination

[f] Ask subject a standard nonleading question to elicit any medically related changes in their well-being. Ask whether they have been hospitalized, had any accidents, used any new medications, or changed concomitant medication regimens (both prescription and over the counter medications).

[g] Diary Cards I and III will be used to collect unsolicited and solicited local and systemic reactions 8 days after each vaccination, inclusive of vaccination day (from Day 1 through Day 8 and Day 29 through Day 36); Diary Card II will be used to collect unsolicited AEs from Day 8 through Day 29; Diary Card IV will be used to collect unsolicited AEs from Day 36 through Day 50. Thermometers and measuring tools will be provided to subjects at Visit 1 and replenished as needed.

[h] If applicable, according to the protocol period, collect and review diary card with subject.

[i] Use of all concomitant medications that the subject is taking at the time of the Screening Visit and vaccines received 3 months prior to the Screening Visit will be recorded in the subject's eCRF.

[j] Obtained by interview and by review of diary card completed by the subject.

[k] Any physical examination findings will be collected in the eCRF for medical history data presentation.

[l] Targeted physical examination will be performed if clinically indicated based on review of interim health status and/or clinical laboratory assessments. Any findings from this examination will be collected in the eCRF for AEs.

[m] Onsite urine pregnancy test must be performed at the Screening Visit and within 24 hours prior to each vaccination, and pregnancy test results must be final and negative prior to vaccination.

[n] Clinical safety laboratory tests include a complete blood count with differential, coagulation (partial thromboplastin time and prothrombin time (international normalized ratio) and chemistry including alanine aminotransferase, albumin, alkaline phosphatase, aspartate aminotransferase, bicarbonate, blood urea nitrogen, calcium, chloride, creatine kinase, creatinine, glucose (random, serum), potassium, sodium, total bilirubin, and total protein.

[o] At Visits 1 and 4, subjects will be monitored for solicited and unsolicited AEs, SAEs, and MAAEs for at least 30 minutes after vaccination. Any concomitant medications taken for these events will be recorded.

[p] At Visits 1 and 4, complete a 30 ±5 minute postvaccination injection site examination for erythema/redness and induration/swelling and monitor for pain. If a visible injection site abnormality is observed, document with a photograph, if subject consented.

[q] Examine vaccination site for erythema/redness and induration/swelling and monitor for pain. If a visible injection site abnormality is observed, document with a photograph, if subject consented.

[r] If the ET or unscheduled visit occurs within 7 days after either of the 2 study vaccinations.

[s] Only SAEs, MAAEs, and PIMMCs will be collected after Day 50 until the EoS Visit (13 months following the first vaccination).

[t] If subject declines to participate in an onsite ET visit, study staff should attempt to collect AE/SAE/MAAE/PIMMC and pregnancy information by phone.

APPENDIX 2. TABLES FOR CLINICAL AND LABORATORY ABNORMALITIES

The tables for clinical and laboratory abnormalities are presented in Table 12 and Table 13, respectively, according to the FDA Guidance for Industry: *Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials (September 2007).*

Table 12: Tables for Clinical Abnormalities

Local Reaction to Injectable Product	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**	**Potentially Life Threatening (Grade 4)**
Pain	Does not interfere with activity	Repeated use of non-narcotic pain reliever >24 hours or interferes with activity	Any use of narcotic pain reliever or prevents daily activity	Emergency room visit or hospitalization
Tenderness	Mild discomfort to touch	Discomfort with movement	Significant discomfort at rest	Emergency room visit or hospitalization
Erythema/redness[a]	2.5-5 cm	5.1-10 cm	>10 cm	Necrosis or exfoliative dermatitis
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	Necrosis

[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.

Vital Signs[a]	**Mild (Grade 1)**	**Moderate (Grade 2)**	**Severe (Grade 3)**	**Potentially Life Threatening (Grade 4)**
Fever (°C)[b] (°F)[b]	38.0-38.4 100.4-101.1	38.5-38.9 101.2-102.0	39.0-40 102.1-104	>40 >104
Tachycardia – beats per minute	101-115	116-130	>130	ER visit or hospitalization for arrhythmia
Bradycardia – beats per minute[c]	50-54	45-49	<45	ER visit or hospitalization for arrhythmia
Hypertension (systolic) – mm Hg	141-150	151-155	>155	ER visit or hospitalization for malignant hypertension
Hypertension (diastolic) – mm Hg	91-95	96-100	>100	ER visit or hospitalization for malignant hypertension
Hypotension (systolic) – mm Hg	85-89	80-84	<80	ER visit or hospitalization for hypotensive shock

Vital Signs[a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Respiratory rate – breaths per minute	17-20	21-25	>25	Intubation

ER = emergency room
[a] Subject should be at rest for all vital sign measurements.
[b] Oral temperature; no recent hot or cold beverages or smoking.
[c] When resting heart rate is between 60 to 100 beats per minute. Use clinical judgment when characterizing bradycardia among some healthy subject populations, for example, conditioned athletes.

Systemic (General)	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Nausea/vomiting	No interference with activity or 1-2 episodes/24 hours	Some interference with activity or >2 episodes/24 hours	Prevents daily activity, requires outpatient IV hydration	ER visit or hospitalization for hypotensive shock
Diarrhea	2-3 loose stools or <400 g/24 hours	4-5 stools or 400-800 g/24 hours	6 or more watery stools or >800 g/24 hours or requires outpatient IV hydration	ER visit or hospitalization
Headache	No interference with activity	Repeated use of non-narcotic pain reliever >24 hours or some interference with activity	Significant; any use of narcotic pain reliever or prevents daily activity	ER visit or hospitalization
Fatigue	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Myalgia	No interference with activity	Some interference with activity	Significant; prevents daily activity	ER visit or hospitalization
Illness or clinical adverse event (as defined according to applicable regulations)	No interference with activity	Some interference with activity not requiring medical intervention	Prevents daily activity and requires medical intervention	ER visit or hospitalization

ER = emergency room; IV = intravenous

Table 13: Tables for Laboratory Abnormalities

Serum[a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)[b]
Sodium – hyponatremia mEq/L	132-134	130-131	125-129	<125
Sodium – hypernatremia mEq/L	144-145	146-147	148-150	>150
Potassium – hyperkalemia mEq/L	5.1-5.2	5.3-5.4	5.5-5.6	>5.6
Potassium – hypokalemia mEq/L	3.5-3.6	3.3-3.4	3.1-3.2	<3.1

Serum[a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)[b]
Glucose – hypoglycemia mg/dL	65-69	55-64	45-54	<45
Glucose – hyperglycemia Fasting - mg/dL Random - mg/dL	 100-110 110-125	 111-125 126-200	 >125 >200	Insulin requirements or hyperosmolar coma
Blood urea nitrogen mg/dL	23-26	27-31	>31	Requires dialysis
Creatinine mg/dL	1.5-1.7	1.8-2.0	2.1-2.5	>2.5 or requires dialysis
Calcium-hypocalcemia mg/dL	8.0-8.4	7.5-7.9	7.0-7.4	<7.0
Calcium-hypercalcemia mg/dL	10.5-11.0	11.1-11.5	11.6-12.0	>12.0
Magnesium-hypomagnesemia mg/dL	1.3-1.5	1.1-1.2	0.9-1.0	<0.9
Phosphorus – hypophosphatemia mg/dL	2.3-2.5	2.0-2.2	1.6-1.9	<1.6
CPK - mg/dL	1.25-1.5 × ULN	1.6-3.0 × ULN	3.1-10 × ULN	>10 × ULN
Albumin – hypoalbuminemia g/dL	2.8-3.1	2.5-2.7	<2.5	–
Total protein – hypoproteinemia g/dL	5.5-6.0	5.0-5.4	<5.0	–
Alkaline phosphate – increase by factor	1.1-2.0 × ULN	2.1-3.0 × ULN	3.1-10 × ULN	>10 × ULN
Liver function tests – ALT, AST increase by factor	1.1-2.5 × ULN	2.6-5.0 × ULN	5.1-10 × ULN	>10 × ULN
Bilirubin – when accompanied by any increase in liver function test increase by factor	1.1-1.25 × ULN	1.26-1.5 × ULN	1.51-1.75 × ULN	>1.75 × ULN
Bilirubin – when liver function test is normal; increase by factor	1.1-1.5 × ULN	1.6-2.0 × ULN	2.0-3.0 × ULN	>3.0 × ULN
Cholesterol mg/dL	201-210	211-225	>226	-
Pancreatic enzymes – amylase, lipase	1.1-1.5 × ULN	1.6-2.0 × ULN	2.1-5.0 × ULN	>5.0 × ULN

ALT = alanine aminotransferase; AST = aspartate aminotransferase; CPK = creatine phosphokinase; ULN = upper limit of normal.

[a] The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

[b] The clinical signs or symptoms associated with laboratory abnormalities might result in characterization of the laboratory abnormalities as Potentially Life Threatening (Grade 4). For example, a low sodium value that falls within a grade 3 parameter (125-129 mE/L) should be recorded as a grade 4 hyponatremia event if the subject had a new seizure associated with the low sodium value.

Hematology[a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Hemoglobin (female) – g/dL	11.0-12.0	9.5-10.9	8.0-9.4	<8.0
Hemoglobin (female) change from baseline value – g/dL	Any decrease – 1.5	1.6-2.0	2.1-5.0	>5.0
Hemoglobin (male) – g/dL	12.5-13.5	10.5-12.4	8.5-10.4	<8.5
Hemoglobin (male) change from baseline value – g/dL	Any decrease – 1.5	1.6-2.0	2.1-5.0	>5.0
WBC increase – cell/mm^3	10,800-15,000	15,001-20,000	20,001-25,000	>25,000
WBC decrease – cell/mm^3	2,500-3,500	1,500-2,499	1,000-1,499	<1,000
Lymphocytes decrease – cell/mm^3	750-1 000	500-749	250-499	<250
Neutrophils decrease – cell/mm^3	1,500-2,000	1,000-1,499	500-999	<500
Eosinophils – cell/mm^3	650-1 500	1501-5 000	>5 000	Hypereosinophilic
Platelets decrease – cell/mm^3	125,000-140,000	100,000-124,000	25,000-99,000	<25,000
PT – increase by factor	1.0-1.10 × ULN	1.11-1.20 × ULN	1.21-1.25 × ULN	>1.25 x ULN
PTT – increase by factor	1.0-1.2 × ULN	1.21-1.4 × ULN	1.41-1.5 × ULN	>1.5 x ULN
Fibrinogen increase – mg/dL	400-500	501-600	>600	–
Fibrinogen decrease – mg/dL	150-200	125-149	100-124	<100 or associated with gross bleeding or disseminated intravascular coagulation

PPT = partial thromboplastin time; PT = prothrombin time; ULN = upper limit of normal; WBC = white blood cell.
[a] The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

Urine[a]	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Protein	Trace	1+	2+	Hospitalization or dialysis
Glucose	Trace	1+	2+	Hospitalization or hyperglycemia
Blood (microscopic) – red blood cells per high power field	1-10	11-50	>50 and/or gross blood	Hospitalization or packed red blood cells transfusion

[a] The laboratory values provided in the tables serve as guidelines and are dependent upon institutional normal parameters. Institutional normal reference ranges should be provided to demonstrate that they are appropriate.

APPENDIX 3. LIST OF POTENTIALLY IMMUNE-MEDIATED MEDICAL CONDITIONS

Gastrointestinal disorders

- Celiac disease
- Crohn's disease
- Ulcerative colitis
- Ulcerative proctitis

Liver disorders

- Autoimmune cholangitis
- Autoimmune hepatitis
- Primary biliary cirrhosis
- Primary sclerosing cholangitis

Metabolic diseases

- Addison's disease
- Autoimmune thyroiditis (including Hashimoto thyroiditis)
- Diabetes mellitus type I
- Grave's or Basedow's disease

Musculoskeletal disorders

- Antisynthetase syndrome
- Dermatomyositis
- Juvenile chronic arthritis (including Still's disease)
- Mixed connective tissue disorder
- Polymyalgia rheumatic
- Polymyositis
- Psoriatic arthropathy
- Relapsing polychondritis
- Rheumatoid arthritis
- Scleroderma, including diffuse systemic form and CREST (calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia) syndrome
- Spondyloarthritis, including ankylosing spondylitis, reactive arthritis (Reiter's Syndrome) and undifferentiated spondyloarthritis
- Systemic lupus erythematosus
- Systemic sclerosis

Neuroinflammatory disorders

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

Skin disorders

- Alopeci areata
- Autoimmune bullous skin diseases, including pemphigus, pemphigoid, and dermatitis herpetiformis
- Cutaneous lupus erythematosus
- Erythema nodosum
- Morphoea
- Lichen planus
- Psoriasis
- Sweet's syndrome
- Vitiligo

Vasculitides

- Large vessel vasculitis including giant cell arteritis such as Takayasu's arteritis and temporal arteritis
- Medium sized and/or small vessel vasculitis including polyarteritis nodosa, Kawasaki's disease, microscopic polyangiitis, Wegener's granulomatosis, Churg–Strauss syndrome (allergic granulomatous angiitis), Buerger's disease (thromboangiitis obliterans), necrotizing vasculitis and antineutrophil cytoplasmic antibody (ANCA)-positive vasculitis (type unspecified), Henoch-Schonlein purpura, Behcet's syndrome, leukocytoclastic vasculitis

Others

- Antiphospholipid syndrome

- Autoimmune hemolytic anemia
- Autoimmune glomerulonephritis (including IgA nephropathy, glomerulonephritis rapidly progressive, membranous glomerulonephritis, membranoproliferative glomerulonephritis, and mesangioproliferative glomerulonephritis)
- Autoimmune myocarditis/cardiomyopathy
- Autoimmune thrombocytopenia
- Goodpasture syndrome
- Idiopathic pulmonary fibrosis
- Pernicious anemia
- Raynaud's phenomenon
- Sarcoidosis
- Sjögren's syndrome
- Stevens-Johnson syndrome
- Uveitis