

CLINICAL STUDY PROTOCOL V122_01 Version 1.0

A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults

EUDRACT No. 2014-000145-69

BB-IND No. TBD

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PROTOCOL SYNOPSIS V122_01 V1.0

Name of Sponsor Novartis Vaccines and Diagnostics	Protocol number: V122_01	Health authority trial registration number(s): EudraCT No. 2014-000145-69 BB-IND No. TBD
Title of Study: A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults		
Study Period: Approximately 13 months for each subject.	Clinical Phase: Phase 1	
Rationale: Respiratory syncytial virus (RSV) is the most important cause of acute lower respiratory tract infections (ALRIs) that result in hospital visits during infancy and early childhood. In the United States, more than 60% of infants are infected by RSV during their first RSV season, and nearly all have been infected by two to three years of age (Breese, 2010; Glezen et al., 1986). Approximately 2.1 million US children less than five years of age are treated for RSV disease each year: 3% as inpatients, 25% in emergency departments, and 73% in pediatric practices (Hall et al., 2009). Disease burden is similar in Europe, with more than 1% of all children less than three years of age hospitalized annually and more than 7% receiving outpatient medical care (Forster et al., 2004). Globally, among children less than five years of age, RSV causes an estimated 33.8 million ALRIs each year (more than 22% of all ALRIs), resulting in 66,000-199,000 deaths, 99% of which occur in developing countries (Nair et al., 2010). In addition, RSV is a common cause of respiratory disease among the elderly, resulting in as many hospitalizations as influenza in a heavily influenza-immunized population (Falsey et al., 2005). RSV spreads by respiratory droplets and close contact with infected persons or contaminated objects. In temperate climates there is an annual winter epidemic (Brandt et al., 1973), while in tropical areas seasonality is less distinct, but infection is most common during the rainy season (Weber et al., 1998). Infants are at highest risk for severe RSV disease in their first six months (Iwane et al., 2004; Hall et al., 2009), and hospitalization peaks at two to three months of age (Parrott et al., 1973). Preterm birth and cardiopulmonary disease are risk factors for severe disease, with 33.8% of infants with bronchopulmonary dysplasia hospitalized for RSV in their first year of life (Boyce et al., 2000). Bronchiolitis is the classic presentation, but RSV also causes pneumonia, rhinitis, and otitis media. Severe RSV infection in infancy is associated with higher rates		

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of asthma later in childhood (Stein et al., 1999; Wu & Hartert, 2011). Recommended treatment of RSV bronchiolitis consists primarily of respiratory support and hydration (Subcommittee on diagnosis and management of bronchiolitis, 2006). No specific anti-viral therapy is recommended. The neutralizing monoclonal antibody (mAb), Palivizumab[®], is used for prophylaxis of infants at highest risk for severe RSV disease, and although the monoclonal antibody can protect these infants, it is too expensive and impractical for universal use (Prescott et al., 2010). Induction of neutralizing antibody (NAb) should be the primary goal of vaccination because the severity of RSV disease is largely determined by the extent of viral replication (Graham, 2011). Although developing a safe and effective RSV vaccine is a global public health priority and will have a significant impact on RSV prevention, there is no licensed RSV vaccine.

Novartis Vaccines and Diagnostics (NVD) has developed an investigational RSV subunit vaccine from an engineered recombinant RSV fusion (F) glycoprotein. The clinical program will evaluate the feasibility of passively protecting infants by immunizing pregnant women with the RSV F subunit vaccine. If maternal immunization during 24 to 32 weeks of gestation increases antibody titers eight-fold, the median peak of RSV disease in infants would be delayed from its current peak at two to three months of age to a new peak at approximately five to six months of age. This delay could significantly decrease the burden of RSV disease in infants in the first months of life and open an RSV disease-free interval during which active immunization of infants could further extend protection beyond six months of age. In the current study, we will test one dosage of the investigational RSV F subunit vaccine that is presumed to be below the dosage-response plateau (to establish that a maximal neutralizing antibody response cannot be achieved with a low dosage) and two higher dosages (to either establish the dosage-response plateau or indicate that higher dosages are needed in a subsequent study to reach the plateau).

The purpose of this study is to evaluate the safety and immunogenicity of two doses of the investigational RSV F subunit vaccine administered intramuscularly (IM). In this current Phase 1, first-in-human study, the three different antigen amounts that have been selected will be evaluated in a stepwise manner in three different cohorts (cohort 1: low dosage of RSV F subunit vaccine [45 µg], cohort 2: middle dosage of RSV F subunit vaccine [90 µg], and cohort 3: high dosage of RSV F subunit vaccine [135 µg]). In addition, the effect of an adjuvant, either aluminum hydroxide [Al(OH)₃] or MF59, and antibody kinetics post-vaccination at different time points will be evaluated as compared

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to unadjuvanted RSV F subunit vaccine at the same dosage levels.		
Objectives: <i>Primary Safety Objective:</i> 1. To assess the safety of the RSV F subunit vaccine compared to placebo. <i>Primary Immunogenicity Objective:</i> 1. To evaluate the serum neutralizing antibody (NAb) response to the RSV F subunit vaccine or placebo at Day 57 (28 days after the second dose). <i>Secondary Immunogenicity Objectives:</i> 1. To evaluate the serum neutralizing antibody (NAb) response to the RSV F subunit vaccine or placebo at Day 1 (baseline), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose). 2. To evaluate the total serum antibody responses to the RSV F subunit vaccine or placebo at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 3. To compare the ratio of RSV F subunit serum neutralizing antibody (NAb) titer to RSV F subunit serum total binding antibody titers to the RSV protein F in vaccine or placebo recipients at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 4. To compare the ratio of RSV F subunit serum neutralizing antibody (NAb) titer to RSV F subunit serum total binding antibody titers to each of the RSV proteins G and N in vaccine or placebo recipients at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).		

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<p>Exploratory Objectives:</p> <ol style="list-style-type: none"> 1. To characterize the serum neutralizing antibody (NAb) and binding antibody responses against additional RSV strains and native or engineered RSV antigens at baseline and following vaccination in a subset of subjects. 2. To determine the frequency of B cells specific for RSV proteins in a subset of subjects, and subsequently, explore the baseline immunity to RSV and the immune response to the RSV F subunit vaccine by analyzing the RSV-specific B-cell repertoire in a selected group of the subset of subjects. 		
<p>Methodology:</p> <p><i>Study Design:</i> In total, approximately 288 healthy non-pregnant female and male adults (18 to 45 years of age) will be enrolled in the study in the ratio of 3:1. There is a higher ratio of female to male subjects because the RSV F subunit vaccine is ultimately intended for use in pregnant women. Approximately 216 subjects will be assigned to receive three different formulations of the investigational vaccine and 72 will be assigned to receive placebo.</p> <p>The approximately 216 healthy subjects assigned to receive the investigational vaccine will be further randomized (1:1:1) in a stepwise dosage-escalation manner and assigned to one of three cohorts (cohort 1: low dosage of RSV F subunit vaccine [45 µg], cohort 2: middle dosage of RSV F subunit vaccine [90 µg], and cohort 3: high dosage of RSV F subunit vaccine [135 µg]). Within each cohort subjects will be randomly allocated (1:1:1) to receive vaccine with no adjuvant, with aluminum hydroxide, or with MF59, as outlined in Table 1. All subjects will receive two doses of a 0.5 mL intramuscular injection of the vaccine, with adjuvant (aluminum hydroxide [1 mg] or MF59 [0.25 mL; 9.75 mg squalene and surfactants]) or without adjuvant. There will be approximately 24 subjects in each treatment group. All the remaining 72 subjects (approximately 24 subjects per cohort) will be assigned to receive placebo (saline) (Table 1). The minimum interval between the first and the second doses will be 28 days.</p> <p><i>Vaccination Plan:</i> Eighteen (18) treatment groups (9 active treatment groups and 3 placebo [saline] groups in each cohort) will be evaluated as shown in Table 1:</p>		

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Table 1. Number of Subjects Randomized per Vaccine Treatment Group in Each Cohort

Cohort	Dosage	Route of Administration	Regimen: Two Doses †		
			No Adjuvant	Al(OH) ₃ ‡	MF59 ‡
1	45 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8
2	90 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8
3	135 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8

Abbreviations: IM, intramuscular; mL, milliliter; Al(OH)₃, aluminum hydroxide.

† Administered at least 28 days apart.

‡ Concentration of Al(OH)₃ will be 1 mg per dose and that of MF59 will be 0.25 mL (1X, which contains 9.75 mg squalene and surfactants).

Safety Measurements:

Solicited local and systemic adverse events (AEs) and unsolicited AEs will be collected using diary cards for 7 days following any dose of vaccine or placebo. Solicited local AEs include injection site pain, injection site induration, injection site swelling, and injection site erythema. Solicited systemic AEs include body temperature (oral or axillary), chills, nausea, generalized myalgia, generalized arthralgia, headache, fatigue, diarrhea, loss of appetite, cough, rhinorrhea, and wheezing. Safety telephone calls at 14 and 21 days after each dose of vaccine or placebo will be used to collect unsolicited AEs occurring since the last study contact, as well as any solicited AEs continuing beyond the 7 day period after vaccination. See [section 3.2.5](#) for further details.

In addition, all SAEs, new onset of chronic disease (NOCDs), and adverse events of special interest (AESIs) will be collected from the date of signed informed consent until the end of the study (approximately 393 days after the first dose of vaccine or placebo [saline]). Monitoring for AESIs (see [section 6.6.1.1](#) and [Appendix A](#)) will be extended through 12 months after the second vaccination and will be accomplished by clinic visits or telephone follow-up calls as detailed in [Table 2](#). All AESIs will be reported in the same manner as SAEs. A tabulation of all AESIs, categorized by MedDRA preferred terms and assessed relationship to study vaccine will be performed.

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Schedule of safety data collection:

- Solicited local and systemic AEs, body temperature, and other indicators of reactogenicity will be collected for 7 days following receipt of first and second dose of vaccine (using diary cards).
- All unsolicited AEs and solicited local and systemic AEs that continue beyond 7 days after vaccination will be collected for 28 days following receipt of first and second dose of vaccine.
- All SAEs, non-scheduled physician visits, unsolicited medically attended AEs, unsolicited AEs leading to study withdrawal, NOCDs, and AESIs will be collected from first vaccination to study completion.
- All concomitant medications administered in relation to the reported AEs will be collected from first vaccination to study completion.

Primary and Secondary Immunogenicity Measurements:

Four blood samples per subject will be collected for serum preparation and determination of immune responses to the RSV F subunit vaccine at Visit 1, Visit 5, Visit 9, and Visit 12 as outlined in [Table 2](#). Serum anti-RSV neutralizing antibody (NAb) titers will be evaluated for all subjects by a plaque reduction neutralization assay (PRNA); binding antibody to the RSV proteins F, G (subgroups G_a and G_b), and N will be evaluated by a microsphere-based fluorescence linked immunosorbent assay, multiplexed using Luminex® technology. See [section 6.3](#) for further details.

Safety Laboratory Parameters:

To assess laboratory AEs, blood and urine samples will be collected for each subject at Screening, Visit 1, Visit 2, Visit 5, Visit 6, and Visit 9 as outlined in [Table 2](#) to perform blood chemistry, hematology, and urine analyses for all subjects.

Exploratory Measurements:

The serum neutralizing antibody (NAb) and binding antibody responses against additional RSV strains and native or engineered RSV antigens at baseline and following vaccination will be performed in a subset of subjects, following evaluation of the primary and secondary immunogenicity evaluations and depending on volume of serum remaining for additional testing.

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<p>In approximately the first 10 subjects enrolled in each of the four treatment groups of cohort 3 (three vaccine groups and placebo) who agree to a separate Informed Consent, additional blood will be collected at Visit 1, Visit 2, Visit 5, and Visit 12 for cell mediated immunity assays. The purpose of these assays is to determine the frequency of B cells specific for RSV proteins by enzyme-linked immunosorbent spot (ELISPOT). Subsequently, the analysis of the RSV-specific B-cell repertoire will be performed in a subset of these subjects. See section 6.8 for further details.</p>		
<p>Number of Subjects planned:</p> <p>Sample size is not driven by statistical assumptions for formal hypothesis testing as previous clinical trial data are not available and clinically meaningful group differences are not established. In addition, values for a correlate of protection are not known at present stage. Nevertheless, some information from the neutralization assay for human serum was available.</p> <p>Hence, the number of proposed subjects will be sufficient to provide a descriptive summary of the safety and immunogenicity of the RSV F subunit vaccine. Anticipating that about 10% of the subjects will be non-evaluable (due to being lost to follow-up, having insufficient samples, incomplete laboratory results, or protocol violations, etc.), approximately 288 subjects will be randomized to 12 different treatment groups (approximately 24 subjects in each vaccine treatment group and approximately 24 subjects in each of the placebo cohorts), in order to obtain approximately 260 evaluable healthy adult subjects.</p>		
<p>Subject Population:</p> <p>Healthy adult volunteers between 18-45 years of age, inclusive.</p>		
<p>Subject Characteristics and Main Criteria for Inclusion and Exclusion:</p> <p>Key Inclusion (<i>abridged for the synopsis</i>):</p> <p>In order to participate in this study, all subjects must meet ALL of the inclusion criteria described.</p>		

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<p>1. Healthy males and non-pregnant females 18 to 45 years of age at time of enrollment.</p> <p>Key Exclusion (<i>abridged for the synopsis</i>):</p> <p>In order to participate in this study, all subjects must meet NONE of the exclusion criteria described.</p> <ol style="list-style-type: none">1. Individuals with any severe chronic or acute disease.2. Individuals with a history of illness or with an ongoing illness that may pose additional risk to the subject if he/she participates in the study, including the following:<ul style="list-style-type: none">- History of any chronic respiratory illness, including current diagnosis of asthma within 2 years, exercise induced wheezing, reactive airway disease, emphysema, chronic bronchitis, cystic fibrosis or chronic obstructive pulmonary disease (COPD).- Any respiratory illness (e.g. cough, sore throat, dyspnea, wheezing or nocturnal awakenings to respiratory symptoms) within 7 days prior to receiving the first study vaccination.- Any active pulmonary infection or other inflammatory conditions, even in the absence of febrile episodes, within 14 days prior to the first study vaccination.- Hepatitis B or hepatitis C infection.3. Individuals participating in any clinical trial with another investigational product 28 days prior to receiving the first study vaccination or intent to participate in another clinical study at any time during the conduct of this study.4. Individuals who have received any vaccine 28 days prior to enrollment in this study, or who plan to receive any non-study vaccines within 28 days of the second dose of study vaccine.5. If female, 'of childbearing potential', sexually active and has not used any of the 'acceptable contraceptive methods' for at least two months prior to study entry. <p>Childbearing potential is defined as status post onset of menarche and not meeting any of the following conditions: menopausal for at least two years; sterile status</p>		

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<p>after bilateral tubal ligation for at least one year, immediately after bilateral oophorectomy or after hysterectomy.</p> <p>Acceptable methods of birth control are defined as one or more of the following:</p> <ul style="list-style-type: none">- Hormonal contraceptives (such as oral, injection, transdermal patch, implant, cervical ring).- Barrier (condom with spermicide or diaphragm with spermicide) each and every time during intercourse.- Intrauterine device (IUD).- Monogamous relationship with vasectomized partner. Partner must have been vasectomized for at least six months prior to subject's study entry. <p>6. If female subject of childbearing potential and have a positive urine pregnancy test prior to study injections, or are currently lactating.</p> <p>7. If female of childbearing potential and sexually active, refusal to use an 'acceptable contraceptive method' through to three weeks after last study vaccination.</p> <p>The full list of inclusion and exclusion criteria is included in protocol section 4.</p>		
<p>Vaccines:</p> <p>The RSV F subunit vaccine is a lyophilized vaccine consisting of 135 µg of an engineered recombinant RSV fusion (F) glycoprotein. The vaccine for this study will be provided in a lyophilized pellet and reconstituted and mixed in saline and/or adjuvant by the unblinded designated site staff who is to follow a procedure as described in the Reconstitution Instructions in the Investigator Site File (i.e., the vaccine is prepared in the clinic prior administration).</p> <p>The lyophilized product is reconstituted with (a) sterile saline 0.9% NaCl to generate the non-adjuvanted RSV F subunit vaccine formulation, (b) in aluminum hydroxide [Al(OH)₃] to generate the RSV F subunit vaccine formulated in Al(OH)₃, or (c) sterile saline 0.9% NaCl and adjuvant MF59 to generate the RSV F subunit vaccine formulated with MF59. Preparation for each formulation of the RSV F subunit vaccine will require a separate kit and detailed instructions for reconstitution and dilution steps. See section</p>		

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<p>5.3 for further details.</p> <p>In each cohort, subjects will be randomized to receive two injections of the vaccine or placebo (saline). The vaccine will be reconstituted in 0.5 mL of saline and/or adjuvant and injected intramuscularly (IM) in the deltoid muscle, with preference that the injection is administered in the non-dominant arm.</p>		
<p>Safety Endpoints:</p> <p>Safety will be assessed by measuring the frequency of local and systemic solicited adverse events (AEs), unsolicited AEs, serious adverse events (SAEs), new onset of chronic diseases (NOCD), adverse events of special interest (AESIs), and safety laboratory data for all subjects. Tolerability will be assessed by the joint evaluation of both reactogenicity and unsolicited AEs.</p> <ul style="list-style-type: none">▫ Reactogenicity will be assessed by the percentage and frequency of subjects with solicited local and solicited systemic adverse events up to 7 days after each vaccination.▫ Unsolicited AEs will be assessed by the percentage and frequency of subjects with any unsolicited AEs for 28 days after each vaccination.▫ Percentage and frequency of subjects with SAEs, non-scheduled physician visits, unsolicited medically attended AEs, unsolicited AEs leading to study withdrawal, NOCDs, and AESIs will be collected from study start until study completion. <p>Primary Immunogenicity Endpoints:</p> <ol style="list-style-type: none">1. Geometric mean titer (GMT) of the serum anti-RSV neutralizing antibody (NAb) titer at Day 57 (28 days after the second dose).2. Proportion of subjects with a ≥ 4-fold increase in serum anti-RSV NAb titer from Day 1 (baseline) to Day 57 (28 days after the second dose). <p>Secondary Immunogenicity Endpoints:</p> <ol style="list-style-type: none">1. GMT of the serum anti-RSV NAb titer at Day 1 (baseline), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose).2. Proportion of subjects with a ≥ 4-fold increase in serum anti-RSV NAb titer from		

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<p>Day 1 (baseline) to all other time points (Day 29 [28 days after the first dose] and Day 181 [six months after the first dose]).</p> <ol style="list-style-type: none"> 3. Proportion of subjects at Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose) who achieve serum anti-RSV NAb titers greater than the 3rd quartile of the serum anti-RSV NAb titers overall distribution at Day 1 (baseline). 4. Reverse cumulative distribution of serum anti-RSV NAb titers at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 5. GMT of the serum total binding antibody to each of the RSV proteins F, G, and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 6. Proportion of subjects with a ≥ 4-fold increase in serum total binding antibody titers to each of the RSV proteins F, G, and N from Day 1 (baseline) to all time points (Day 29 [28 days after the first dose], Day 57 [28 days after the second dose], and Day 181 [six months after the first dose]). 7. Proportion of subjects at Days 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose) who achieve serum total binding antibody titers to each of the RSV proteins F, G, and N greater than the 3rd quartile of serum total binding antibody titers to RSV protein F at Day 1 (baseline). 8. Reverse cumulative distribution of serum total binding antibody to each of the RSV proteins F, G, and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 9. Ratio of RSV F subunit serum neutralizing antibody (NAb) titer to each of the RSV F subunit serum total binding antibody titers to RSV proteins F, G, and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose). 		
<p>Exploratory Endpoints:</p> <ol style="list-style-type: none"> 1. Predominant isotype of the RSV-specific serum antibody at multiple time points. 2. RSV-specific immune response against different RSV group A and group B strains 		

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<p>or engineered RSV antigens at multiple time points.</p> <ol style="list-style-type: none"> 3. Frequency of B cells secreting RSV-specific antibodies at Day 1 (baseline), Day 8 (seven days after the first dose), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose). 4. Diversity of the B-cell repertoire at Day 1 (baseline), Day 8 (seven days after the first dose), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose). 		
<p>Statistical Analysis of Primary Objective(s):</p> <p>Values below the detection limit (1:100) will be set at 1:50.</p> <p><u>GMTs:</u> Antibody concentrations will be logarithmically transformed (base 10). For each treatment group within each cohort, GMTs of the serum anti-RSV NAb along with their associated 95% confidence intervals (CIs) will be computed by exponentiation of the corresponding log-transformed means and 95% CIs.</p> <p><u>Fold-rise:</u> Proportions of subjects with a ≥ 4-fold increase in serum anti-RSV NAb titer from baseline will be presented, for each treatment group within each cohort, together with their two-sided 95% Clopper-Pearson CIs.</p> <p>The statistical analyses for GMTs will be conducted using an ANCOVA model with dosage and adjuvant as factors and baseline antibody level as covariate. Interactions between factors will be investigated and further described in the statistical analysis plan to be finalized and approved before the start of the trial.</p> <p>The primary immunogenicity analyses will be based on a per-protocol (PP) population. If the full analysis set (FAS) and PP differ by more than 10%, primary immunogenicity analyses will also be conducted on the FAS.</p>		
<p>Interim Analysis:</p> <p>An Interim Analysis will be performed on the immunogenicity and safety data collected from all subjects at Visit 1, Visit 5, and Visit 9. Further details regarding the interim</p>		

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analysis are contained in [section 7.5](#) of the protocol.

Data Monitoring Committee:

A Data Monitoring Committee (DMC) will be implemented to review safety data during scheduled periodic reviews. The DMC will review safety data, as described in the DMC charter and in the statistical analysis plan, after enrollment of the first 12 subjects in each cohort, and shortly after Visit 2 (seven days after the first dose), before proceeding with enrollment of the remaining subjects in each cohort. In addition, the DMC will review safety data after enrollment of each cohort has been completed before proceeding with enrollment of the subsequent cohort. Please see [section 6.9](#) of the protocol for more details.

Table 2. Times and Events Table

Study Period	Screen	Vaccine Dose #1 & Safety Follow-up				Vaccine Dose #2 & Safety Follow-up					Post-vaccination Follow-up				
		Clinic Visit	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Clinic Visit	Clinic Visit	Clinic Visit	Phone Call
Study Day	-21 to -3	1	3, 5	8	15, 22	29	31, 33	36	43, 50	57	85	113	181	209	271, 366, 394
Days post 1 st dose		0	2, 4	7	14, 21	28	30, 32	35	42, 49	56					
Days post 2 nd dose						0	2, 4	7	14, 21	28	56				
Study Visit Window (min-max) in Days (d)	n/a	n/a	n/a	-1/ +4 d	n/a	+14 d	n/a	-3/ +3 d	n/a	-7/ +7 d	-7/ +7 d	-7/ +7 d	-7/ +7 d	-14/ +14 d	-7/ +7 d
Visit Number	Screening	1		2	3, 4	5		6	7, 8	9	10	11	12	13	14, 15, 16
Informed Consent	X														
Medical History	X														
Physical Exam ^a	X	X ^b		X		X ^b		X		X	X	X	X	X	
Safety Laboratory Blood draw [max: 10 mL whole blood] ^c	X	X ^b		X		X ^b		X		X					
Urinalysis ^d	X	X ^b		X		X ^b		X		X					
Pregnancy Test	X	X ^b				X ^b									
Exclusion/Inclusion Criteria	X														
Randomization		X ^b													
Serology Blood draw [max: 15 mL whole blood] ^e		X ^b				X ^b				X			X		

Study Period	Screen	Vaccine Dose #1 & Safety Follow-up				Vaccine Dose #2 & Safety Follow-up					Post-vaccination Follow-up				
Type of Visit	Clinic Visit	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Clinic Visit	Clinic Visit	Clinic Visit	Phone Call	
Study Day	-21 to -3	1	3, 5	8	15, 22	29	31, 33	36	43, 50	57	85	113	181	209	271, 366, 394
Days post 1 st dose		0	2, 4	7	14, 21	28	30, 32	35	42, 49	56					
Days post 2 nd dose						0	2, 4	7	14, 21	28	56				
Study Visit Window (min-max) in Days (d)	n/a	n/a	n/a	-1/ +4 d	n/a	+14 d	n/a	-3/ +3 d	n/a	-7/ +7 d	-7/ +7 d	-7/ +7 d	-7/ +7 d	-14/ +14 d	-7/ +7 d
Visit Number	Screening	1		2	3, 4	5		6	7, 8	9	10	11	12	13	14, 15, 16
2° Exploratory Obj. Blood Draw [max: 50 mL whole blood] ^f		X ^b		X		X ^b							X		
Study Vaccine Administered ^g		V				V									
30 Minutes Post Injection Assessment (Local/ Systemic AEs, Body Temperature) ^h		X				X									
Local/Systemic AEs, Body Temperature, Other Indicators of Reactogenicity ⁱ		X				X									
Diary Card Training ^j		X				X									
Diary Card Dispensed ^k		X				X									
Diary Card Reminder Call ^l			X				X								

Study Period	Screen	Vaccine Dose #1 & Safety Follow-up				Vaccine Dose #2 & Safety Follow-up					Post-vaccination Follow-up				
Type of Visit	Clinic Visit	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Reminder Call	Clinic Visit	Phone Call	Clinic Visit	Clinic Visit	Clinic Visit	Clinic Visit	Clinic Visit	Phone Call
Study Day	-21 to -3	1	3, 5	8	15, 22	29	31, 33	36	43, 50	57	85	113	181	209	271, 366, 394
Days post 1 st dose		0	2, 4	7	14, 21	28	30, 32	35	42, 49	56					
Days post 2 nd dose						0	2, 4	7	14, 21	28	56				
Study Visit Window (min-max) in Days (d)	n/a	n/a	n/a	-1/ +4 d	n/a	+14 d	n/a	-3/ +3 d	n/a	-7/ +7 d	-7/ +7 d	-7/ +7 d	-7/ +7 d	-14/ +14 d	-7/ +7 d
Visit Number	Screening	1		2	3, 4	5		6	7, 8	9	10	11	12	13	14, 15, 16
Diary Card Reviewed and Collected ^m				X				X							
Telephone Contact for Review of Safety Data ⁿ					X				X						
Assess all AEs, including SAEs and NOCDs Leading or not to Study Withdrawal ^o		X		X		X		X		X	X	X	X	X	X
Assess for AESIs ^p		X		X		X		X		X	X	X	X	X	X
Prior/Concomitant Medications/vaccines ^q	X	X		X		X		X		X	X	X	X	X	
Study Termination ^r															X

- Physical examination must be performed by a qualified health professional in accordance with local regulations and licensing requirements designated within the Site Responsibility Delegation Log. See [section 6.2](#) for components of physical examination by visit.
- Procedure to be performed prior to vaccination.
- Safety laboratory assays that will be included are listed in [section 3.5.3](#).

- d. Urinalysis testing will include at a minimum protein, glucose, and presence of red blood cells.
- e. Maximal blood draw refers to volume drawn for each type of assay at each specified visit. See [section 3.5.1](#) for greater detail regarding blood sampling volumes.
- f. The first 10 subjects enrolled in each treatment group of Cohort 3 who agree to the additional blood draws will need to sign an additional consent prior to the blood draw for the secondary exploratory objective.
- g. Subjects will receive two doses of vaccine or placebo according to the study randomization scheme.
- h. A 30 minute post-injection local and systemic adverse event and body temperature measurement will be performed by the subject under site staff supervision at the clinic during Visit 1 and Visit 5.
- i. Beginning 6 hours following study vaccine administration at Visit 1 and Visit 5, and daily thereafter through 7 days after each vaccination, solicited local and systemic adverse events including other reactions (i.e. body temperature measurements and use of analgesics/antipyretics) will be reported daily by the subject on a diary card.
- j. Subjects will receive training on the diary card at Visit 1 and Visit 5.
- k. The diary card will be dispensed at Visit 1 and Visit 5, and subjects will be reminded that diary cards must be returned at the next clinic visit (Visit 2 or Visit 6).
- l. Site staff will contact subjects by phone 2 and 4 days after each vaccination to remind them to complete their diary cards each day, and to bring the diary cards to their next clinic visit.
- m. Review of safety data captured on diary cards will be completed at Visit 2 and Visit 6. Diary cards will be collected and stored with subject files.
- n. Safety data will be collected for 28 days following each vaccination. At 14 and 21 days after each dose, subjects will be interviewed by site staff using a scripted interview for collection of safety data. These safety data will be transcribed on source documents by the site staff performing the interviews.
- o. All medically attended AEs that lead to an unscheduled visit to a healthcare practitioner and/or a visit to the emergency department or its equivalent will be collected for 28 days following signing of the informed consent. SAEs, NOCDs and AEs leading to study or vaccine withdrawal will be collected through 1 year after receipt of the second dose. Please see [section 6.6](#) for greater detail regarding methods for SAE and AEs leading to study or vaccine withdrawal collection.
- p. Adverse events of special interest (AESIs) will also be documented in all study subjects for the duration of the study. Monitoring for AESIs will be extended through 1 year after receipt of the second dose and will be accomplished at clinic visits or by a telephone follow-up call. A tabulation of all AESIs, categorized by MedDRA preferred terms and assessed relationship to study vaccine will be submitted as an addendum to the Clinical Study Report (CSR) if not included in the CSR.
- q. Collect concomitant medications and vaccination history according to the study procedures outlined in protocol [section 3.2.5](#) and [5.4](#).
- r. Any subject who terminates the study after receipt of vaccine (prior to Visit 9) is recommended to undergo study-related procedures required at Visit 9. For subjects who terminate after Visit 9, a telephone contact to assess for SAEs/AEs and associated concomitant medications is required.

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LIST OF ABBREVIATIONS

ACIP	Advisory Committee on Immunization Practices
AE	Adverse Event
AESI	Adverse Events of Special Interest
ALRI	Acute Lower Respiratory Infection
ALT	Alanine Aminotransferase
Al(OH) ₃	Aluminum hydroxide
ANCOVA	Analysis of Covariance
AP	(Statistical) Analysis Plan
AST	Aspartate Aminotransferase
BCDM	Biostatistics and Clinical Data Management
B&SR	Biostatistics and Statistical Reporting
BMI	Body Mass Index
BUN	Blood Urea Nitrogen
CBC	Complete Blood Count
CBER	Center for Biologics Evaluation, Research and Review
CDC	Centers for Disease Control and Prevention
cDNA	Complementary Deoxyribonucleic acid
CFR	Code of Federal Regulations
CHMP	Committee for Human Medicinal Products for Human Use
CI	Confidence Interval
CMI	Cell Mediated Immunity
COPD	Chronic Obstructive Pulmonary Disease
CPK	Creatinine Phosphokinase
CRF	Case Report Form
CRO	Contract Research Organization
CRP	C-reactive Protein
CSR	Clinical Study Report
DCF	Data Clarification Form
DMC	Data Monitoring Committee
EC	Ethics Committee
eCRF	Electronic Case Report Form
EDC	Electronic Data Capture
eDiary	Electronic Diary
EDT	Electronic Data Transfer
ELISA	Enzyme-linked Immunosorbent Assay
ELISPOT	Enzyme-linked Immunosorbent Spot
EMA	European Medicines Agency
F	Fusion (glycoprotein)
FAS	Full Analysis Set

FDA	Food and Drug Administration
FI	Formalin-inactivated
FSFV	First Subject First Visit
γ GT	Gamma-glutamyl Transpeptidase
GCP	Good Clinical Practices
GLP	Good Laboratory Practices
GMC	Geometric Mean Concentration
GMR	Geometric Mean Ratio
GMT	Geometric Mean Titer
HEENT	Head, Ears, Eyes, Nose, and Throat
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human Immunodeficiency Virus
IB	Investigator's Brochure
ICF	Informed Consent Form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ID	Identification (Subject ID), Intradermal
Ig	Immunoglobulin
IUD	Intrauterine Device
IM	Intramuscular
IND	Investigational New Drug
INR	International Normalized Ratio
IRB	Institutional Review Board
ITT	Intention-To-Treat
IVRS	Interactive Voice Response System
IWRS	Interactive Web Response System
LDH	Lactate Dehydrogenase
LSLV	Last Subject Last Visit
mAb	Monoclonal antibody
MedDRA	Medical Dictionary for Regulatory Activities
mg	Milligrams
MITT	Modified Intention-To-Treat
mL	Milliliters
MPV	Mean Platelet Volume
NAb	Neutralizing antibody
NCR	Non Carbon-Copy Paper
NOCD	New onset of chronic disease
NVD	Novartis Vaccines and Diagnostics
PBMC	Peripheral Blood Mononuclear Cells
PCR	Polymerase Chain Reaction
PIV	Paramyxovirus

PP	Per Protocol
PPT	Partial Thromboplastin Time
PRNA	Plaque Reduction Neutralization Assay
PRO	Patient Reported Outcome
PT	Prothrombin Time
RBC	Red Blood Cell
RSV	Respiratory Syncytial Virus
SAE	Serious Adverse Event
SDA	Source Data Agreement
SOC	System Organ Class
SOP	Standard Operating Procedure
SPC	Summary of Product Characteristics
SUSAR	Suspected Unexpected Serious Adverse Reaction
VSAE	Vaccine Serious Adverse Event
WBC	White Blood Cell
WHO	World Health Organization
µg	Microgram

1.0 BACKGROUND AND RATIONALE

A comprehensive review of the RSV F subunit vaccine is contained in the Investigator's Brochure (IB) supplied by NVD; this document should be reviewed prior to initiating the study.

Respiratory syncytial virus (RSV) is the most important cause of acute lower respiratory tract infections (ALRIs) that result in hospital visits during infancy and early childhood. In the United States, more than 60% of infants are infected by RSV during their first RSV season, and nearly all have been infected by two to three years of age (Breese, 2010; Glezen et al., 1986). Approximately 2.1 million US children less than five years of age are treated for RSV disease each year: 3% as inpatients, 25% in emergency departments, and 73% in pediatric practices (Hall et al., 2009). Disease burden is similar in Europe, with more than 1% of all children less than three years of age hospitalized annually and more than 7% receiving outpatient medical care (Forster et al., 2004). Globally, among children less than five years of age, RSV causes an estimated 33.8 million ALRIs each year (more than 22% of all ALRIs), resulting in 66,000-199,000 deaths, 99% of which occur in developing countries (Nair et al., 2010). In addition, RSV is a common cause of respiratory disease among the elderly, resulting in as many hospitalizations as influenza in a heavily influenza-immunized population (Falsey et al., 2005).

RSV spreads by respiratory droplets and close contact with infected persons or contaminated objects. In temperate climates there is an annual winter epidemic (Brandt et al., 1973). In tropical areas seasonality is less distinct, but infection is most common during the rainy season (Weber et al., 1998). Infants are at highest risk for severe RSV disease in their first six months (Iwane et al., 2004; Hall et al., 2009), and hospitalization peaks at two to three months of age (Parrott et al., 1973). Preterm birth and cardiopulmonary disease are risk factors for severe disease, with 33.8% of infants with bronchopulmonary dysplasia hospitalized for RSV in their first year of life (Boyce et al., 2000). Bronchiolitis is the classic presentation of RSV. RSV also causes pneumonia, rhinitis, and otitis media. Severe RSV infection in infancy is associated with higher rates of asthma later in childhood (Stein et al., 1999; Wu & Hartert, 2006). Recommended treatment of RSV bronchiolitis consists primarily of respiratory support and hydration (Subcommittee on diagnosis and management of bronchiolitis, 2006). No specific anti-viral therapy is recommended. The neutralizing monoclonal antibody (mAb), Palivizumab®, directed to a single epitope of the RSV fusion glycoprotein (F), is used for prophylaxis of infants at highest risk for severe RSV disease, and although the monoclonal antibody can protect these infants, it is too expensive and impractical for universal use (Prescott et al., 2010). Induction of neutralizing antibody (NAb) should be the primary goal of vaccination because the severity of RSV disease is largely determined by the extent of viral replication (Graham, 2011). Although developing a safe and

effective RSV vaccine is a global public health priority and will have a significant impact on RSV prevention, there is no licensed RSV vaccine.

A history of RSV vaccine-mediated disease enhancement raises safety concerns for vaccine studies in RSV-naive infants. In a key RSV vaccine clinical trial in the late 1960s, infants and young children were immunized with a formalin-inactivated whole virion RSV preparation (FI-RSV) or an equivalent paramyxovirus control preparation (FI-PIV). Five percent of the subjects who were immunized with FI-PIV and then naturally infected by RSV during the next RSV season were hospitalized; 80% of those who were immunized with FI-RSV and then infected by RSV were hospitalized, and two of them died (Kim et al., 1969). FI-RSV-mediated RSV disease enhancement has been attributed to FI-RSV's failure to elicit neutralizing antibodies combined with its priming for an exaggerated, Th2-predominant immune response to subsequent RSV infection (Bouhvalova et al., 2006; Graham et al., 1993; Kalina et al., 2004; Connors et al., 1994; De Swart et al., 2002; Polack et al., 2002; The Impact-RSV Study Group, 1998; Shaw et al., 2013).

Given the early age of peak RSV disease (Iwane et al., 2004; Hall et al., 2009; Parrott et al., 1973), an RSV vaccine would ideally be administered to neonates and young infants, elicit neutralizing antibodies, and prime for an immune response to RSV infection that prevents rather than enhances RSV disease. However, it is difficult to actively immunize infants to protect them by two to three months of age because time must elapse after immunization for a neutralizing antibody response, and more than one dose of vaccine may be needed. Direct infant immunization is also challenged by the inhibitory effects of maternal antibody (Crowe, 2001; Wood & Siegrist, 2011; Philbin & Levy, 2009; Shaw et al., 2013) and the immaturity of the neonatal immune system, which has impaired antigen-presenting cell responses, Th2-biased cellular responses, and impaired antibody affinity maturation.

Novartis Vaccines and Diagnostics (NVD) has developed an investigational RSV subunit vaccine from an engineered recombinant RSV fusion (F) glycoprotein. The engineered recombinant F antigen is modified from the wild type F glycoprotein of RSV strain A2, a laboratory group A prototype strain isolated in 1961 from an infant with bronchiolitis. The wild type RSV F glycoprotein is a homotrimer that is anchored in the viral envelope by a C-terminal transmembrane region. The RSV F protein mediates virus-cell fusion, is highly conserved among RSV isolates, and is a major target of neutralizing antibodies (Shaw et al., 2013). The clinical program will evaluate the feasibility of passively protecting infants by immunizing pregnant women with the RSV F subunit vaccine. The inverse correlation between maternal, cord blood, or infant serum RSV neutralizing titers and early RSV disease in infants demonstrates that maternal antibody can protect infants (Glezen et al., 1981; Piedra et al., 2003). Although a precise correlate of protection has not yet been agreed, studies of passive protection by maternal antibody or administered

immunoglobulin and studies of RSV infection of cotton rats suggest that infant serum neutralizing titers of approximately 1:200 to 1:400 prevent severe RSV disease (Piedra et al., 2003; Groothuis et al., 1993; Prince et al., 1985; Siber et al., 1994; Shaw et al., 2013). The half-life of maternal antibody in infants predicts that each doubling of titer should extend passive infant protection by approximately one month (Ochola et al., 2009; Brandenburg et al., 1997). If maternal immunization during 24 to 32 weeks of gestation increases antibody titers eight-fold, the median peak of RSV disease in infants would be delayed by approximately three months from its current peak at two to three months of age to a new peak at approximately five to six months of age. This delay could significantly decrease the burden of RSV disease in infants in the first months of life and open an RSV disease-free interval during which active immunization of infants could further extend protection beyond six months of age.

Because the duration of protection of an infant receiving transplacental antibody from an immunized pregnant woman is expected to be directly dependent on the woman's neutralizing titer to RSV, there is no maximum desirable neutralization titer. This is an important distinction between immunization of a subject to actively protect that subject and immunization of a pregnant woman to passively protect her infant from a disease with an extended period of vulnerability after birth. With immunization for active immunity, achievement of a threshold immune response indicates that the subject's immune system is sufficiently primed to protect against future disease, often remote future disease, and the goal is to achieve that threshold response. On the other hand, with immunization for passive protection, the infant receives a finite placental transfer of antibody before birth, and that protective antibody decays to zero over time, leaving no priming of the immune system of the infant to protect against future and distant infections. As a consequence, the goal for passive immunization is to elicit the highest level of neutralizing antibodies in the pregnant subject that can be achieved safely, tolerably, and practically, so that a maximal neutralizing antibody titer is transferred to the infant before birth, providing the longest possible period of passive protection after birth. Given the extended vulnerability of infants to severe RSV infections, at least through two years of age, and the approximately one month half-life of passively transferred antibody (Breese, 2010; Glezen et al., 1986; Hall et al., 2009; Ochola et al., 2009; Brandenburg et al., 1997), any achievable maternal neutralization titer is unlikely to protect throughout the complete period of vulnerability.

Preclinical animal experiments with the investigational RSV F subunit vaccine have shown different effects of dosage levels, number of doses, and need for adjuvant, depending on the RSV infection history of the animal. The difference between the responses of RSV-naïve and recently RSV-infected animals complicates extrapolation of the results to immunization of adult humans, who are likely to have been more remotely infected. In RSV-naïve cotton rats or mice, aluminum hydroxide [Al(OH)₃] or MF59 was required for high titer neutralizing responses. There was a clear dosage level-response for

both adjuvanted and unadjuvanted RSV F subunit vaccine (two intramuscular [IM] injections at a three week interval) in mice, with a plateau achieved with approximately 3.3 µg of aluminum hydroxide-adjuvanted and 10 µg of unadjuvanted RSV F subunit vaccine, respectively. Maximum neutralizing titers of approximately 1:1,000 in mice and 1:7,000 in cotton rats were less than two-fold higher with aluminum hydroxide than with MF59 in both species. However, cotton rats and mice infected intranasally with RSV that were subsequently immunized once IM with 5 µg, 0.5 µg, or 0.05 µg of RSV F subunit vaccine seven weeks later (cotton rats) or eleven weeks later (mice), showed a four-fold increase (cotton rats) and 10 to 50-fold increase (mice) in neutralizing titers after immunization – but the neutralizing titers were minimally impacted by the RSV F subunit vaccine dosage level, by the inclusion of adjuvant (none, aluminum hydroxide, or MF59), or by a second vaccine injection given three weeks after the first.

The discrepancy between the effect of dosage level in previously RSV-infected and RSV-naïve rodents, the unknown (and possibly long) expected interval between natural RSV infection and immunization of adult humans, the lack of predictable scaling of dosage-response with body size, and species-specific adjuvant effects make the animal data a poor guide to dosage level and regimen determination for testing the RSV F subunit vaccine in humans. Therefore, previous clinical trial experience with other F-containing, non-replicating RSV vaccines, rather than the animal data, serve as the primary guide for the initial selection of dosage levels for testing of Novartis's RSV F subunit vaccine candidate in humans. Available clinical trial results suggest that adult humans respond in a manner that is intermediate between the RSV-naïve and previously RSV-infected mice and cotton rats, and require higher dosage levels than those used to immunize rodents (Glenn et al., 2013; Muñoz et al., 2003; Falsey & Walsh, 1996; Falsey & Walsh, 1997; Falsey et al., 2008; Langley et al., 2009).

Because the adult human population is seropositive for RSV, RSV vaccination in adults will act as a booster. Boosting immunity in RSV-seropositive individuals may be challenging and may require use of adjuvant. Aluminum hydroxide [Al(OH)₃] and aluminum phosphate salts are the most commonly used adjuvant in licensed vaccines worldwide and they have a well-established safety profile, including in pregnant women (Baylor et al., 2002; Silveira et al., 1995; Blencowe et al., 2010; Murphy et al., 2008; ACIP, 2011; Mast et al., 2005; Levy & Koren, 1991; Tsai et al., 2010; Vecchi et al., 2012). MF59 is a nanoemulsion of squalene, polysorbate 80, sorbitan trioleate, and citrate. MF59 is used in subunit influenza vaccines that are licensed in European and other countries (not including the United States), such as Flud[®] (seasonal), Aflunov[®] (H5N1 pre-pandemic), Foclivia[®] (H5N1 pandemic), Focetria[®] (H1N1 pandemic), and Celtura[®] (cell culture-produced H1N1 pandemic). More than 100 million doses of MF59-adjuvanted influenza vaccines have been distributed in licensed products. Although MF59-adjuvanted vaccines are somewhat more reactogenic than equivalent non-adjuvanted vaccines, clinical trials and post-marketing surveillance demonstrate MF59's

good safety and tolerability profile (O'Hagan 2007; 2013; Pellegrini et al., 2009; Tsai et al., 2010; Schultze et al., 2008; Black et al., 2010). An estimate of >90,000 pregnant women were vaccinated with Focetria during the 2009 pandemic. Preliminary data from observational studies, spontaneously reported events, and ongoing post-marketing studies (a pregnancy registry and a prospective interventional study) have provided no evidence of direct or indirect harmful effects to mother or fetus from the use of MF59-adjuvanted vaccines during pregnancy (Banzhoff et al., 2011; Parretta et al., 2011; Zuccotti et al., 2010; Huang et al., 2011). The mechanism of action of adjuvants is not completely understood, but both aluminum hydroxide and MF59 are thought to increase local cytokines and chemokines and increase cell recruitment (including eosinophils for aluminum hydroxide, neutrophils for MF59, and monocytes and macrophages for both) (Awate et al., 2013). Aluminum hydroxide is also proposed to increase antigen presentation, while MF59 increases antigen uptake, increases antigen-loaded neutrophils and monocytes, and activates muscle cells (Awate et al., 2013). In total, approximately 150 million doses of MF59-adjuvanted vaccines have been distributed (O'Hagan et al., 2013). No safety concerns have been observed related to MF59 (O'Hagan et al., 2013). Similarly, hundreds of millions of doses of aluminum-containing adjuvants have been administered without any major safety findings (Lamprecht et al., 2009).

Recently, Novavax (Rockville, MD) performed a Phase 1 randomized, observer-blinded, placebo-controlled clinical trial with a similar recombinant RSV F subunit vaccine to evaluate its safety and immunogenicity in healthy adults (Glenn et al., 2013). The results of the Phase 1 clinical trial showed that 30 µg and 60 µg (with or without aluminum phosphate) induced a two-fold increase in neutralizing titers after two IM injections; the two-fold increase in neutralizing titers was negatively correlated with baseline neutralizing titers. The small magnitude of the increase of neutralizing titers and large range of baseline neutralizing titers led to variable increases in neutralizing titers with dosage level or use of adjuvant. Dosage level-responses for RSV F binding antibodies (including those that compete with Palivizumab®) covered a broader range (Glenn et al., 2013). There was no plateau in binding antibody titers between the two highest dosage levels tested, 30 and 60 µg, but there were increases in binding titers with aluminum phosphate adjuvant and a modest increase with a second injection (a maximum 1.6-fold increase at 5 µg or 15 µg with aluminum phosphate and a smaller increase at higher dosage levels) (Glenn et al., 2013). A subsequent clinical trial is currently being performed in women of child bearing age with 60 and 90 µg of their RSV F subunit vaccine candidate. Although other trials in the literature do not provide as clearly relevant information (Muñoz et al., 2003; Falsey & Walsh, 1996; Falsey & Walsh, 1997; Falsey et al., 2008; Langley et al., 2009; Tristram et al., 1993; Belshe et al., 1993; Paradiso et al., 1994; Piedra et al., 2003), their results are broadly consistent with those of the Novavax Phase 1 clinical trial (Glenn et al., 2013), which closely followed the intended immunization regimen for the NVD investigational RSV F subunit vaccine

candidate. In previous clinical trials of former (or still current) non-replicating RSV F subunit vaccine candidates, including trials conducted in RSV seropositive infants, typical dosage levels of RSV antigens tested IM have been in the range of 5 to 100 µg per dose (Glenn et al., 2013; Muñoz et al., 2003; Falsey & Walsh, 1996; Falsey & Walsh, 1997; Falsey et al., 2008; Langley et al., 2009; Tristram et al., 1993; Belshe et al., 1993; Paradiso et al., 1994; Piedra et al., 2003).

To provide the longest period of protection from RSV disease in infants, the goal is to maximize neutralizing antibody titers in pregnant women through immunization. In the current study, we will test one dosage of the investigational RSV F subunit vaccine that is presumed to be below the dosage-response plateau (to establish that a maximal neutralizing antibody response cannot be achieved with a low dosage) and two higher dosages (to either establish the dosage-response plateau or indicate that higher dosages are needed in a subsequent study to reach the plateau). Therefore, for the current study, two doses (administered IM at least 28 days apart) of three dosage levels of the investigational RSV F subunit vaccine will be tested in healthy adults: 45 µg, 90 µg, and 135 µg. The two-dose regimen was selected because clinical data with a similar vaccine candidate (Glenn et al., 2013) have shown an increase in titers following the second dose. Based on the previous clinical experience with similar vaccine candidates, the lowest dosage level (45 µg) is not anticipated to be on the dosage-response plateau. In a non-GLP preclinical toxicology study in rabbits, two simultaneous injections of 135 µg of the RSV F subunit vaccine candidate were well-tolerated, and the preclinical safety and tolerability of a 135 µg dosage will be confirmed in the GLP preclinical toxicology study before this Phase 1 study starts. Based on these constraints, the two upper dosages, 90 µg and 135 µg, have been selected to maximize the chances that a dosage-response plateau will be established in this study.

The purpose of this study is to evaluate the safety and immunogenicity of two doses of the investigational RSV F subunit vaccine administered IM. In this current Phase 1, first-in-human study, the three different antigen amounts that have been selected will be evaluated in a stepwise manner in three different cohorts (cohort 1: low dosage of RSV F subunit vaccine [45 µg], cohort 2: middle dosage of RSV F subunit vaccine [90 µg], and cohort 3: high dosage of RSV F subunit vaccine [135 µg]). In addition, the effect of an adjuvant, either aluminum hydroxide [Al(OH)₃] or MF59, and antibody kinetics post-vaccination at different time points will be evaluated as compared to unadjuvanted RSV F subunit vaccine at the same dosage levels. The trial will be conducted in compliance with the protocol, GCP and applicable regulatory requirement(s).

2.0 OBJECTIVES

2.1 Primary Objective(s)

Primary Immunogenicity Objective

1. To evaluate the serum neutralizing antibody (NAb) response to the RSV F subunit vaccine or placebo at Day 57 (28 days after the second dose).

Primary Safety Objective

1. To assess the safety of the RSV F subunit vaccine compared to placebo.

2.2 Secondary Objectives

Secondary Immunogenicity Objectives

1. To evaluate the serum neutralizing antibody (NAb) response to the RSV F subunit vaccine or placebo at Day 1 (baseline), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose).
2. To evaluate the total serum antibody responses to the RSV F subunit vaccine or placebo at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).
3. To compare the ratio of RSV F subunit serum neutralizing antibody (NAb) titer to RSV F subunit serum total binding antibody titers to the RSV protein F in vaccine or placebo recipients at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).
4. To compare the ratio of RSV F subunit serum neutralizing antibody (NAb) titer to RSV F subunit serum total binding antibody titers to each of the RSV proteins G and N in vaccine or placebo recipients at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).

Secondary Exploratory Objectives

1. To characterize the serum neutralizing antibody (NAb) and binding antibody responses against additional RSV strains and native or engineered RSV antigens at baseline and following vaccination in a subset of subjects.

2. To determine the frequency of B cells specific for RSV proteins in a subset of subjects, and subsequently, explore the baseline immunity to RSV and the immune response to the RSV F subunit vaccine by analyzing the RSV-specific B-cell repertoire in a selected group of the subset of subjects.

3.0 STUDY DESIGN AND INVESTIGATIONAL PLAN

3.1 Overview of Study Design

This is a Phase 1, randomized, observer blind, placebo-controlled, dosage-escalation, single center study, enrolling healthy adults. In total, approximately 288 healthy non-pregnant female and male adults (18 to 45 years of age) will be enrolled in the study in the ratio of 3:1. There is a higher ratio of female to male subjects because the RSV F subunit vaccine is ultimately intended for use in pregnant women. Men are still being included in this study because the vaccine may also be tested in the future in elderly subjects.

Approximately 216 healthy subjects will be randomized to receive the investigational vaccine. Subjects will be enrolled (1:1:1) in a stepwise dosage-escalation manner in to one of three cohorts (cohort 1: low dosage of RSV F subunit vaccine [45 µg], cohort 2: middle dosage of RSV F subunit vaccine [90 µg], and cohort 3: high dosage of RSV F subunit vaccine [135 µg]). Cohort 1 will be enrolled first, followed by cohort 2, and finally cohort 3. Within each cohort subjects will be randomly allocated (1:1:1) to receive vaccine with no adjuvant, with aluminum hydroxide, or with MF59, as outlined in [Table 1](#). All subjects will receive two doses of a 0.5 mL intramuscular injection of the vaccine, with adjuvant (aluminum hydroxide [1 mg] or MF59 [0.25 mL; 9.75 mg squalene and surfactants]) or without adjuvant. There will be approximately 24 subjects in each treatment group. All the remaining 72 subjects will be assigned to receive placebo (saline) (approximately 24 subjects per cohort) ([Table 1](#)).

The minimum interval between doses will be 28 days. The study duration for a given subject will be approximately 13 months. An Interim Analysis will be performed on the immunogenicity and safety data collected from all subjects at Visit 1 (baseline), Visit 5 (28 days after the first dose), and Visit 9 (28 days after the second dose). Further details regarding the interim analysis are contained in [section 7.5](#).

Vaccination Plan: Eighteen (18) treatment groups (9 active treatment groups and 3 placebo [saline] groups in each cohort) will be evaluated as shown in [Table 1](#):

Table 3.1-1: Number of Subjects Randomized per Vaccine Treatment Group in Each Cohort

Cohort	Dosage	Route of Administration	Regimen: Two Doses †		
			No Adjuvant	Al(OH) ₃ ‡	MF59 ‡
1	45 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8
2	90 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8
3	135 µg	IM (0.5 mL)	24	24	24
	Placebo (saline)	IM (0.5 mL)	8	8	8

Abbreviations: IM, intramuscular; mL, milliliter; Al(OH)₃, aluminum hydroxide.

† Administered at least 28 days apart.

‡ Concentration of Al(OH)₃ will be 1 mg per dose and that of MF59 will be 0.25 mL (1X, which contains 9.75 mg squalene and surfactants).

3.1.1 Study Period

The expected duration of the study for an individual subject is approximately 13 months.

3.2 Study Visit Procedures

The sections that follow provide an overview of the procedures that are to be followed in enrolling, evaluating, and following subjects who participate in this clinical trial.

3.2.1 Informed Consent

"Informed consent" is the voluntary agreement of an individual or his/her legal guardian to participate in research. Consent must be given with free will of choice, and without undue inducement. The individual must have sufficient knowledge and understanding of the nature of the proposed research, the anticipated risks and potential benefits, and the requirements of the research to be able to make an informed decision.

Informed consent following local IRB/EC guidance **must** be obtained before conducting any study-specific procedure (i.e., all of the procedures described in the protocol). The process of obtaining informed consent should be documented in the subject source documents in addition to maintaining a copy of the signed and dated informed consent.

If a subject is unable to read, an impartial witness should be present during the entire informed consent discussion. An impartial witness is defined as a person who is independent from trial conduct, who cannot be unfairly influenced by those involved with

the trial, who attends the informed consent process if the subject or the subject's legally acceptable representative cannot read, and who reads the informed consent form and any other written information supplied to the subject. After the written informed consent form and any other written information to be provided to subjects, is read and explained to the subject and after the subject has verbally consented to the subject's participation in the trial and, if capable of doing so, has signed and personally dated the informed consent form, the witness should sign and personally date the consent form. By signing the consent form, the witness attests that the information in the consent form and any other written information was accurately explained to, and apparently understood by, the subject and that informed consent was freely given by the subject.

3.2.2 Screening Procedures

After an individual has consented to participate in the study and informed consent/assent is signed, that individual will be given a unique screening number which is documented in the Screening & Enrollment Log. Screening procedures will include the following:

- Medical history, including but not limited to:
 - Prior vaccinations
 - Concomitant medications
 - Previous and ongoing illnesses or injuries
- Physical examination, including but not limited to:
 - Vital signs
 - Assessment of general appearance
 - Examination of head, ears, eyes, nose, and throat (HEENT)
 - Examination of skin
 - Auscultation of heart and lungs
 - Palpation of abdomen
 - Examination of extremities
- Safety laboratory blood draw
- Urinalysis
- Pregnancy test (for female subjects)

See [section 3.5.3](#), [section 6.2](#), and [Table 2](#) for more information.

In the event that the individual is determined ineligible for study participation, he/she is considered a “screen failure”. The reason for screen failure must be documented in the Screening & Enrollment Log. If the individual is determined to be eligible for the study, he/she should be enrolled into the study as described in [section 3.2.3](#).

3.2.3 Enrollment

After an individual is determined to be eligible for study participation, the subject will be enrolled using a computerized randomization system that automatically assigns a unique Subject ID. The Subject ID consists of a 7 digit number resulting from the combination of the site number and the subject’s order of randomization at the site. Access to the system can be obtained by the site staff either via web or telephone (as back up).

Enrollment of all three cohorts will not occur simultaneously. Instead, subjects will be enrolled in to each cohort in a step-wise manner based on the dosage-escalation design. The first 12 subjects will be enrolled in cohort 1, a safety review of cohort 1 data through Visit 2 will be performed by the DMC, and then the remaining subjects in cohort 1 will be enrolled. After a safety review of cohort 1 data through Visit 9 is performed by the DMC, the first 12 subjects will be enrolled in cohort 2. After the safety review of cohort 2 data through Visit 2 is completed the remainder of cohort 2 subjects will be enrolled. Once safety review of cohort 2 data through Visit 9 has been completed by the DMC, the first 12 subjects of cohort 3 will be enrolled. Finally, cohort 3 enrollment will be completed after DMC review of cohort 3 safety data through Visit 2.

3.2.4 Randomization

Subjects will be randomly assigned to study groups in a pre-specified ratio as described in [section 3.1](#). The computerized randomization system will allocate the subject to one of the study groups and assign a Pack Number corresponding to the pack containing the treatment to be administered to the subject according to the randomization list. The list of randomization assignments will be produced by the service provider and approved by NVD Biostatistics and Clinical Data Management department (BCDM) according to applicable NVD SOP.

If for any reason, after signing the informed consent form (ICF), the subject (who has passed screening) fails to be randomized, the reason for not being randomized should be recorded on source documents as specified in the source data agreement. The information on these subjects, who are randomization failures, should be kept distinct in the source documentation from screen failures, which are described in [section 3.2.2](#).

3.2.5 Visit Procedures

The following sections provide an overview of procedures to be performed during the study. Where applicable, references to other sections of the protocol are provided for further details. Timing of procedures can also be found in [Table 2](#), Times and Events Table. Unless otherwise stated, all tests and procedures described in the protocol are required.

3.2.5.1 Pre-vaccination Procedures

The following procedures will be performed at a Screening visit prior to the day of first vaccination:

1. Informed consent (refer to [sections 3.2.1](#) and [12.2](#) for further detail)
2. Demography: refer to [section 6.2](#) for details
 - a. prior vaccination and concomitant medication (refer to [section 5.4](#) for further detail)
 - b. medical history: refer to [section 6.2](#) for details
3. Review of systems: a structured interview that queries the subject as to any complaints the subject has experienced across each organ system
4. “General” physical examination: refer to [section 6.2](#) for details
5. Height and weight: height will be measured in centimeters while standing; weight will be measured in kilograms
6. Safety laboratory blood draw: blood will be drawn to perform safety laboratory assessments (refer to [section 3.5.3](#) for further detail)
7. Urinalysis: a urine sample will be collected (refer to [section 3.5.3](#) for further detail)
8. Pregnancy testing: all women of child-bearing potential will be tested; refer to [section 3.5.2](#) for guidance regarding the procedure
9. Review of eligibility criteria: refer to [sections 4.1](#) and [4.2](#) for the list

The following procedures will be performed at Visit 1 and Visit 5 prior to vaccination:

1. Randomization (Visit 1 only): refer to [section 3.2.4](#)
2. Limited physical examination: refer to [section 6.2](#) for details
3. Blood sampling: Prior to study vaccination, blood will be drawn from the subjects for serology and safety laboratory assessments. For subjects in cohort 3 who sign an

additional Informed Consent for exploratory immunology assays, additional blood will be collected (see [section 3.5.4](#)). Details regarding the volume of blood and testing to be performed are in [sections 3.5.1](#), [3.5.3](#), and [3.5.4](#).

4. Urinalysis: a urine sample will be collected (refer to [section 3.5.3](#) for further detail)
5. Pregnancy testing: all women of child-bearing potential will be tested; refer to [section 3.5.2](#) for guidance regarding the procedure

3.2.5.2 Vaccination Procedures

Subjects will receive two (2) doses of vaccine according to their assigned treatment in a blinded fashion. Vaccination will be performed at Visit 1 and Visit 5. The minimum interval between vaccinations will be 28 days.

After confirming eligibility and enrolling subject into the study at Visit 1, perform vaccination of the subject according to the assigned study vaccine and according to the procedures described in [section 5.3](#) and observing the blinding procedures described in [section 3.3](#). At Visit 5, confirm that the subject does not meet any criteria for delaying or cancelling additional study vaccinations, as described in [section 4.3](#) and [section 4.4](#) of the protocol.

3.2.5.3 Post-vaccination Procedures

The following post-vaccination procedures will be performed at Visit 1 and Visit 5.

1. Subjects will be provided with a diary card at Visit 1 and Visit 5.
2. Careful training of the subject on how to measure local reactions and body temperature, how to complete and how often to complete the diary card is crucial. Training should be directed at the individual(s) who will perform the measurements of reactions and those who will enter the information into the diary card. This individual can be someone other than the subject, but if a person other than the subject enters information into the diary card, this person's identity must be documented in the study file and this person must receive training on the diary card. Training of the subject on how to measure an injection site reaction should be performed while the subject is under observation after vaccination.
3. The subject must understand that timely completion of the diary card on a daily basis is a critical component to study participation. The subject should also be instructed to write clearly and to complete the diary card in pen. Any corrections to the diary card that are performed by the person completing the diary card should include a single

strikethrough line with a brief explanation for any change. No changes can be made to the diary card when it is returned to the clinic.

4. Starting on the day of vaccination, the subject will check in the evening for specific types of reactions at the injection site (solicited local adverse events), any specific generalized symptoms (solicited systemic adverse events), body temperature (taken preferably orally), any other symptoms or change in the subject's health status, and any medications taken (excluding vitamins and minerals). These solicited adverse events and body temperature will be recorded in the "six hour" location on the diary card.
5. Body temperature measurement is to be performed using the thermometer provided by the site. If the subject feels unusually hot or cold during the day, the subject should check body temperature. If the subject has a fever, the highest body temperature observed that day should be recorded on the diary card. The measurement of solicited local adverse events is to be performed using the ruler provided by the site. The collection of body temperature, solicited local adverse events, and solicited systemic adverse events will continue for a total of 7 days on the diary card. The collection of unsolicited adverse events and medications will continue for 7 days on the diary card, (or until the subject returns for the next clinic visit).
6. After vaccination, the subject will be observed for at least 30 minutes including observation for unsolicited adverse events, solicited adverse events, and body temperature measurement. The site staff should take the opportunity to remind the subject how to measure solicited reactions and body temperature as part of this observation period. Record all safety data collected in the subject's source documents.
7. The site should schedule the next study activity (reminder call) with the subject.
8. The subject will receive a written reminder of the next planned study activity. The subject will be reminded to complete the diary card daily and to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.

3.2.5.4 Reminder Telephone Calls

1. Reminder calls will be performed 2 and 4 days after each vaccination.
2. The purpose of this call is to remind the subject about completion of the diary card. It is a conversation that follows the reminder telephone call script provided to the site, and it is not intended to be a call for collection of safety data. If the subject wishes to describe safety information, this information should only be collected by a trained

healthcare professional at the site, and the safety data described must be written down on source documents. The subject should be reminded to write the information down in the diary card and to contact the site via the telephone number provided in the informed consent to discuss medical questions.

3.2.5.5 Clinic Visits after Vaccination

Clinic visits that do NOT include vaccine administration include Visits 2, 6, 9, 10, 11, 12, and 13. The following procedures will be performed at the indicated clinic visits:

1. Visit 2 and Visit 6:
 - a. The diary card will be reviewed and collected. Please see [section 3.4.1](#) for additional guidance on diary card review. The subject will be interviewed to determine if any unsolicited adverse events occurred and if any concomitant medications or vaccines were taken/received in the time since the last clinic visit. The healthcare professional reviewing these data will discuss the symptoms (if any) reported by the subject and will determine if any additional diagnoses and/or adverse events are present. All AEs, SAEs, NOCDs (see [section 6.6.1.2](#)), or AESIs (see [section 6.6.1.1](#)) will be recorded on source documents. All medications taken or vaccines received will also be recorded on the source documents.
 - b. A symptom-directed physical exam will be performed (see [section 6.2](#)).
 - c. Blood will be collected for safety laboratory assessments (see [section 3.5.3](#)).
 - d. A urine sample will be collected for urinalysis (see [section 3.5.3](#)).
 - e. For subjects in cohort 3 who sign an additional Informed Consent for exploratory immunology assays, additional blood will be collected (Visit 2 only; see [section 3.5.4](#)).
 - f. The site should schedule the next study activity (safety call) with the subject.
 - g. The subject will receive a written reminder of the next planned study activity. The subject will be reminded to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.
2. Visit 9:
 - a. The subject will be interviewed to determine if any unsolicited adverse events occurred and if any concomitant medications or vaccines were taken/received in

the time since the last clinic visit. The healthcare professional reviewing these data will discuss the symptoms (if any) reported by the subject and will determine if any additional diagnoses and/or adverse events are present. All AEs, SAEs, NOCDs, or AESIs will be recorded on source documents. All medications taken or vaccines received will also be recorded on the source documents.

- b. A symptom-directed physical exam will be performed (see [section 6.2](#)).
 - c. Blood will be collected for safety laboratory assessments (see [section 3.5.3](#)).
 - d. A urine sample will be collected for urinalysis (see [section 3.5.3](#)).
 - e. Blood for serology will be drawn (see [section 3.5.1](#)).
 - f. The site should schedule the next study activity (clinic visit) with the subject.
 - g. The subject will receive a written reminder of the next planned study activity. The subject will be reminded to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.
3. Visits 10, 11, and 13:
- a. The subject will be interviewed to determine if any unsolicited adverse events occurred and if any concomitant medications or vaccines were taken/received in the time since the last clinic visit. The healthcare professional reviewing these data will discuss the symptoms (if any) reported by the subject and will determine if any additional diagnoses and/or adverse events are present. All AEs, SAEs, NOCDs, or AESIs will be recorded on source documents. All medications taken or vaccines received will also be recorded on the source documents.
 - b. A symptom-directed physical exam will be performed (see [section 6.2](#)).
 - c. The site should schedule the next study activity (clinic visit or safety call) with the subject.
 - d. The subject will receive a written reminder of the next planned study activity. The subject will be reminded to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.
4. Visit 12:
- a. The subject will be interviewed to determine if any unsolicited adverse events occurred and if any concomitant medications or vaccines were taken/received in the time since the last clinic visit. The healthcare professional reviewing these data will discuss the symptoms (if any) reported by the subject and will determine

if any additional diagnoses and/or adverse events are present. All AEs, SAEs, NOCDs, or AESIs will be recorded on source documents. All medications taken or vaccines received will also be recorded on the source documents.

- b. A symptom-directed physical exam will be performed (see [section 6.2](#)).
- c. Blood for serology will be drawn (see [section 3.5.1](#)).
- d. For subjects in cohort 3 who sign an additional Informed Consent for exploratory immunology assays (approximately 40 subjects in total, 10 from each treatment group), additional blood will be collected (see [section 3.5.4](#)).
- e. The site should schedule the next study activity (clinic visit) with the subject.
- f. The subject will receive a written reminder of the next planned study activity. The subject will be reminded to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.

3.2.5.6 Safety Calls

Safety telephone calls will be performed for Visits 3, 4, 7, and 8 (14 and 21 days after each dose). Additional safety telephone calls will be performed for Visits 14 and 15 (9 and 12 months after the first dose).

1. Safety telephone calls are calls made to the subject by a trained healthcare provider. These calls will follow a script which will facilitate the collection of relevant safety information. The subject will be interviewed according to the script, and information relating to unsolicited adverse events, including serious adverse events (SAEs), adverse events of special interest (AESIs), new onset of chronic disease, and AEs leading to study or vaccine withdrawal, and concomitant medications or vaccinations associated with those events. All safety information described by the subject must be written down in a designated location within the source documents and not written on the script used for the telephone call.
2. The site should schedule the next study activity (clinic visit, reminder call, or safety call) with the subject.
3. The subject will be reminded to contact the site if there are any questions and to contact the site immediately (or as soon as the subject is medically stable) if the subject has a medical condition that leads to a hospitalization or an emergency room visit.

3.2.5.7 “For cause” Visits

Not applicable for this study.

3.2.5.8 Termination Visits

1. The termination visit will occur at Visit 16 (12 months after the second dose). For visit procedures to be performed for a subject whose planned study participation ends prematurely, please see [section 3.8](#).
2. During the phone call, the following procedures will be performed:
 - a. The subject will be interviewed to determine if any unsolicited adverse events occurred and if any concomitant medications or vaccines were taken/received in the time since the last clinic visit.
 - b. The healthcare professional reviewing these data will discuss the symptoms (if any) reported by the subject and will determine if any additional diagnoses and/or adverse events are present.
 - c. All AEs, SAEs, NOCDs, or AESIs will be recorded on source documents.
 - d. All medications taken or vaccines received will be recorded on the source documents.
3. After thanking the subject for study participation, the site will review the plan of when information relating to the subject’s participation in the study may be available (e.g., study results, treatment assignments). It will also be discussed how information relating to the subject’s participation in the study will be shared with the subject’s healthcare provider, if the subject chooses to share this information.

The site will complete the termination CRF page and this will mark the completion of the subject’s participation in the study.

3.3 Blinding Procedures

This is an observer-blind study. The investigator, subjects, monitors, site personnel, and laboratory staff will be blinded to the vaccine administered, with the following exceptions:

- An unblinded designee (i.e., unblinded pharmacist or equivalent) will be assigned to administer the vaccine to all subjects. If deemed necessary, a second unblinded designee may be allowed to assist with vaccine preparation. The designee(s) will have no other study functions than vaccine management, documentation, accountability,

and administration. They will not be involved in subject evaluations. They will not reveal the vaccine treatment assignment to either the subject or the site personnel involved in the conduct of the study, unless this information is necessary in the case of an emergency.

The vaccine treatment assignment will be concealed to the extent possible by having the unblinded designee(s) (i.e., unblinded pharmacist) prepare the vaccine out of view of the subject or other site personnel. Each vaccine treatment group will receive a single 0.5 mL dose at each vaccination visit. In case the color or appearance of different dosages of the vaccine (or the placebo) differ, the subject will be shielded from view of the syringe and injected arm.

A Data Monitoring Committee (DMC) will perform a review of safety data after Visit 2 (seven days after the first dose) has been completed by the first 12 subjects in each cohort, and after Visit 9 (28 days after the second dose) has been completed for all subjects in each cohort. Details of the DMC review of safety data and stopping guidelines can be found in the DMC Charter.

An interim analysis of immunogenicity and safety data will be performed after results of serology assays for all samples through Visit 9 (28 days after the second dose) from all 3 cohorts are available. The analysis will include serology results from all subjects at Visits 1, 5, and 9. This interim analysis will be used to evaluate the optimal dosage and/or adjuvant for use in future clinical trials. An unblinded statistician from NVD will perform the interim analysis; results of the interim analysis will be confirmed by the DMC. Interim safety assessments will be performed by the DMC according to the DMC charter. The DMC will determine whether any safety issues exist. An unblinded statistician from NVD may assist with the evaluations. In the event a safety issue arises, the cluster physician and NVD pharmacovigilance department will be informed.

Note: Emergency unblinding should only be undertaken when it is essential to protect an individual subject's safety. There are limited circumstances in a vaccine study where unblinding would change the course of medical treatment or care of a subject.

For an emergency unblinding, the investigator will retrieve the subject's assigned vaccine from the computerized randomization system either via web or phone (as backup). Details of the unblinding procedures will be described in the system's Site User manual.

3.4 Data Collection

3.4.1 Data Collected From Subjects

All data collected from subjects and provided to the sponsor for analysis must be stripped of any identifiers that reveal the identity of that individual (beyond the use of subject ID, as described in [section 3.2.3](#)).

The use of any written or verbal information identifying the subject such as name, initials, photos or testimonials, requires separate and appropriate documented consent from each subject.

Diary cards will be the only source document allowed for solicited systemic and local adverse events (including body temperature measurements). The following additional rules apply to documentation of safety information collected by diary card:

1. No corrections or additions to the diary card will be allowed after it is delivered to the site.
2. Any blank or illegible fields on the diary card must be described as missing in the CRF.
3. The site must enter all readable entries in the diary card into the CRF, including those values that may be biologically implausible (e.g. body temperature: 400°C).
4. Any illegible or implausible data should be reviewed with the subject to determine if an underlying solicited or unsolicited adverse event is described. For example, if the subject with a body temperature of 400°C describes that the body temperature was actually 40°C on the day in which body temperature: 400°C was written into the diary card, this fever of 40°C should be described in the study file and reported as an unsolicited adverse event **in the adverse event CRF**.
5. Any newly described safety information (including a solicited reaction) must **NOT** be written into the diary card and must be described in the study file as a verbally reported event. Any adverse reaction reported in this fashion must be described as an unsolicited reaction and therefore entered on the adverse event CRF.

3.4.2 Electronic Case Report Forms

Electronic CRFs will be used in this study. Data will be entered into the database from the paper source documents, diaries, or the VSAE form. Paper CRFs will be used in this study for pregnancy and pregnancy follow-up CRFs. All data will be recorded by the investigator and/or the investigator's dedicated site staff. See [section 9.1](#) for more details.

3.5 Laboratory Assessments

3.5.1 Processing, Labeling and Storage of Serum Samples for Serology

A minimum of approximately 10 mL sample of blood will be drawn from all subjects at Visit 1 before vaccination, and at Visits 5, 9, and 12 for serology assays. The blood volume drawn for serology will not exceed 15 mL at each time point in order to provide the necessary serum volume (approximately half of the blood draw volume) for the serology assays. Additional blood samples may be drawn at these time points for safety labs and additional immunology assays.

Please refer to the Laboratory Manual in the Investigator Site File for instructions on drawing and processing blood samples, including the minimum volume of blood required and what to do in the case of failed attempts at drawing blood. Please see [section 6.3](#) for details on the serology assays and analysis of serology data.

Samples will be retained in accordance with regulatory guidance for retention of essential study documents as described in [section 10](#).

3.5.2 Pregnancy Testing

A urine pregnancy test will be performed at Screening, Visit 1, and Visit 5, at the study site prior to vaccination for all female subjects of childbearing potential (see [section 4.2](#)). Subjects must agree in the informed consent to use an appropriate method of birth control through 3 weeks after the last study vaccination at Visit 5 (i.e., until the Visit 8 safety call). Please refer to the instructions included with the test kits when performing urine pregnancy tests. Any subject with a positive urine pregnancy test at screening or Visit 1 will be excluded from the study. Any subject with a positive urine pregnancy test prior to receipt of the second vaccination at Visit 5 will not be vaccinated, and must be followed for safety and pregnancy outcomes. Results of pregnancy testing must be recorded in the source documents.

3.5.3 Safety Laboratory Assessments

A minimum of approximately 8 mL sample of blood will be drawn from all subjects at Screening, Visit 1 before vaccination, Visit 2, Visit 5 before vaccination, Visit 6, and Visit 9 for safety labs. The blood volume drawn for safety labs will not exceed 10 mL at each time point. Additional blood samples may be drawn at these time points for serology and immunology assays. Please refer to the Laboratory Manual in the Investigator Site File for instructions on drawing blood and shipping samples to the safety laboratory, including the minimum volume of blood required and what to do in the case of failed attempts at drawing blood.

Safety laboratory assessments from serum will include:

- Sodium
- Potassium
- Chloride
- Bicarbonate (or CO₂)
- Glucose
- Blood urea nitrogen (BUN)
- Calcium
- Magnesium
- Phosphorous
- Creatinine
- Creatine phosphokinase (CPK)
- Albumin
- Total protein
- Alkaline phosphatase
- Liver function tests (ALT, AST, γGT)
- Total bilirubin
- Direct bilirubin
- Indirect bilirubin
- Cholesterol
- LDH
- Pancreatic enzymes (amylase, lipase)

Hematology tests will include:

- CBC test (WBC, WBC differential, RBC, RBC indices, hemoglobin, hematocrit, platelets, and mean platelet volume [MPV])
- Fibrinogen
- Prothrombin time (PT)

- Partial thromboplastin time (PPT)
- International Normalized Ratio (INR)
- C-reactive protein (CRP)
- D-dimer

A urine sample will be collected from all subjects at Screening, Visit 1 before vaccination, Visit 2, Visit 5 before vaccination, Visit 6, and Visit 9. Please refer to the Laboratory Manual in the Investigator Site File for instructions on collecting urine samples and shipping samples to the safety laboratory, including the minimum volume required.

Safety laboratory assessments from urine will include (but may not be limited to):

- Protein
- Glucose
- Presence of red blood cells

The investigator MUST assess all safety laboratory results. If a laboratory result is outside the normal range based on the institution's normal values or CBER guidance (see [Appendix D, E, and F](#)), it must be classified as clinically significant or not. If the laboratory result is determined to be clinically significant it must be reported in the subject's medical history (in the case of screening and baseline samples tested prior to vaccination) or as an adverse event.

3.5.4 Cell Mediated Immunity Assessments

Cell mediated immunity assays will be performed in approximately the first 10 subjects of each treatment group (no adjuvant, aluminum hydroxide, MF59, or placebo) enrolled in cohort 3 who sign an additional informed consent for the additional blood draws needed to perform these assays. Approximately 40 subjects total will be included in these assays. Blood samples from subjects in cohort 3 will be tested because these subjects will receive the highest dosage of vaccine (excluding placebo recipients) and are most likely to develop an immune response. Approximately 50 mL of blood will be drawn from subjects at Visit 1 before vaccination, Visit 2, Visit 5 before vaccination, and Visit 12. The blood volume drawn for cell mediated immunity assays will not exceed 50 mL at each time point in order to provide the necessary blood volume to collect peripheral blood mononuclear cells (PBMCs) for the cell mediated immunity assays. Additional blood samples may be drawn at these time points for safety labs and serology assays.

Please refer to the Laboratory Manual in the Investigator Site File for instructions on drawing and processing blood samples, including the minimum volume of blood required and what to do in the case of failed attempts at drawing blood. Please see [section 6.8](#) for details on the cell mediated immunity assays and analysis of their data.

Samples will be retained in accordance with regulatory guidance for retention of essential study documents as described in [section 10](#).

3.5.5 Culture/PCR/Genotyping Assessments

Not applicable.

3.6 Stopping/Pausing Guidelines

Stopping/pausing guidelines are predefined criteria that halt the conduct of a study (either a vaccine group or the entire study). These guidelines are used to ensure the safety of study subjects. The following criteria, based on FDA guidance and with grading scales from the CBER Guidance for Industry “Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive vaccine Clinical Trials”, will be used for this study:

1. If one or more subjects experience a Grade 4 AE (see [Appendix D, E, and F](#)), vital sign or laboratory abnormality that cannot be clearly attributed to another cause, vaccination will be suspended until a full safety review is performed.
2. If six or more subjects experience a Grade 3 AE (see [Appendix D, E, and F](#)), vital sign or laboratory abnormality, dosage escalation will be suspended for that vaccine until a full safety review is performed.
3. The study will be halted (no new enrollments and no further investigational product administered until a full safety review and consultation with the health authorities completed) if one of the following occurs
 - a. One subject experiences a serious adverse event (SAE) assessed as possibly or probably related to investigational product or,
 - b. There is a subject death assessed as possibly or probably related to the investigational product.

If any of the halting rules are met, vaccination of subjects will be suspended until after full review of safety data by the DMC and health authorities. The investigator must inform the sponsor and the EC/IRB if any of the halting rules are met. The sponsor then must inform the DMC and health authorities. For further information on the DMC please see [section 6.9](#) and the DMC Charter.

The sponsor or the investigator (following consultation with the sponsor) has the right to discontinue the study at any time. If the clinical study is prematurely terminated, the investigator must promptly inform the study subjects and must assure appropriate therapy and follow-up for the subjects. All procedures and requirements pertaining to the archiving of the documents must be followed. All other study materials (such as study vaccines) must be returned to the sponsor.

3.7 Premature Withdrawal and Early Study Termination

A subject may discontinue study participation at any time prior to the last planned study visit. This is referred to as **premature withdrawal from the study** (see below for a description of withdrawal from study vaccine for subjects which refers to those subjects who do not receive additional vaccine doses but continue in the study for safety follow-up and/or other procedures). The reasons for premature withdrawal from the study include:

- Adverse event
- Death
- Withdrawal of consent
- Lost to follow-up
- Administrative reason
- Protocol deviation
- Other

NOTE: Before entering any alternate category as the reason for the subject's discontinuation from the study, the investigator should make every effort to investigate whether or not safety concerns (adverse event or death) may have been related to the subject's discontinuation from the study. If a safety concern has been associated with the subject's discontinuation, this must be described on the Termination CRF page, even if it is not the primary reason for the subject's discontinuation.

For any subject withdrawing from study participation prior to the planned Termination visit, it is important to determine if an AE was associated with the reason for discontinuing the study. This AE must be identified on the AE CRF page by indicating "Withdrawn from study due to AE".

For any subject withdrawn from study participation due to death, this should be noted on the Termination CRF page and the associated SAE that led to the death must be reported.

The subject can withdraw consent for participation in the study at any time without penalty or loss of benefit to which the subject is otherwise entitled. Reason for early termination should be deemed as “withdrawal of consent” if the subject withdraws from participation due to a non-medical reason (i.e., reason other than AE). If the subject intends to withdraw consent from the study, the investigator should clarify if the subject will withdraw completely from the study or if the subject will continue study participation for safety or a subset of other study procedures. If complete withdrawal from the study by the subject is specified, no further study interventions will be performed with the subject.

The date of termination is the date of the last contact (clinic visit or telephone) in which the subject’s health status was assessed or, in cases where the subject does not agree to any further safety follow-up; it is the date consent is withdrawn.

For subjects who fail to show up for scheduled visits (clinic or telephone contacts), study staff are encouraged to make at least three documented attempts to contact the subject by telephone and at least one documented written attempt to contact the subject and encourage the completion of study termination procedures. These efforts to contact the subject should be recorded in the source documents. The termination date for the subject to be captured on the Termination CRF page is the date of the last successful visit (clinic or telephone) with the subject.

For subjects who are withdrawn from the study due to sponsor decision (e.g., meeting pre-specified withdrawal criteria or termination of study by the sponsor), this reason should be noted in the Termination CRF page and any ongoing AEs at the time of study withdrawal must be followed until resolution/stabilization.

In general, subjects associated with protocol deviations may remain in the study unless continuation in the study jeopardizes the subject’s health, safety, or rights. For subjects who are withdrawn from the study due to receipt of an excluded medication/vaccination or due to significant protocol non-compliance, this reason should be noted in the Termination CRF page. Any ongoing AEs at the time of study withdrawal must be followed until resolution/stabilization. See below for greater detail.

If a subject is withdrawn prematurely from the study for a reason other than those outlined above, this reason must be documented in the Termination CRF page.

In studies that involve more than 1 consecutive dose of study vaccine, a separate event is “**withdrawal of study vaccination**”. This event may occur if subjects are expected to receive more than 1 consecutive dose of vaccine as part of study participation. The act of withholding additional study vaccinations is referred to as withdrawal of study vaccination. Subjects may be withdrawn from study vaccination for several reasons

including but not limited to: AE related to earlier vaccinations, failure to meet criteria for revaccination (see [section 4.4](#)), or pregnancy. **Subjects who are withdrawn from study vaccination should be encouraged to continue in the study for safety follow-up and other procedures as appropriate until the scheduled termination visit.** If the subject is withdrawn from study vaccination(s) due to adverse event, this event must be linked to the withdrawal from vaccination on the AE CRF page.

The sponsor or the investigator (following consultation with the sponsor) has the right to discontinue this study at any time. If the clinical study is prematurely terminated, the investigator is to promptly inform the study subjects and local EC/IRB and should assure appropriate therapy and follow up for the subjects. All procedures and requirements pertaining to the archiving of study documents should be followed. All other study materials (such as vaccines) must be returned to the sponsor.

Any subject who, despite the requirement for adequate contraception, becomes pregnant during the trial will not receive further vaccination but should be encouraged to continue participation in the study. The site should complete a Pregnancy Report CRF (initial report) as soon as possible (see [section 6.6.4](#) further details). If the subject withdraws from the study for any of the categories listed above except death, the site will obtain permission from the subject to continue to remain in contact with her until the outcome of the pregnancy is known, even if the outcome is not known until after the subject reaches the end of the routine study period.

Withdrawn subjects will not be replaced.

When a subject is withdrawn or withdraws from the study, the procedures described in [section 3.8](#) Early Termination Visit should be completed if possible.

3.8 Early Termination Visit

When a subject is withdrawn or withdraws from the study, the investigator will notify the sponsor and, when possible, will perform the procedures listed below:

- Collect the diary card (if not already collected).
- Review the subject's solicited and unsolicited safety data.
- Perform review of systems (see [section 3.2.5](#) for explanation) and review of concomitant medications/vaccinations since last visit.
- Collect vital sign measurements, including respiratory rate, blood pressure, pulse rate, and temperature (oral).

- Perform a directed physical examination including the respiratory system and any organ system warranting further examination based on reporting in the subject's diary and review of system.
- Draw a blood sample of approximately 10 mL for safety laboratory assessment.
- Obtain a urine sample for safety assessment.
- Perform a pregnancy test for female subjects of childbearing potential.

The data for the early termination visit should be recorded on a designated early termination visit eCRF.

4.0 SELECTION OF STUDY POPULATION

4.1 Inclusion Criteria

In order to participate in this study, all subjects must meet ALL of the inclusion criteria described.

1. Healthy males and non-pregnant females 18 to 45 years of age at time of enrollment.
2. Individuals who have given written consent after the nature of the study has been explained according to local regulatory requirements.
3. Individuals in good health as determined by the outcome of the medical history, physical examination and clinical judgment of the investigator.
4. Individuals who can comply with the study procedures and are available for follow up.

4.2 Exclusion Criteria

In order to participate in this study, all subjects must meet NONE of the exclusion criteria described.

1. Individuals with any severe chronic or acute disease.
2. Individuals with a history of illness or with an ongoing illness that may pose additional risk to the subject if he/she participates in the study, including the following:
 - History of any chronic respiratory illness, including current diagnosis of asthma within 2 years, exercise induced wheezing, reactive airway disease, emphysema, chronic bronchitis, cystic fibrosis or chronic obstructive pulmonary disease (COPD).
 - Any respiratory illness (e.g. cough, sore throat, dyspnea, wheezing or nocturnal awakenings to respiratory symptoms) within 7 days prior to receiving the first study injection.
 - Any active pulmonary infection or other inflammatory conditions, even in the absence of febrile episodes, within 14 days prior to the first study injection.
 - Hepatitis B or hepatitis C infection.
3. Individuals who have had a malignancy (excluding nonmelanotic skin cancer) or lymphoproliferative disorder within the past 5 years.

4. Individuals with known or suspected impairment of the immune system including but not limited to HIV, autoimmune disorders, immunosuppressive therapy, and diabetes mellitus.
5. Individuals with any history of progressive or severe neurologic disorder, seizure disorder or Guillian-Barré syndrome.
6. Individuals with a known bleeding diathesis, or any condition that may be associated with a prolonged bleeding time.
7. Individuals with a BMI > 35 kg/m². BMI is to be calculated by the following formula: subject weight at baseline divided by subject height in meters multiplied by the subject height in meters. The numerical result will be rounded to the nearest 0.1.
8. Individuals who are allergic to any of the vaccine components, or with a history of anaphylaxis after vaccination.
9. Individuals who during the 90 days prior to enrollment receive any medications or other treatments that may adversely affect the immune system such as allergy injections, immune globulin, interferon, immunomodulators, cytotoxic drugs or other drugs known to be frequently associated with significant major organ toxicity.
10. Individuals who receive systemic immunosuppressive agents including steroids. Prior corticosteroid therapy should be discontinued 28 days prior to enrollment. Individuals using inhaled or topical corticosteroids will be permitted.
11. Receipt or donation of blood or blood products 8 weeks prior to vaccination or planned receipt or donation during the study period.
12. Individuals participating in any clinical trial with another investigational product 28 days prior to receiving the first study vaccination or intent to participate in another clinical study at any time during the conduct of this study.
13. Individuals who have received any vaccine 28 days prior to enrollment in this study, or who plan to receive any non-study vaccines within 28 days of the second dose of study vaccine.
14. Individuals with any abnormal safety laboratory result at the screening visit.
15. If female, 'of childbearing potential', sexually active and has not used any of the 'acceptable contraceptive methods' for at least two months prior to study entry.

Childbearing potential is defined as status post onset of menarche and not meeting any of the following conditions: menopausal for at least two years; sterile status after bilateral tubal ligation for at least one year, immediately after bilateral oophorectomy or after hysterectomy.

Acceptable methods of birth control are defined as one or more of the following:

- Hormonal contraceptives (such as oral, injection, transdermal patch, implant, cervical ring).
 - Barrier (condom with spermicide or diaphragm with spermicide) each and every time during intercourse.
 - Intrauterine device (IUD).
 - Monogamous relationship with vasectomized partner. Partner must have been vasectomized for at least six months prior to subject's study entry.
16. If female subject of childbearing potential and have a positive urine pregnancy test prior to study vaccinations, or are currently lactating.
 17. If female of childbearing potential and sexually active, refusal to use an 'acceptable contraceptive method' through to three weeks after last study vaccination.
 18. Individuals with behavioral or cognitive impairment or psychiatric disease that, in the opinion of the investigator, may interfere with the subject's ability to participate in the study.
 19. Individuals with a history of drug or alcohol abuse within the past 2 years.
 20. Individuals who are acting as study personnel or immediate family members (brother, sister, child, parent) or the spouse of study personnel.
 21. Individuals with a body temperature ≥ 38 °C (≥ 100.4 °F) within 3 days of intended study vaccination.
 22. Individuals with any condition that, in the opinion of the investigator, would interfere with the primary study objectives.

There may be instances when individuals meet all entry criteria except one that relates to transient clinical circumstances (e.g., body temperature elevation or recent use of excluded medication or vaccine). Under these circumstances, a subject may be considered eligible for study enrollment if the appropriate window for delay has passed, inclusion/exclusion criteria have been rechecked, and if the subject is confirmed to be eligible.

4.3 Criteria for Delay of Vaccination and/or Blood Sampling

After enrollment, subjects may encounter clinical circumstances that warrant a delay in subsequent study vaccination. These situations are listed below. In the event that a subject meets a criterion for delay of vaccination, the subject may receive study vaccination once the window for delay has passed as long as the subject is otherwise eligible for study participation.

- Individuals with a body temperature $>38.0^{\circ}\text{C}$ ($>100.4^{\circ}\text{F}$) within 3 days of intended study vaccination.

There are also circumstances under which repeat vaccination is a contraindication in this study. These circumstances include anaphylaxis or severe hypersensitivity reactions following vaccination. If these reactions occur, the subject must not receive additional vaccinations, but should be encouraged to continue in study participation.

4.4 Criteria for Repeat Vaccination in the Study

Prior to receipt of second study vaccination, subjects must be evaluated to confirm that they are eligible for subsequent vaccination. If subjects meet any of the original exclusion criteria or the criteria listed below, they should not receive additional vaccinations.

- Subjects who experience any serious adverse event judged to be possibly or probably related to study vaccine or non-study vaccines, including hypersensitivity reactions.
- Subjects who develop any new condition which, in the opinion of the investigator, may pose additional risk to the subject if he/she continues to participate in the study.

Subjects who meet any of these criteria must not receive further study vaccinations. However, these subjects should be encouraged to continue study participation, as discussed in [section 3.7](#).

5.0 TREATMENT OF SUBJECTS

All vaccines associated with this study are to be stored separately from other vaccines and medications in a secure location under appropriate storage conditions with temperature monitoring. **All vaccines associated with this study must be checked for expiration date prior to use. Expired vaccines must not be administered to subjects.**

5.1 Study Vaccine(s)

The term 'study vaccine' refers to those vaccines provided by the Sponsor, which will be evaluated as part of the study objectives. The study vaccines specific to this study are described below.

Three dosage levels of the RSV F subunit vaccine will be tested in this study, including formulations with and without adjuvant (see [Table 5.1](#)). Subjects will be randomized into three different cohorts, which will be assigned to receive one of the following dosage levels:

1. Cohort 1: 45 µg
2. Cohort 2: 90 µg
3. Cohort 3: 135 µg

Within each cohort subjects will be further randomized to receive the RSV F subunit vaccine without adjuvant, with 1 mg aluminum hydroxide, or with MF59 (which contains 9.75 mg squalene and surfactants in 0.25 mL). Additionally, approximately 24 subjects in each cohort will be randomized to receive placebo (saline).

Details of vaccine composition in each cohort are summarized in [Table 5.1](#).

Table 5.1 Study Vaccine Composition

Cohort	Vaccine Group	Antigen Name (Dosage)	Volume (Route of Administration)	Adjuvant Name (Dosage)	Excipients
1	A	RSV F subunit 45 µg	0.50 mL (IM)	None	Sucrose, Potassium phosphate
	B	RSV F subunit 45 µg	0.50 mL (IM)	Aluminum hydroxide (1 mg)	
	C	RSV F subunit 45 µg	0.50 mL (IM)	MF59 (9.75 mg squalene)	
2	D	RSV F subunit 90 µg	0.50 mL (IM)	None	
	E	RSV F subunit 90 µg	0.50 mL (IM)	Aluminum hydroxide (1 mg)	
	F	RSV F subunit 90 µg	0.50 mL (IM)	MF59 (9.75 mg squalene)	
3	G	RSV F subunit 135 µg	0.50 mL (IM)	None	
	H	RSV F subunit 135 µg	0.50 mL (IM)	Aluminum hydroxide (1 mg)	
	I	RSV F subunit 135 µg	0.50 mL (IM)	MF59 (9.75 mg squalene)	
1-3	Placebo	Sterile saline 0.9%	0.50 mL (IM)	none	

5.2 Non-Study Vaccines

The term ‘non-study vaccine’ refers to those vaccines which will be intentionally given to study subjects but not formally included in the analysis of study objectives. No non-study vaccines are planned for this trial. Non-study vaccines which are given to subjects during the trial will be recorded in the Prior and Concomitant Medications/Blood Products eCRF.

5.3 Vaccines Preparation and Administration

Detailed descriptions for vaccine handling and preparation will be placed in the Investigator Site File and the investigator must review these materials prior to study start.

The investigator or designee will be responsible for oversight of the administration of vaccine to subjects enrolled in the study according to the procedures stipulated in this study protocol. All vaccines will be administered only by unblinded personnel who are qualified to perform that function under applicable local laws and regulations for the specific study site.

The RSV subunit vaccine will be provided as a lyophilized pellet at the 135 µg dosage. The lyophilized product is reconstituted with (a) sterile saline 0.9% NaCl to generate the non-adjuvanted RSV F subunit vaccine formulation, (b) aluminum hydroxide [Al(OH)₃] to generate the RSV F subunit vaccine formulated in Al(OH)₃, or (c) sterile saline 0.9% NaCl and adjuvant MF59 to generate the RSV F subunit vaccine formulated with MF59. In addition, the vaccine will need to be reconstituted in saline and diluted to the proper dosage for Cohort 1 and Cohort 2, and mixed with adjuvant as needed for each treatment group (for example, diluted and mixed with aluminum hydroxide for vaccine group E in [Table 5.1](#)), while also producing the correct final volume of 0.5 mL. Complete reconstitution and mixing instructions can be found in the Investigator Site File. The vaccine will be injected intramuscularly as a 0.5 mL dose using a standard syringe. A new syringe will be used for each subject.

PRECAUTIONS TO BE OBSERVED IN ADMINISTERING STUDY VACCINE:

Prior to vaccination, subjects must be determined to be eligible for study vaccination and it must be clinically appropriate in the judgment of the investigator to vaccinate. Eligibility for vaccination prior to first study vaccine administration is determined by evaluating the entry criteria outlined in protocol [section 4.1](#) through [4.2](#).

Eligibility for subsequent study vaccination is determined by following the criteria outlined in [sections 4.3](#) and [4.4](#).

Study vaccines should not be administered to individuals with known hypersensitivity to any component of the vaccines.

Standard immunization practices are to be observed and care should be taken to administer the injection intramuscularly. Before administering vaccine, the vaccination site is to be disinfected with a skin disinfectant (e.g., 70% alcohol). Allow the skin to dry. **DO NOT inject intravascularly.**

As with all injectable vaccines, trained medical personnel and appropriate medical treatment should be readily available in case of anaphylactic reactions following vaccine administration. For example, epinephrine 1:1000, diphenhydramine, and/or other medications for treating anaphylaxis should be available.

5.4 Prior and Concomitant Medications and Vaccines

All medications, vaccines and blood products taken or received by the subject within 28 days prior to the start of the study are to be recorded on the Prior and Concomitant Medications/Blood Products eCRF. The use of antipyretics and/or analgesic medications within 24 hours prior to vaccination must be identified and the reason for their use

(prophylaxis versus treatment) must be described in the source documents and/or diary card and Concomitant Medications eCRF.

Medications taken for prophylaxis are those intended to prevent the onset of symptoms. Medications taken for treatment are intended to reduce or eliminate the presence of symptoms that are present.

In addition, the following are considered prior medications for this protocol: all medication/vaccines described in the inclusion and exclusion criteria of this protocol including those that may adversely affect the immune system.

Concomitant medications include all medications (including vaccines) taken by/administered to the subject at and after enrollment and must be documented on the Concomitant Medications/Blood Products eCRF.

When recording concomitant medications/vaccines, they should be checked against the study entry and continuation criteria in [sections 4.1](#) through [4.4](#) to ensure that the subject should be enrolled/continue in the study.

5.5 Vaccine Supply, Labeling, Storage, and Tracking

The sponsor will ensure the following:

- supply the study vaccine
- appropriate labeling of all study vaccines provided that complies with the legal requirements of each country where the study is to be performed

The investigator must ensure the following:

- acknowledge receipt of the study vaccines by a designated staff member at the site, including confirmation that the vaccines:
 - were received in good condition
 - remained within the appropriate temperature range during shipment from the sponsor to the investigator's designated storage location
 - have been confirmed by the sponsor as authorized for use
- proper storage of the study vaccines, including:
 - storage in a secure, locked, temperature-controlled location
 - proper storage according to the instructions specified on the labels

- appropriate record keeping and inventory of the study vaccines, including regular documentation of adequate storage temperature
- appropriate use of the study vaccines, including:
 - use only in accordance with the approved protocol
 - proper handling, including confirmation that the vaccine has not expired prior to administration
 - appropriate documentation of administration of vaccines to study subjects including:
 - date, dosage, batch/serial numbers, expiration dates, unique identifying numbers assigned to subjects and study vaccines, and time of vaccine administration. This information will be maintained in an accountability log that will be reviewed by the site monitor.
 - proper reconciliation of all study and non-study vaccines received from the sponsor. Reconciliation is defined as maintaining records of which and how many vaccines were received, which vaccines (and volume thereof) were administered to subjects, which vaccines were destroyed at the site, and which vaccines were returned to the sponsor, as applicable.
- proper adherence to the local institutional policy with respect to destruction of study vaccines.
- complete record keeping of vaccine use, wastage, return or destruction, including documentation of:
 - copy of the site's procedure for destruction of hazardous material
 - number of doses destroyed, date of destruction, destruction code (if available), method of destruction, and name of individual performing destruction

Vaccines that have been stored differently from the sponsor's instructions **must not** be used unless the sponsor provides written authorization for use. In the event that the use cannot be authorized, the sponsor will make every effort to replace the vaccine supply. All vaccines used in conjunction with this protocol must be stored separately from normal hospital/practice stocks to prevent unintentional use of study vaccines outside of the clinical trial setting.

Monitoring of vaccine accountability will be performed by the study monitor during site visits and at the completion of the trial as applicable dependent on the use of a computerized randomization system.

At the conclusion of the study, and as appropriate during the course of the study, the investigator must return all unused study vaccines, packaging and supplementary labels to the sponsor.

6.0 MEASUREMENTS

6.1 Appropriateness of Measurements

The measures of immunogenicity used in this study have been described based on scientific consensus and have been deemed appropriate to describe the immune response against RSV in this study. Exploratory immunogenicity assays will further understanding of the immune response to RSV and RSV vaccines.

The measures of safety used in this study are based on previous study data and on comparable routine clinical and laboratory procedures. They include a close vigilance for, and stringent reporting of selected local and systemic adverse events routinely monitored in vaccine trials as indicators of reactogenicity. In addition, selected laboratory tests will be performed pre- and post-vaccination.

6.2 Demographics, Medical History and Physical Examination

Prior to study enrollment, demographic data will be collected from the subject, including: gender, race, ethnicity, age, height, weight, and body mass index.

Medical history will also be collected, including but not limited to any medical history that may be relevant to subject eligibility for study participation such as prior vaccinations, concomitant medications, and previous and ongoing illnesses or injuries. Relevant medical history can also include any medical history that contributes to the understanding of an adverse event that occurs during study participation, if it represents an exacerbation of an underlying disease/preexisting problem.

A general physical examination is to be performed by a qualified health care practitioner and will include, at a minimum, measurement of the following vital signs: respiratory rate, heart rate, blood pressure, and body temperature (oral). It will also include assessment of general appearance; examination of the head, ears, eyes, nose and throat (HEENT); examination of skin; auscultation of heart and lungs; palpation of the abdomen; and examination of extremities. "Qualified health care practitioner" refers to any licensed health care professional who is permitted by institutional policy to perform physical examinations and who is identified within the site's roles and responsibilities log.

At clinic visits after enrollment subjects will undergo a symptom-directed physical examination. This is a physical examination that will include at a minimum measurement of vital signs (respiratory rate, heart rate, and blood pressure), body temperature (oral), a check of general appearance, as well as examination of organ systems that are relevant to

the investigator based on review of the subject's reported adverse events, review of systems, or concomitant medication use.

Data on subject demographics, medical history, and physical exams will be recorded in the source documents and in the associated eCRFs as included in the eCRF instructions provided by NVD.

6.3 Immunogenicity Measurements

Serum anti-RSV neutralizing antibody titers will be evaluated for all subjects by plaque reduction neutralization assay (PRNA). Samples will be collected at the following time points:

- Visit 1 (baseline, pre-vaccination)
- Visit 5 (28 days after the first dose)
- Visit 9 (28 days after the second dose)
- Visit 12 (6 months after the first dose)

Binding antibody to the RSV proteins F, G (G_a and G_b), and N by a microsphere-based fluorescence linked immunosorbent assay, multiplexed using Luminex® technology will be evaluated using the same samples.

The binding assays are needed to (i) distinguish immune responses following immunization versus natural infection, and (ii) assess the ratio of neutralizing antibodies (NAb) and non-NAb to the F protein.

All testing will be conducted by qualified and certified laboratories. See [section 3.5.1](#) and the Laboratory Manual in the Investigator Site File for more information. For assays not performed internally by NVD, data will be provided by external laboratories by EDT to NVD (see [section 9](#)).

6.4 Efficacy Measurements

This study has no efficacy measurements.

6.5 Solicited Safety Measurements

The term “reactogenicity” refers to selected signs and symptoms (“adverse events”) occurring in the hours and days following a vaccination, to be collected by the subject for 7 consecutive days, using a pre-defined checklist in a diary card (i.e., solicited adverse events).

The following adverse events are included in the diary check list. Each adverse event is to be assessed using the scoring system shown in [Appendix B](#) (local AEs) and [Appendix C](#) (systemic AEs).

Solicited local adverse events:

Solicited local AEs include:

- injection site induration
- injection site swelling
- injection site erythema
- injection site pain

AEs will be recorded as absent, mild, moderate, or severe. Please see [Appendix B](#) for grading scales.

Solicited systemic adverse events:

Solicited systemic AEs include:

- body temperature (oral preferred)
- chills
- nausea
- generalized myalgia
- generalized arthralgia
- headache
- fatigue
- diarrhea
- loss of appetite
- cough
- rhinorrhea
- wheezing

AEs will be recorded as absent, mild, moderate, or severe. Please see [Appendix C](#) for grading scales.

Other solicited reactions:

Use of analgesics/antipyretics will be recorded as absent or present, and it will be indicated if they were used for treatment or for prophylaxis.

Solicited local and systemic AEs and unsolicited AEs will be collected using diary cards for 7 days following any dose of vaccine or placebo. Subjects will be asked to take an oral temperature for the first 7 days after each dose and to record other systemic and local AEs that may develop, as well as any medications subjects have received. Reminder telephone calls at two and four days after each dose of vaccine or placebo will be used to remind the subject to record AEs in the diary cards and bring the diary cards to their next clinic visit. Telephone calls at 14 and 21 days after each dose of vaccine or placebo will be used to collect unsolicited AEs occurring since the last study contact, as well as any solicited AEs continuing beyond the 7 day period after vaccination.

All AEs necessitating a non-scheduled physician visit, medical attention, or leading to withdrawal from the study will also be collected throughout the study, and all AEs will be monitored until resolution or, if the AE becomes chronic, a cause is identified. If an AE is unresolved at the conclusion of the study, a clinical assessment will be made by the investigator and the medical monitor to determine whether or not continued follow-up of the AE is warranted. The relationship of the study treatment to any AE and any SAE will be determined by the investigator as probably related, possibly related, or not related. Any SAEs will further be reported as related/suspected or not related. All AEs resulting in withdrawal of the subjects from the study will be summarized.

In addition, all SAEs, NOCDs, and AESIs will be collected from the date of signed informed consent and at scheduled periodic reviews and will be followed up until the end of the study (approximately 393 days after the first dose of vaccine or placebo [saline]). This may include a period before and after an active treatment of investigational product. On an individual basis, SAE follow-up may take longer than the study duration, if the SAE is not resolved at the completion of the study. Monitoring for AESIs (see [section 6.6.1.1](#) and [Appendix A](#)) will be extended through 12 months after the second vaccination and will be accomplished by clinic visits or telephone follow-up calls as detailed in [Table 2](#). All AESIs will be reported in the same manner as SAEs. A tabulation of all AESIs, categorized by MedDRA preferred terms and assessed relationship to study vaccine will be performed.

The study staff must review the diary card with the subject at the next clinic visit (see [section 3.2.5](#)) and must directly record the solicited local and systemic adverse events, and other solicited reactions on the appropriate Local and Systemic Reactions eCRF. As described in [Section 3.4.1](#), all solicited adverse events that are legible must be recorded verbatim in the eCRFs, even if the values do not appear to be plausible (see [section 3.2.5.3](#) and [section 8.1](#) for more detail).

If a solicited local or systemic adverse event continues beyond day 7 after vaccination, it will also be recorded as an Adverse Event on the Adverse Events eCRF.

6.6 Unsolicited Safety Measurements

6.6.1 Adverse Events

An adverse event (AE) is defined as any untoward medical occurrence in a subject or clinical investigation subject administered a pharmaceutical product at any dose that does not necessarily have to have a causal relationship with this treatment. Therefore, an AE can be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of an investigational product, whether or not considered related to the investigational product. This definition includes intercurrent illnesses or injuries and exacerbation of pre-existing conditions.

NOTE: Every effort should be made by the investigator to evaluate new safety information reported by a subject (solicited and unsolicited AEs) for an underlying diagnosis and to capture this diagnosis as the event in the AE page. In other words, the practice of reporting only symptoms (e.g., “cough” or “ear pain”) are better reported according to the underlying cause (e.g., “asthma exacerbation” or “otitis media”).

All AEs will be monitored until resolution or, if the AE becomes chronic, a cause identified. If an AE is unresolved at the conclusion of the study, a clinical assessment will be made by the investigator and medical monitor whether continued follow-up of the AE is warranted.

The severity of events reported on the Adverse Events eCRF will be determined by the investigator as:

Mild: transient with no limitation in normal daily activity.
Moderate: some limitation in normal daily activity.
Severe: unable to perform normal daily activity.

The relationship of the study treatment to an AE will be determined by the investigator based on the following definitions:

1. Not Related

The AE is not related to an investigational vaccine if there is evidence that clearly indicates an alternative explanation. If the subject has not received the vaccine, the timing of the exposure to the vaccine and the onset of the AE are not reasonably related in

time, or other facts, evidence or arguments exist that reasonably suggest an alternative explanation, then the AE is not related.

2. Possibly Related

The administration of the investigational vaccine and AE are considered reasonably related in time and the AE could be explained by exposure to the investigational vaccine or by other causes.

3. Probably Related

Exposure to the investigational vaccine and AE are reasonably related in time and no alternative explanation has been identified.

The relationship of the study treatment to an unsolicited AE will be determined by the investigator. Solicited AEs will not be evaluated for relationship to study vaccine and severity of solicited AEs is defined as described in [section 6.5](#).

Adverse events will also be evaluated by the investigator for the co-existence of any of the other following conditions:

- “Medically attended adverse event”: an adverse event that leads to an unscheduled visit to a healthcare practitioner.
- “New onset of chronic disease”: an adverse event that represents a new diagnosis of a chronic medical condition that was not present or suspected in a subject prior to study enrollment.
- “New onset of autoimmune disorder”: an adverse event that represents a new diagnosis of an autoimmune disease that was not present or suspected in a subject prior to study enrollment.

Please note: any solicited adverse event that meets any of the following criteria must also be entered as an adverse event on the Adverse Event eCRF:

- Solicited local or systemic adverse event leading to the subject withdrawing from the study or the subject being withdrawn from the study by the investigator.
- Solicited local or systemic adverse event lasting beyond 7 days’ duration.
- Solicited local or systemic adverse events that lead to subject withdrawal from study vaccination.
- Solicited local or systemic adverse event that otherwise meets the definition of a serious adverse event (see [section 6.6.2](#)).

6.6.1.1 Adverse Events of Special Interest

Certain adverse events of special interest (AESIs) are monitored for after the administration of immunostimulatory agents. All subjects enrolled in the study will be monitored for AESIs for the entire follow-up period. The AESIs will be defined according to the MedDRA preferred terms listed in [Appendix A](#). The occurrence of any of these adverse events will be treated as a serious adverse event (SAE), meeting the criterion of a “medically important event.”

Subjects will be assessed for diagnosis of an AESI at the following clinic visits:

- Visit 1 (baseline, pre-vaccination)
- Visit 2 (7 days after the first dose)
- Visit 5 (28 days after the first dose; pre-second dose)
- Visit 6 (7 days after the second dose)
- Visit 9 (28 days after the second dose)
- Visit 10 (56 days after the second dose)
- Visit 11 (4 months after the first dose)
- Visit 12 (6 months after the first dose)
- Visit 13 (6 months after the second dose)

At these visits a qualified health care practitioner listed on the site’s responsibilities and delegation logs will conduct a review of organ systems and a targeted physical exam, and will evaluate subjects for any new signs or symptoms that could indicate one of the AESIs listed in [Appendix A](#). They will also interview the subject about recent medical history and any new diagnosis that could indicate an AESI. Medical records related to any new medical event or diagnosis will be requested, reviewed by the site staff, and recorded in the subject’s source documents.

In addition, subjects will be interviewed by phone at the following time points:

- Visit 3 (14 days after the first dose)
- Visit 4 (21 days after the first dose)
- Visit 7 (14 days after the second dose)
- Visit 8 (21 days after the second dose)
- Visit 14 (9 months after the first dose)
- Visit 15 (1 year after the first dose)

- Visit 16 (1 year after the second dose)

These phone calls will include a scripted interview to determine if the subject has had a new medical issue for which they visited a doctor, and if they had any new diagnosis since the last clinic visit. Any new diagnosis deemed to potentially be an AESI will be recorded in the subject's source documents, and the subject will be instructed to visit the site within the next 7 days. The subject will be instructed to bring any new medical records with them to the clinic for review.

A diagnosis of an AESI will be reported in the same manner and time frame as an SAE using the VSAE form. The investigator must notify NVD within 24 hours. The AESI diagnosis, as well as any medications taken to treat the condition, will be recorded in the subject's source documents and on the Adverse Events eCRF.

6.6.1.2 New Onset of Chronic Disease

New onset of chronic disease is defined as an illness that starts during the course of the study that did not exist prior to enrollment into the study and is likely to persist throughout the lifetime of the subject. A chronic disease is one that can be treated but for which no cure exists. For example, a new onset of asthma (occurs for the first time during the study and has been evaluated longitudinally to ensure that a diagnosis of asthma is appropriate) is classified as a New Onset of Chronic Disease (NOCD). This is applicable for any chronic disease affecting any organ class that arises during the study that is likely to persist throughout the subject's lifetime (even if in remission). Infectious diseases, such as Hepatitis C virus or tuberculosis, are not considered NOCDs as there are antibiotics and antivirals that are potentially able to cure such diseases (this is a generalizable cure and not specific to the subject). In case of doubt, diagnoses of new onset of chronic disease should be discussed with the study medical monitor.

Any NOCD will be described in the medical chart/source documents and entered into the Adverse Events eCRF.

6.6.2 Serious Adverse Events

A serious adverse event (SAE) is defined as any untoward medical occurrence that at any dose results in one or more of the following:

- Death.
- Is life-threatening (i.e., the subject was, in the opinion of the investigator, at immediate risk of death from the event as it occurred); it does not refer to an event which hypothetically might have caused death if it were more severe.
- Required or prolonged hospitalization.

- Persistent or significant disability/incapacity (i.e., the event causes a substantial disruption of a person's ability to conduct normal life functions).
- Congenital anomaly or birth defect.
- An important and significant medical event that may not be immediately life threatening or resulting in death or hospitalization but, based upon appropriate medical judgment, may jeopardize the subject or may require intervention to prevent one of the other outcomes listed above.

Adverse events which do not fall into these categories are defined as **non-serious**.

It should be noted that a severe adverse event need not be serious in nature and that a serious adverse event need not, by definition, be severe.

Serious adverse events will be captured both on the VSAE form as well as on the AE eCRF. All SAEs will be evaluated by the investigator for relationship of the event to study vaccine. SAEs that are judged to be possibly or probably related to the study vaccine should be reported to the sponsor as related (i.e., suspected) events.

The relationship of the study treatment to an SAE will be determined by the investigator based on the following definitions:

1. Related/suspected

The SAE is judged by the investigator to be possibly or probably related to the study vaccine on the AE CRF page (see [section 6.6.1](#)).

2. Not Related

The SAE is not related if exposure to the study vaccine has not occurred, **or** the occurrence of the SAE is not reasonably related in time, **or** the SAE is considered unlikely to be related to use of the study vaccine, i.e., there are no facts (evidence) or arguments to suggest a causal relationship.

The relationship of the study vaccine to an SAE will be determined by the investigator.

In addition, SAEs will be evaluated by the sponsor or designee for "expectedness." An unexpected AE is one that is not listed in the Investigator's Brochure or an event that is by nature more specific or more severe than a listed event.

In addition, a pre-existing event or condition that results in hospitalization should be recorded on the Medical History eCRF. If the onset of an event occurred before the subject entered the trial (e.g., any pre-planned hospitalization for conditions like cosmetic

treatments or for non-emergency routine visits for a pre-existing condition), the hospitalization would not lead to an AE being classified as serious unless, in the view of the investigator, hospitalization was prolonged as a result of participation in the clinical trial or was necessary due to a worsening of the pre-existing condition.

6.6.3 Methods for Assessing and Recording AEs and SAEs

The period of observation for AEs extends from the time the subject signs informed consent until he or she completes the specified safety follow-up period of approximately 1 year after receipt of the second dose of vaccine or terminates the study early (whichever comes first). AEs occurring after the informed consent form is signed but prior to receiving study vaccine/product will be documented as an adverse event and recorded on the Adverse Events eCRF and on source documents. However, AEs occurring prior to receipt of any study vaccine will be analyzed separately from “treatment emergent” AEs (AEs occurring after administration of the first study vaccine).

All AEs meeting criteria for reporting, regardless of severity, will be monitored by the investigator until resolution or stabilization. All subjects experiencing AEs - whether considered associated with the use of the study vaccine or not - must be monitored until symptoms subside and any abnormal laboratory values have returned to baseline, or until there is a satisfactory explanation for the changes observed, or until death, in which case a full pathologist’s report should be supplied, if possible. All findings must be reported on an Adverse Events CRF and on the VSAE form, if necessary, which is part of the Investigator Site File. All findings in subjects experiencing AEs must be reported also in the subject's medical records.

All SAEs which occur during the course of the trial, whether considered to be associated with the study vaccination or not, must be reported **within 24 hours of the site becoming aware of the event** by telephone or fax to NVD. Contact details for submitting SAEs to NVD or its designee and instructions for completion of documentation will be provided in a handout located in the Investigator Site File.

All SAEs are also to be documented on the Adverse Events eCRF. Any medication or other therapeutic measures used to treat the AE will be recorded on the appropriate eCRF(s) in addition to the outcome of the AE.

After receipt of the initial report, representatives of NVD will contact the investigator if it is necessary to obtain further information for assessment of the event.

All SAEs must be reported by the investigator to his/her corresponding EC or IRB and applicable regulatory authorities in accordance with institutional policy/regulatory

requirements and adequate documentation of this notification must be provided to the sponsor.

NVD or its designee must also comply with the applicable regulatory requirement(s) related to the reporting of unexpected serious and non-serious adverse vaccine reactions (also referred to as “SUSARs”) to the regulatory authority(ies) and the IRB/EC. If a SUSAR or other safety signal relating to use of one of the study vaccines is reported to NVD or its designee, the sponsor will communicate the information to the investigator and the investigator will be responsible for submitting this information to the EC or IRB and other relevant authorities.

Post-Study Events

Any suspected SAE that occurs outside of the protocol-specified follow-up period or after the end of the study but considered to be caused by the study vaccine must be reported to Novartis or its designee. These SAEs will be processed by Novartis or its designee as during the course of the study, until 3 months after the last scheduled study visit. Instructions and contact details for collecting and reporting these suspected SAEs will be provided to the investigator.

6.6.4 Pregnancies

To ensure subjects’ safety, each pregnancy in a subject on study vaccine must be reported to NVD within 24 hours of the site learning of its occurrence. If the subject agrees to submit this information, the pregnancy must be followed to determine outcome, including spontaneous or voluntary termination, details of the birth, and the presence or absence of any birth defects, congenital abnormalities, or maternal and/or newborn complications. This follow-up should occur even if intended duration of safety follow-up for the study has ended.

Pregnancy data must be recorded on a Pregnancy Report CRF (initial report) and Pregnancy Follow-Up CRF (outcome report) and reported to NVD. Contact details for submitting the case report forms will be described in the Investigator Site File.

Any pregnancy outcome meeting the definition of a SAE (see [section 6.6.2](#)) must also be reported on the VSAE Report Form.

6.7 Safety Laboratory Measurements

To assess laboratory AEs, blood and urine samples will be collected for each subject at Screening and Visits 1, 2, 5, 6, and 9 as outlined in [Table 2](#) and in [section 3.5.3](#) to perform blood chemistry, hematology, and urine analyses for all subjects.

Safety laboratory assessments from serum will include sodium, potassium, chloride, bicarbonate (or CO₂), glucose, blood urea nitrogen (BUN), calcium, magnesium, phosphorous, creatinine, creatine phosphokinase (CPK), albumin, total protein, alkaline phosphatase, liver function tests (ALT, AST, γGT), total bilirubin, direct bilirubin, indirect bilirubin, cholesterol, LDH, and pancreatic enzymes (amylase, lipase). Hematology tests will include CBC test (WBC, WBC differential, RBC, RBC indices, hemoglobin, hematocrit, platelets, and mean platelet volume [MPV]), fibrinogen, prothrombin time (PT), partial thromboplastin time (PPT), INR, C-reactive protein (CRP), D-dimer.

Safety laboratory assessments from urine will include at a minimum protein, glucose, and presence of red blood cells.

All testing will be conducted by qualified and certified laboratories as outlined in the Laboratory Manual. Abnormal laboratory values will be defined following the FDA CBER Guidance for Industry “Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive vaccine Clinical Trials” dated September 2007, or the institution’s normal ranges if they differ from CBER guidance. Please [see Appendix D, E, and F](#). Retesting will not be performed in the case of abnormal values.

6.8 Other Measurements

The serum neutralizing antibody (NAb) and binding antibody responses against additional RSV strains and native or engineered RSV antigens at baseline and following vaccination will be performed in a subset of subjects, following evaluation of the primary and secondary immunogenicity evaluations and depending on the volume of serum remaining for additional testing. The predominant isotype of the RSV-specific serum antibody may be determined and the RSV-specific immune response against different RSV subgroup A and subgroup B strains may be evaluated. RSV group A and group B strains could include those with engineered alterations in the F and G glycoproteins and strains that lack the G glycoprotein. Different RSV antigens may include engineered, recombinant RSV antigens, such as an F antigen in which one or more known neutralizing epitopes has been blocked by the addition of a glycosylation site or F antigens locked in a defined conformation, such as a pre-fusion conformation. To make this determination, the serum

samples may be mixed with a recombinant RSV antigen (potentially an antigen with engineered changes) before use in a neutralization assay. Alternatively, the serum may be bound to an immobilized recombinant RSV antigen and then eluted before use in a neutralization assay or binding assay, or the fraction of serum that does not bind to an immobilized recombinant RSV antigen may be used in a neutralization assay or binding assay.

These data will be entered into Source Documents if the assays are conducted internally at NVD, or will be transferred via EDT if the assays are conducted by external laboratories (see [section 9.1](#)). Testing and analyses to characterize the serum neutralizing antibody (NAb) to additional RSV strains may be performed after the Clinical Study Report (CSR) has been completed, in which case these data will be submitted as an addendum to the CSR.

The serum NAb and binding Ab assays described above will be performed in approximately the first 10 subjects enrolled in each of the four treatment groups of cohort 3 (135 µg of RSV F subunit vaccine without adjuvant, with MF59, or with aluminum hydroxide; or placebo), who agree to a separate Informed Consent and additional collection of blood samples. The time points for blood collection will be:

- Visit 1 (baseline, pre-vaccination)
- Visit 2 (seven days after the first dose)
- Visit 5 (28 days after the first dose)
- Visit 12 (six months after the first dose)

In addition, cell mediated immunity assays will be performed. The frequency of B cells specific for RSV proteins will be determined by enzyme-linked immunosorbent spot (ELISPOT) at the same time points, in order to evaluate the peak of plasmablast responses (Visit 2), the B cell memory responses (Visit 5), and for persistence of antibody responses (Visit 12).

Subsequently, the analysis of the RSV-specific B-cell repertoire to explore the baseline immunity to RSV and the immune response to the RSV F subunit vaccine will be performed in a subset of these subjects, including those with the most pronounced response to the RSV F subunit vaccine in the non-adjuvanted, aluminum hydroxide-adjuvanted, and MF59-adjuvanted treatment groups. The samples for the exploratory B cell repertoire analysis will be chosen based on the levels of baseline immunity and the level of response to the vaccine (as determined serologically and by ELISPOT), and on the distribution of samples between subjects who received placebo, unadjuvanted vaccine,

aluminum hydroxide-adjuvanted vaccine, or MF59-adjuvanted vaccine. The selection will be based on the goal of the exploratory objective to understand the differences in the RSV-specific B cell repertoires elicited by natural infection and by the adjuvanted or unadjuvanted vaccine. Testing and analyses of the RSV-specific B-cell repertoire may be or may not be performed after the Clinical Study Report (CSR) has been completed, and these data will be submitted as an addendum to the CSR.

B cells secreting RSV-specific antibodies will be enumerated by ELISPOT according to treatment group and time point evaluated for subjects in cohort 3 only. The diversity of the elicited B-cell receptors will be accomplished through sequence analysis of complementary DNA (cDNA) from immunoglobulin (Ig) messenger RNA (mRNA) and functional, binding, and epitope mapping analyses of antibodies or antibody fragments expressed from those cDNAs. The Ig cDNAs will be cloned from plasmablasts obtained approximately one week after dosing and from memory B cells obtained at Visit 1 and at time points more than one week after dosing.

6.9 Data Monitoring Committee

A Data Monitoring Committee (DMC) will be implemented to review safety data during scheduled periodic reviews. DMC membership will consist of a minimum of 3 individuals who are external to the site and sponsor, including at least 1 statistician. Subjects will be enrolled in a stepwise manner in each of the three vaccine dosage cohorts (cohort 1: low dosage of RSV F subunit vaccine [45 µg], cohort 2: middle dosage of RSV F subunit vaccine [90 µg], and cohort 3: high dosage of RSV F subunit vaccine [135 µg]). The DMC will review safety data, as described in the DMC charter and in the statistical analysis plan, after enrollment of the first 12 subjects in each cohort, and shortly after Visit 2 (seven days after the first dose), before proceeding with enrollment of the remaining subjects in each cohort. In addition, the DMC will review safety data, as described in the DMC charter and in the statistical analysis plan, after enrollment of each cohort has been completed before proceeding with enrollment of the subsequent cohort. Preferably, the DMC will firstly receive blinded data and only in case it is deemed necessary to evaluate unblinded data (either at group or subject level), unblinded information will be released to the DMC members only. In such a circumstance, an NVD independent statistician and a randomization programmer will have access to unblinded data.

Criteria to assess safety will include, but may not be limited to, immediate AEs within 30 minutes, solicited and unsolicited AEs within 7 days and 28 days after each vaccination, respectively, SAEs (including AESI and NOCD), and clinical laboratory safety parameters. The specific roles and responsibilities of the DMC members and other personnel involved will be documented in the DMC charter. The DMC charter will be

finalized before the start of the study and will pre-define all safety criteria to be met, as well as any stopping rules set by the DMC, if necessary.

7.0 ENDPOINTS AND STATISTICAL ANALYSES

7.1 Endpoints

For the immunogenicity evaluation:

- Values below the detection limit (1:100) will be set at 1:50.
- Antibody concentrations will be logarithmically transformed (base 10).

7.1.1 Primary Endpoint(s)

Primary Immunogenicity Endpoints:

1. Geometric mean titer (GMT) of the serum anti-RSV neutralizing antibody (NAb) titer at Day 57 (28 days after the second dose).
2. Proportion of subjects with a ≥ 4 -fold increase in serum anti-RSV NAb titer from Day 1 (baseline) to Day 57 (28 days after the second dose).

7.1.2 Secondary Immunogenicity Endpoints

1. GMT of the serum anti-RSV NAb titer at Day 1 (baseline), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose).
2. Proportion of subjects with a ≥ 4 -fold increase in serum anti-RSV NAb titer from Day 1 (baseline) to all other time points (Day 29 [28 days after the first dose] and Day 181 [six months after the first dose]).
3. Proportion of subjects at Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose) who achieve serum anti-RSV NAb titers greater than the 3rd quartile of serum anti-RSV NAb titers at Day 1 (baseline).
4. Reverse cumulative distribution of serum anti-RSV NAb titers at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).
5. GMT of the serum total binding antibody to each of the RSV proteins F, G and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).

6. Proportion of subjects with a ≥ 4 -fold increase in serum total binding antibody titers to each of the RSV proteins F, G and N from Day 1 (baseline) to all time points (Day 29 [28 days after the first dose], Day 57 [28 days after the second dose], and Day 181 [six months after the first dose]).
7. Proportion of subjects at Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose) who achieve serum total binding antibody titers to each of the RSV proteins F, G, and N greater than the 3rd quartile of serum total binding antibody titers to RSV protein F at Day 1 (baseline).
8. Reverse cumulative distribution of serum total binding antibody to each of the RSV proteins F, G, and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose) and Day 181 (six months after the first dose).
9. Ratio of RSV F subunit serum neutralizing antibody (NAb) titer to each of the RSV F subunit serum total binding antibody titers to RSV proteins F, G and N at Day 1 (baseline), Day 29 (28 days after the first dose), Day 57 (28 days after the second dose), and Day 181 (six months after the first dose).

7.1.3 Secondary Efficacy Endpoints

There are no secondary efficacy endpoints for this study.

7.1.4 Safety Endpoints

Safety will be assessed by measuring the frequency of local and systemic solicited adverse events (AEs), unsolicited AEs, serious adverse events (SAEs), new onset of chronic diseases (NOCD), adverse event of special interest (AESIs), and safety laboratory data for all subjects.

- Reactogenicity will be assessed by the percentage and frequency of subjects with solicited local and solicited systemic adverse events up to 7 days after each vaccination and calculated for four time intervals after each vaccination: 30 minutes, Days 1-3 (without 30 min), Days 4-7 (without 30 min), Days 1-7 (without 30 min).
- Unsolicited AEs will be assessed by the percentage and frequency of subjects with any unsolicited AEs for 28 days after each vaccination.
- Percentage and frequency of subjects with SAEs, non-scheduled physician visits, unsolicited medically attended AEs, unsolicited AEs leading to study withdrawal, NOCDs, and AESIs will be collected from study start until study completion.

7.1.5 Other Endpoints

No other endpoints are included in this study.

7.1.6 Exploratory Endpoints

1. Predominant isotype of the RSV-specific serum antibody at multiple time points.
2. RSV-specific immune response against different RSV group A and group B strains or engineered RSV antigens at multiple time points.
3. Frequency of B cells secreting RSV-specific antibodies at Day 1 (baseline), Day 8 (seven days after the first dose), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose).
4. Diversity of the B-cell repertoire at Day 1 (baseline), Day 8 (seven days after the first dose), Day 29 (28 days after the first dose), and Day 181 (six months after the first dose).

7.2 Success Criteria

In the absence of formal statistical hypotheses, the study will not be declared positive or negative according to given rules. The selection of an appropriate vaccine dosage and adjuvant for future studies will be determined by the joint evaluation of the immunogenicity and safety profiles of each dosage/adjuvant group.

7.2.1 Success Criteria for Primary Objectives

Not applicable.

7.2.2 Success Criteria for Secondary Immunogenicity Objectives

Not applicable.

7.2.3 Success Criteria for Secondary Efficacy Objectives

There are no secondary efficacy objectives in this study.

7.2.4 Success Criteria for Safety Objectives

Not applicable.

7.3 Analysis Sets

7.3.1 All Enrolled Set

All screened subjects who provide informed consent and provide demographic and/or baseline screening assessments, regardless of the subject's randomization and treatment status in the trial.

7.3.2 Exposed Set

All subjects in the Enrolled Population who receive a study vaccination.

7.3.3 Full Analysis Set (FAS) Immunogenicity Set

All subjects in the Enrolled Population who:

- receive a study vaccination AND provide immunogenicity data at relevant time points.

Subjects will be included in the FAS if they have any evaluable serum samples at pre- and post-baseline.

FAS populations will be analyzed "as randomized" (i.e., according to the vaccine a subject was designated to receive, which may be different from the vaccine the subject actually received).

7.3.4 Per Protocol (PP) Population, Immunogenicity Set

All subjects in the FAS Immunogenicity Population who:

- Are not excluded due to reasons (see [section 7.3.8](#)) defined prior to unblinding or analysis
- Examples for subjects excluded due to other reasons than major protocol deviations are:
 - Subjects who withdrew informed consent
 - Premature withdrawals due to adverse events
- Exclusions need to be considered by objective/time point, i.e., sometimes not all data of a subject but only part of the subject's data will be removed from the PPS analysis.

If a subject shows evidence of natural RSV infection during the trial they may not be part of the PPS. However, if evidence of natural RSV infection occurs at Visit 9 or

later, that subject will be included in the PPS analysis through Visit 5, but will not be included in the PPS analysis from Visit 9 through the end of the study.

The complete list of all PPS, based on objective and time points, will be provided in the Statistical Analysis Plan.

7.3.5 Safety Set

Safety Set (solicited adverse events and other solicited reactions)

All subjects in the Exposed Population who:

- Provide post-vaccination reactogenicity data

Safety Set (unsolicited adverse events)

All subjects in the Exposed Population who:

- Have post-vaccination unsolicited adverse event records

Safety Set (overall)

All subjects in the Exposed Population who:

- Have either post-vaccination adverse event or reactogenicity records

Subjects will be analyzed as "treated" (i.e., according to the vaccine a subject received, rather than the vaccine to which the subject may have been randomized).

7.3.6 Other Analysis Sets

All subjects who are screened, i.e., consented but not yet randomized/enrolled.

7.3.7 Subgroups

The subgroup of subjects undergoing additional evaluations for exploratory objectives will not be part of this analysis. Results will be available after the Clinical Study Report is issued.

7.3.8 Protocol Deviations

A protocol deviation is any change, divergence, or departure from the study design or procedures of a study protocol. An exclusion refers to a protocol deviation that is used to remove data from an analysis population at the time of analysis. Relevant protocol

deviations will be defined as exclusionary from the analysis according to protocol objectives and endpoints, which will be specified in the statistical analysis plan.

Any deviation that affects the subject's rights, safety, or well-being and/or the completeness, accuracy and reliability of the study data constitutes a major protocol deviation. Changes or alterations in the conduct of the trial which do not have a major impact on the subject's rights, safety or well-being, or the completeness, accuracy and reliability of the study data are considered minor protocol deviations. Major and minor deviations will be reviewed to determine the final list of deviations that will be used for exclusion from the analysis set(s). This will be defined in the statistical analysis plan prior to unblinding.

The following deviations are considered major:

- A subject received incorrect study vaccine or dose of study vaccine
- A subject met withdrawal criteria during the study but was not withdrawn
- A subject received an excluded medication or vaccine
- A subject was enrolled but does not meet the protocol's eligibility criteria
- A subject with no safety data
- Inadvertent loss of samples or data that support the analysis of primary objectives
- Failure to obtain informed consent prior to initiation of study-related procedures
- Falsifying research or medical records.

Subjects who terminate study participation prematurely for reasons such as withdrawal of consent, adverse event (including death), or administrative reason do not represent protocol deviations, nor are the missing assessments that should otherwise have been collected for that subject later in the study considered protocol deviations.

Pre-specified reasons for delay or cancellation of study vaccination as reflected in [sections 4.3](#) and [4.4](#) do not constitute protocol deviations.

All protocol deviations will be classified into major and minor. Major protocol deviations will be summarized by vaccine, center (overall) and grouped into the different categories as defined above. The site monitor will keep the investigator informed of minor and major protocol deviations, so that the investigator can comply with reporting these deviations to the local EC/IRB according to their institutional policy.

Prior to unblinding, designated staff at the Sponsor will develop a memo that describes the selected deviations that are identified as exclusions from analysis populations. This memo will be included in the trial master file.

7.4 Analysis Plan

7.4.1 Analysis of Demographic and Baseline Characteristics

Descriptive statistics (mean, standard deviation, median, minimum and maximum) for age, height and weight at enrollment will be calculated by overall and by vaccine group.

Distributions of subjects by sex and ethnic origin will be summarized.

7.4.2 Analysis of Primary Objectives

7.4.2.1 Statistical Hypotheses for Primary Objectives

There are no statistical hypotheses being tested in the study.

7.4.2.2 Analysis Populations for Primary Objectives

PPS will be used for the primary objective analyses. The primary immunogenicity analyses will be based on the per-protocol set. If the full analysis set (FAS) and PP differ by more than 10%, primary immunogenicity analyses will also be conducted on the FAS.

7.4.2.3 Statistical Methods for Primary Objectives

For the primary immunogenicity analyses the following statistical methods will be applied:

GMTs: For each treatment group within each cohort, GMTs of the serum anti-RSV NAb along with their associated 95% confidence intervals (CIs) will be computed by exponentiation of the corresponding log-transformed means and 95% CIs.

The statistical analyses for GMTs will be conducted using an ANCOVA model with dosage and adjuvant as factors and baseline antibody level as covariate. Interactions between factors will be investigated and further described in the statistical analysis plan to be finalized and approved before the start of the trial.

Fold-rise: Proportions of subjects with a ≥ 4 -fold increase in serum anti-RSV NAb titer from baseline will be presented, for each treatment group within each cohort, together with their two-sided 95% Clopper-Pearson CIs.

7.4.2.4 Sample Size and Power Considerations of Primary Objectives

Sample size is not driven by statistical assumptions for formal hypothesis testing as previous clinical trial data are not available and clinically meaningful group differences

are not established. In addition, values for a correlate of protection are not known at present stage. Nevertheless, some information from the neutralization assay for human serum was provided.

Hence, the number of proposed subjects will be sufficient to provide a descriptive summary of the safety and immunogenicity of the RSV F subunit vaccine.

Safety

With 24 subjects in each active vaccine group of each cohort, the probability of observing at least one adverse event per group is 90%, if the actual rate of the event is 9.1%. For placebo, with 72 subjects, for the same actual rate of the event, the probability to detect at least one event increases to 99.9%.

When the frequency of an adverse event is greater than 17.5%, there is 99% probability of observing one or more subjects presenting with such an AE in each of the active vaccine groups.

Immunogenicity

With 22 evaluable subjects per group, a two-sided 95% confidence interval (CI) for a single GMT of the anti-RSV NAb will extend 0.167 from the observed mean, assuming a standard deviation of 0.400 (in log scale). Analogously, with an observed proportion of subjects with ≥ 4 -fold increase in NAb titers from Visit 1 (baseline) to Visit 9 (28 days after the second dose) equal to 50%, the 95% CI around that value will be from 28.2% to 71.8% (Clopper-Pearson method).

7.4.2.5 Analysis of Safety Objectives

7.4.2.5.1 Analysis of Extent of Exposure

The number of subjects who received one and two doses of vaccine will be summarized by vaccine group.

7.4.2.5.2 Analysis of Solicited Local and Systemic Adverse Events and Other Reactions

All solicited adverse events will be summarized according to defined severity grading scales.

Frequencies and percentages of subjects experiencing each adverse event will be presented for each symptom severity. Summary tables showing the occurrence of any local or systemic adverse event overall and at each time point will also be presented.

Post-vaccination solicited adverse events reported from day 1 to day 7 will be summarized for the intervals day 1-3, day 4-7, day 1-7 by maximal severity and by vaccine group, excluding the 30 minute measurement, which will be summarized separately. The severity of solicited local adverse events, including injection site induration, swelling, and erythema will be summarized according to categories based on linear measurement: 25 to 50 mm, 51 to 100 mm, >100 mm.

Injection site pain and systemic reactions (except fever) occurring up to 7 days after each vaccination will be summarized according to “mild”, “moderate” or “severe”.

Each solicited local and systemic adverse event will also be further summarized as “none” versus “any”.

Implausible measurements (for further definition see analysis plan) will be left out of the analysis.

Use of antipyretics and analgesics will be summarized by frequency, by type of use (prophylactic versus treatment), and percentage of subjects reporting use. The influence of antipyretics and analgesics use on the occurrence of specific adverse events (e.g., fever) will be assessed.

Body temperature will be summarized by 0.5 °C increments from 36.0 °C up to ≥ 40 °C and will be broken down according by route of measurement.

7.4.2.5.3 Analysis of Spontaneously Reported Adverse Events

All the adverse events occurring during the study, judged either as probably related, possibly related, or not related to vaccination by the investigator, will be recorded.

The original verbatim terms used by investigators to identify adverse events in the CRFs will be mapped to preferred terms using the MedDRA dictionary. The adverse events will then be grouped by MedDRA preferred terms into frequency tables according to system organ class. All reported adverse events, as well as adverse events judged by the investigator as at least possibly related to study vaccine, will be summarized according to system organ class and preferred term within system organ class. These summaries will be presented by vaccination group. When an adverse event occurs more than once for a subject, the maximal severity and strongest relationship to the vaccine group will be counted.

Separate summaries will be produced for the following categories:

- serious adverse events
- adverse events that are possibly or probably related to vaccine
- adverse events of special interest
- new onset of chronic disease adverse event leading to withdrawal
- adverse events leading to a medically attended visit
- adverse event by data source

Data listings of all adverse events will be provided by subject. In addition, adverse events in the categories above will be provided as listed data.

7.4.2.5.4 Analysis of Safety Laboratory Values

The investigator must assess all safety laboratory results (see section 3.5.3). Clinically significant modifications in blood chemistry, hematology, and urinalysis test values will be assessed by medical judgment based on interpretation of deviations from the institution's normal values and, whenever possible, graded according to the criteria included in CBER Guidance for Industry "Toxicity Grading Scale for Healthy Adult and Adolescent Volunteers Enrolled in Preventive Vaccine Clinical Trials" dated September 2007 (see Appendix D, E, and F).

The following information will be displayed:

- Change in absolute laboratory values from Visit 1 (baseline) to Visit 2 (seven days after the first dose), Visit 5 (28 days after the first dose), Visit 6 (seven days after the second dose), and Visit 9 (28 days after the second dose)
- 3 x 3 shift tables by visit, using the categorization of low, normal, and high

7.4.3 Analysis of Key Secondary Immunogenicity Objectives

This study does not include key secondary immunogenicity objectives.

7.4.4 Analysis of Key Secondary Efficacy Objectives

This study does not include key secondary efficacy objectives.

7.4.5 Analysis of Key Secondary Other Objectives

This study does not include key secondary efficacy objectives.

7.4.6 Analysis of Non-Key Objectives

The same statistical methods as described for primary objectives will be applied to the secondary objectives, which are either GMTs or proportions.

Reverse cumulative distribution plots for antibody titers will be provided by treatment group.

Non-Key Immunogenicity Objectives

Not applicable.

Non-Key Exploratory Objectives

Not applicable.

7.5 Planned Interim Analysis

An Interim Analysis will be performed on the immunogenicity and safety data collected from all subjects at Visits 1, 5, and 9.

There will not be any statistical penalties applied as the interim analysis will not trigger changes in the study conduct (e.g. sample size re-estimation, cancellation of vaccine groups, early stop). The site staff will remain blinded through the end of the study and will not have access to unblinded information.

The study statistician and the statistical programmer will be the only NVD people to be unblinded at subject level (i.e. full unblinding) for the interim analysis. Other NVD personnel will only receive grouped unblinded information with no possibility to guess the treatment allocation for a single subject.

A thorough description of how data will be secured will be reported in the Data Security Plan (DSP).

Details on the interim analysis conduct will be described in the Statistical Analysis Plan. An interim CSR will not be produced.

8.0 SOURCE DOCUMENTATION, STUDY MONITORING, AND AUDITING

Study monitoring and auditing will be standardized and performed in accordance with the sponsor's or delegated contract research organization's (CRO) standard operating procedures and applicable regulatory requirements (e.g., FDA, EMA, and ICH guidelines).

Prior to enrollment of the first study subject, NVD or delegate will train investigators and/or their study staff on the study protocol, all applicable study procedures, documentation practices (including signing of the source data agreement (SDA, see [section 8.1](#)) and all electronic systems. CRFs supplied by the sponsor must be completed for each enrolled subject (see [section 7.3.1](#) for definition of enrolled subject). Documentation of screened but not enrolled subjects must be maintained at the site and made available for review by the site monitor. All data entries as well as study related documents will be checked by the sponsor and/or site monitor. In addition, the investigator and site staff will be made aware of the plans to monitor the data collected at the site.

8.1 Source Documentation

Prior to the start of the study, the site staff participating in the study conduct will be trained on what documents will be required for review as source documentation (i.e., original records, laboratory reports, medical records, subject diaries). The kinds of documents that will serve as source documents will be specified in the SDA. The SDA will be finalized and available for further review prior to first subject, first visit.

In addition, source documentation **must** include all of the following: subject identification (on each page), eligibility and participation, proper informed consent procedures, dates of visits, adherence to protocol procedures, adequate reporting and follow-up of adverse events, documentation of prior/concomitant medication/vaccines, study vaccine receipt/dispensing/return records, study vaccine administration information, any data collected by a telephone conversation with the subject, and date of completion and reason.

The subject or the subject's parents or legal guardian(s) must also allow access to the subject's medical records. Each subject, or the subject's parent(s) or legal guardian(s), must be informed of this prior to the start of the study and consent for access to medical records may be required in accordance with local regulations.

All safety data reported by subjects must be written down on source documents prior to entry of the data into CRFs. If there are multiple sources of information (e.g., diary card, verbal report of the subject, telephone contact details, medical chart) supporting the diagnosis of an adverse event, these sources must be identified in the source documents,

discrepancies between sources clarified, the ultimate diagnosis must be justified and written in the source documents, and this diagnosis must be captured in the adverse event CRF (AE CRF). The AE CRF must also capture which source(s) of information were used to determine the adverse event (e.g., subject recall, medical chart, diary card, and/or other sources).

8.2 Study Monitoring and Source Data Verification

A contract research organization (CRO) may be involved in the monitoring of protocol conduct and data entry. If a CRO is involved in study oversight, the name and address of this CRO will be located in the investigator site file. Prior to enrollment of the first study subject, NVD will develop a Clinical Monitoring Plan to specify how monitoring will be performed for the study.

Study progress will be monitored by NVD or its representative (e.g., a CRO) as frequently as necessary to ensure:

- that the rights and well-being of human subjects are protected
- the reported trial data are accurate, complete, and verifiable from the source documents and
- the conduct of the trial is in compliance with the current approved protocol/amendment(s), GCP and applicable regulatory requirements

Contact details for the team involved in study monitoring will be identified in a handout located in the Investigator Site File. Study data recorded on CRFs will be verified by checking the CRF entries against source documents in order to ensure data completeness and accuracy as required by study protocol. Additional documents such as the investigator site file, pharmacy records, and informed consent documentation must also be available for review if requested. Arrangements for monitoring visits will be made in advance in accordance with the monitoring plan, except in case of emergency.

The investigator and/or site staff must make source documents of subjects enrolled in this study available for inspection by NVD or its representative at the time of each monitoring visit. These documents must also be available for inspection, verification and copying, as required by regulations, by officials of the regulatory health authorities (e.g., FDA, EMA and others) and/or ECs/IRBs. The investigator and study site staff must comply with applicable privacy, data protection and medical confidentiality laws for use and disclosure of information related to the study and enrolled subjects.

9.0 DATA MANAGEMENT

9.1 Data Entry and Management

In this study, all clinical data (including, but not limited to, AE/SAEs, concomitant medications, medical history, and physical assessments), reactogenicity data, and immunogenicity data will be entered onto electronic case report forms (eCRFs) in a timely fashion by the investigator and/or the investigator's dedicated site staff. Data entered onto eCRFs are stored on a secure website. The data collected on this secure website are assimilated into an electronic data capture (EDC) system, which is compliant with 21 Part 11 policies of the Code of Federal Regulations. The data system includes password protection and internal quality checks. The EDC will be designed and validated by NVD prior to activation for data entry by sites. The investigator or designated delegate must review data entered and electronically sign the eCRFs to verify their accuracy.

Access to the EDC system for data entry or review will require training and distinct individual access code assignments to those site staff members who will be entering study data and those involved in study oversight who may review study data. Data are collected within EDC, to which the sponsor and site monitors have exclusively "read only" access. eCRF data will be reviewed routinely by study personnel from NVD and clinical monitors.

All serology results produced by Clinical Serology, NVD will be entered into the Seroad database by NVD's Clinical Serology Laboratory, Marburg. All results will be checked in the laboratory for validity and completeness.

Electronic Data Transfer (EDT) is one method used by NVD for collecting laboratory data. The full-service laboratory (i.e., central laboratory) will send data as electronic files by a secured method (e.g., via diskette, CD, as an encrypted file attachment on electronic mail, or as a direct transfer into a specified server directory) to NVD's BCDM department. The data file is pre-processed and loaded by a member of the BCDM team into the study database. The laboratory will submit a results file containing the tests and the results as specified in the protocol. If the laboratory provides the service, it will also submit a Demography (DEMOG) file containing the subject's demographic information. If the file includes results of data blinded to personnel in clinical research, the source will provide a separate results file that will be loaded into a separate laboratory table.

For this protocol, immunology laboratory data and/or safety laboratory data from external laboratories will be transmitted via EDT.

9.2 Data Clarification

As part of the conduct of the trial, NVD may have questions about the data entered by the site, referred to as queries. The monitors and the sponsor are the only parties that can generate a query.

For eCRF trials, all corrections and clarifications will be entered into the EDC and will be identified by the person entering the information, the reason for the change, as well as the time of the changes made. If changes are made to a previously and electronically signed CRF, the investigator must confirm and endorse the changes.

9.3 Data Coding Procedures

Coding of Adverse Events, Medical History, and Prior and Concomitant Medications will be performed using standard dictionaries as described in the Data Management Plan.

9.4 Data Protection

NVD respects the subjects' rights to privacy and will ensure the confidentiality of their medical information in accordance with all applicable laws and regulations.

The sponsor as Data Controller according to the European Directive on the protection of individuals with regard to the processing of personal data and on the free movement of such data [95/46/EC] confirms herewith compliance to Directive 95/46/EC in all stages of Data Management.

10.0 RECORD RETENTION

Investigators must retain all study records required by NVD and by the applicable regulations in a secure and safe facility. The investigator must consult a NVD representative before disposal of any study records, and must notify the sponsor of any change in the location, disposition, or custody of the study files. Essential documents must be retained until at least 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational product. “Essential documents” are defined as documents that individually and collectively permit evaluation of the conduct of a trial and the quality of the data produced. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by an agreement with the sponsor. The Committee for Human Medicinal Products for Human Use (CHMP) requires retention for the maximum period of time permitted by the institution, but not less than 15 years (ICH E6, 4.9.5). It is the responsibility of the sponsor to inform the investigator/institution as to when these documents no longer need to be retained (ICH E6, 5.5.12).

These principles of record retention will also be applied to the storage of laboratory samples, provided that the integrity of the stored sample permits testing.

11.0 USE OF INFORMATION AND PUBLICATION

NVD assures that the key design elements of this protocol will be posted in a publicly accessible database such as clinicaltrials.gov, and in compliance with current regulations.

NVD also assures that key results of this clinical trial will be posted in a publicly accessible database within the required time-frame from the last subject's last study visit as dictated by applicable regulations.

Further to legislated data disclosure, NVD will ensure that as far as possible results of this study will be published as scientific/clinical papers in high-quality peer-reviewed journals. Preparation of such manuscripts will be made with full collaboration of principal investigators and in accordance with the current guidelines of Good Publication Practice ([Graf 2009](#)).

NVD must be notified of any intent to publish data collected from the study and prior approval from NVD must be obtained prior to publication.

12.0 ETHICS

12.1 Regulatory and Ethical Compliance

This clinical study was designed and shall be implemented and reported in accordance with the ICH Harmonized Tripartite Guidelines for Good Clinical Practice, with applicable local regulations including European Directive 2001/20/EC, US Code of Federal Regulations Title 21, Novartis codes on protection of human rights, and with the ethical principles laid down in the Declaration of Helsinki (European Council 2001, US Code of Federal Regulations, ICH 1997).

12.2 Informed Consent Procedures

Eligible subjects may only be included in the study after providing written informed consent or assent, as described in [section 3.2.1](#). Before the start of the trial, the investigator will have the informed consent and any other materials that will be provided to the subjects reviewed and approved by the IRB/EC. This review and approval will be documented and stored with other study documents. The investigator or designee must fully inform the subject or legal guardian of all pertinent aspects of the trial. A copy of the written informed consent will be given to the subject or the designee. The subject/designee must be allowed ample time to ask about the details of the trial and to make a decision as to whether or not to participate in the study. The subject and/or legal guardian must sign the consent form indicating their agreement to participate in the study before any study-related procedures are conducted. If the subject and/or legal guardian is unable to read and write, a witness must be present during the informed consent discussion and at the time of informed consent signature.

Prior to the start of the study, NVD will provide to investigators a separate document with a proposed informed consent form that complies with the ICH GCP guideline and regulatory requirements and is considered appropriate for this study. Any changes to the proposed consent form suggested by the investigator must be agreed to by NVD before submission to the IRB/EC and a copy of the approved version must be provided to the NVD monitor after IRB/EC approval.

Women of child bearing potential should be informed that taking the study medication may involve unknown risks to the fetus if pregnancy were to occur during the study and agree that in order to participate in the study they must adhere to the contraception requirements indicated in the protocol for the duration of the study. If case of doubts on the ability of a subject to adhere to these requirements, that subject should not be allowed in the study

12.3 Responsibilities of the Investigator and IRB/EC

The protocol and the proposed informed consent form must be reviewed and approved by a properly constituted IRB/EC before study start. Properly constituted IRB/EC is defined in ICH Guideline for Good Clinical Practice E6 (R1), Section 3 (ICH 1997). A signed and dated statement that the protocol and informed consent have been approved by the IRB/EC must be given to NVD before study initiation. Prior to study start and at any time the protocol is amended during study conduct, the investigator is required to sign a protocol signature page confirming his/her agreement to conduct the study in accordance with these documents and all of the instructions and procedures found in this protocol and to give access to all relevant data and records to NVD monitors, auditors, Novartis Clinical Quality Assurance representatives, designated agents of NVD, IRBs/ECs, and regulatory authorities as required. If an inspection of the clinical site is requested by a regulatory authority, the investigator must inform NVD immediately that this request has been made.

The investigator is also responsible for the following:

- maintaining a list of appropriately qualified persons to whom the investigator has delegated significant trial-related duties
- demonstrating the capability of recruiting the required number of suitable subjects within the recruitment period
- demonstrating sufficient time and staffing to properly conduct and complete the study within the agreed trial period
- ensuring that all persons assisting with the study are adequately informed about the protocol, the investigational product(s), and their study-related duties and functions
- ensuring that appropriately trained health care professionals are responsible for all study-related medical decisions and for ensuring appropriate medical care of subjects experiencing any adverse event related to the study
- if permission to do so is given by the subject, ensuring that the subject's primary healthcare provider is informed of the subject's participation in the study.

The investigator should not implement any deviation from, or changes of the protocol without agreement by the sponsor and prior review and documented approval/favourable opinion from the IRB/IEC of an amendment, except where necessary to eliminate an immediate hazard(s) to trial subjects, or when the change(s) involves only logistical or administrative aspects of the trial (e.g., change in monitor(s), change of telephone number(s)). In addition, the investigator, or person designated by the investigator, should document and explain any deviation from the approved protocol.

The investigator may implement a deviation from, or a change of, the protocol to eliminate an immediate hazard(s) to trial subjects without prior IRB/IEC approval/favourable opinion. As soon as possible, the implemented deviation or change, the reasons for it, and, if appropriate, the proposed protocol amendment(s) should be submitted:

- (a) to the IRB/IEC for review and approval/favourable opinion,
- (b) to the sponsor for agreement and, if required,
- (c) to the regulatory authority(ies).

12.4 Protocol Adherence

Investigators will apply due diligence to avoid protocol deviations. Under no circumstances should the investigator contact NVD or its agents, if any, monitoring the trial to request approval of a protocol deviation, as no authorized deviations are permitted. If the investigator feels a change to the protocol would improve the conduct of the study this must be considered a protocol amendment, and unless such an amendment is agreed upon by NVD and approved by the IRB/EC it cannot be implemented. All significant protocol deviations will be recorded and reported in the Clinical Study Report.

12.5 Protocol Amendments

An amendment is a written description of change(s) to or formal clarification of a study protocol which may impact on the conduct of the clinical study, potential benefit of the clinical study, or may affect subject safety, including changes of study objectives, study design, subject population, sample sizes, study procedures, or significant administrative aspects. An administrative change of a study protocol is a minor correction or clarification that has no significant impact on the way the clinical study is to be conducted and no effect on subject safety (e.g., change of telephone number(s), logistical changes). Protocol amendments must be approved by NVD, Health Authorities where required, and the IRB/EC. In cases when the amendment is required in order to protect the subject safety, the amendment can be implemented prior to IRB/EC approval. Notwithstanding the need for formal approval of a protocol amendment, the investigator is expected to take any immediate action required for the safety of any subject included in this study, even if this action represents a deviation from the protocol. In such cases, NVD should be notified of this action, the IRB/EC at the study site, and, if required by local regulations, the relevant health authority) should be informed within 10 working days.

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APPENDIX A: ADVERSE EVENTS OF SPECIAL INTEREST (AESI)

AEs of special interest (AESI) will include those listed in below. The AESIs will be defined according to the following MedDRA preferred terms (underlined).

Neuroinflammatory disorders: optic neuritis, uveitis, multiple sclerosis, demyelinating disease, transverse myelitis, Guillain-Barre syndrome, myasthenia gravis, encephalitis, ADEM, neuritis, Bell's palsy

Musculoskeletal and connective tissue disorders: rheumatoid arthritis, juvenile rheumatoid arthritis, polymyalgia rheumatica, psoriatic arthropathy, ankylosing spondylitis, systemic lupus erythematosus, cutaneous lupus, Sjogren's syndrome, scleroderma, dermatomyositis, polymyositis, mixed connective tissue disease, reactive arthritis

Vasculidites: temporal arteritis, Wegener's granulomatosis, mixed connective tissue disease

Gastrointestinal disorders: Crohn's disease, ulcerative colitis, inflammatory bowel disease (non-specific), celiac disease, autoimmune hepatitis, primary sclerosing cholangitis, primary biliary cirrhosis

Renal disorders: glomerulonephritis, nephritis, renal vasculitis

Cardiac disorders: carditis, pericarditis, myocarditis, cardiomyopathy

Skin disorders: psoriasis, vitiligo, Raynaud's phenomenon, erythema nodosum, autoimmune bullous skin diseases, Stevens-Johnson syndrome

Hematologic disorders: autoimmune hemolytic anemia, idiopathic thrombocytopenic purpura, antiphospholipid syndrome, pernicious anemia

Metabolic disorders: autoimmune thyroiditis, Grave's or Basedow's disease, Hashimoto thyroiditis, insulin-dependent diabetes mellitus, Addison's disease

Others: sarcoidosis

APPENDIX B: TOXICITY GRADING SCALES FOR SOLICITED LOCAL ADVERSE EVENTS

(Adapted from CBER 2007b)

Adverse event Following Administration of Injectable Product	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Pain	Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Induration / Swelling / Erythema ¹	25 – 50 mm	51 – 100 mm	> 100 mm

* This toxicity grading scale is adapted from CBER 2007 to enable ease of reporting by Subjects in the source documents for 'patient reported' solicited adverse events. This toxicity grading scale is an NVD standard that is used for patient reporting. 'Grade 4' is not listed here but will be defined in the statistical analysis plan as necessary

¹ These ranges will be included in analyses, summarized: 1 to 2.4 cm, 2.5 to 5.0 cm, 5.1 to 10.0 cm, > 10.0 cm.

APPENDIX C: TOXICITY SCALES FOR SOLICITED SYSTEMIC ADVERSE EVENTS

(Adapted from CBER 2007b)

Systemic Adverse event		Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)
Fever	°C	38.0 – 38.4	38.5 – 38.9	39.0 – 40
	°F	100.4 – 101.1	101.2 - 102	102.1 - 104
Chills		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Nausea		Nausea present but not interfering with oral intake	Nausea leading to decreased oral intake	Nausea leading to minimal to no oral intake
Myalgia		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Arthralgia		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Headache		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Fatigue		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Diarrhea		2-3 loose stools /24 hours	4-5 loose stools /24 hours	6 or more watery stools /24 hours or requires outpatient IV hydration
Loss of appetite		Loss of appetite without decrease in oral intake	Decreased oral intake without weight loss	Decreased oral intake with weight loss
Cough		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Rhinorrhea		Present but does not interfere with activity	Interferes with activity	Prevents daily activity
Wheezing		Present but does not interfere with activity	Interferes with activity	Prevents daily activity

* This toxicity grading scale is adapted from CBER 2007b to enable ease of reporting by Subjects in the source documents for 'patient reported' solicited adverse events. This toxicity grading scale is an NVD standard that is used for patient reporting. 'Grade 4' is not listed here but will be defined in the statistical analysis plan as necessary

**APPENDIX D: TOXICITY SCALES FOR LABORATORY ABNORMALITIES
(SERUM)**

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)**
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
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 BUN 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
Phosphorous – hypophosphatemia mg/dL	2.3 – 2.5	2.0 – 2.2	1.6 – 1.9	< 1.6
CPK – mg/dL	1.25 – 1.5 x ULN***	1.6 – 3.0 x ULN	3.1 – 10 x ULN	> 10 x 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 x ULN	2.1 – 3.0 x ULN	73.1 – 10 x ULN	> 10 x ULN

Serum *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)**
Liver Function Tests – ALT, AST increase by factor	1.1 – 2.5 x ULN	2.6 – 5.0 x ULN	5.1 – 10 x ULN	> 10 x ULN
Bilirubin – when accompanied by any increase in Liver Function Test increase by factor	1.1 – 1.25 x ULN	1.26 – 1.5 x ULN	1.51 – 1.75 x ULN	> 1.75 x ULN
Bilirubin – when Liver Function Test is normal; increase by factor	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.0 – 3.0 x ULN	> 3.0 x ULN
Cholesterol	201 – 210	211 – 225	> 226	---
Pancreatic enzymes – amylase, lipase	1.1 – 1.5 x ULN	1.6 – 2.0 x ULN	2.1 – 5.0 x ULN	> 5.0 x ULN

* 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.

** 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.

*** ULN” is the upper limit of the normal range.

**** Laboratory values that fall in the normal range may be assigned a category of “Grade 0.”

**APPENDIX E: TOXICITY SCALES FOR LABORATORY ABNORMALITIES
(HEMATOLOGY)**

Hematology *	Mild (Grade 1)	Moderate (Grade 2)	Severe (Grade 3)	Potentially Life Threatening (Grade 4)
Hemoglobin (Female) - gm/dL	11.0 – 12.0	9.5 – 10.9	8.0 – 9.4	< 8.0
Hemoglobin (Female) change from baseline value - gm/dL	Any decrease – 1.5	1.6 – 2.0	2.1 – 5.0	> 5.0
Hemoglobin (Male) - gm/dL	12.5 – 13.5	10.5 – 12.4	8.5 – 10.4	< 8.5
Hemoglobin (Male) change from baseline value – gm/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 – 1500	1501 - 5000	> 5000	Hypereosinophilic
Platelets Decreased - cell/mm ³	125,000 – 140,000	100,000 – 124,000	25,000 – 99,000	< 25,000
PT – increase by factor (prothrombin time)	1.0 – 1.10 x ULN**	1.11 – 1.20 x ULN	1.21 – 1.25 x ULN	> 1.25 ULN
PTT – increase by factor (partial thromboplastin time)	1.0 – 1.2 x ULN	1.21 – 1.4 x ULN	1.41 – 1.5 x 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 (DIC)

* 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.

** "ULN" is the upper limit of the normal range.

*** Laboratory values that fall in the normal range may be assigned a category of "Grade 0."

**APPENDIX F: TOXICITY SCALES FOR LABORATORY ABNORMALITIES
(URINE)**

Urine *	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 for hyperglycemia
Blood (microscopic) – red blood cells per high power field (rbc/hpf)	1 - 10	11 – 50	> 50 and/or gross blood	Hospitalization or packed red blood cells (PRBC) transfusion

* 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.

** Laboratory values that fall in the normal range may be assigned a category of “Grade 0.”

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CLINICAL STUDY PROTOCOL AMENDMENT

Study Number: V122_01

Protocol Title: A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults

Amendment Number 1

Revised Protocol version 2.0 issued on 24 JUN 14

The present amendment reflects changes to the Revised Protocol version 1.0 issued on 08 JAN 14

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DESCRIPTION OF CHANGE(S) AND RATIONALE:

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
Removal of BB-IND	Title page, Synopsis header	Decision taken that V122_01 will not be conducted under IND.
Updated description of randomization. Changed to enrolling all 288 subjects in one of three cohorts, followed by allocation to one of four treatment groups. Combined placebo subjects within each cohort. Clarified wording on enrollment and randomization.	Synopsis Methodology section, Table 1, Section 3.1, Table 3.1-1, Section 3.2.3, Section 3.2.4	Modified wording to more accurately describe how enrollment and randomization will actually be performed.
Removal of “loss of appetite” as a solicited systemic reaction.	Synopsis Safety Measurements section, Section 6.5, Appendix B	Nausea is already included as a solicited AE, and is a better measure than loss of appetite. Including both may be confusing to subjects completing the diary.
Updated list of abbreviations to remove those no longer applicable.	List of Abbreviations	Abbreviations for removed safety labs needed to be deleted.

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
<p>Updated list of Safety labs to remove unnecessary tests. The following tests were deleted: cholesterol, LDH, pancreatic enzymes, fibrinogen, PT, PPT, and D-dimer.</p>	<p>Section 3.5.3, Section 6.7, Appendix C, Appendix D</p>	<p>The removed safety labs did not provide critical information related to subject safety, and added cost and logistical complexity. The list of safety labs has now been reduced to those that provide the most important and useful information for assessing subject safety.</p>
<p>Removed the list of AESIs as an appendix. It will instead be included in the Investigator Site File.</p>	<p>Appendix A, references to Appendix A, and renaming of other appendices</p>	<p>The previous list included in Appendix A was outdated. Moving forward the list of AESIs will be provided outside of the protocol to avoid the need for future amendments.</p>
<p>Added the following statement: “The analysis of the B-cell receptor diversity does not have the intent or the capability to identify or rule out any genetic modifications in the RNA or DNA that are presumably linked to disease or predisposition to disease.”</p>	<p>Section 6.8</p>	<p>This statement is intended as a clarification that no genetic testing is being performed that would provide information related to health status of the subjects.</p>

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CLINICAL STUDY PROTOCOL AMENDMENT

Study Number: V122_01

Protocol Title: A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults

Amendment Number 2

Revised Protocol version 3.0 issued on 26 AUG 14

The present amendment reflects changes to the Revised Protocol version 2.0 issued on 24 JUN 14

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DESCRIPTION OF CHANGE(S) AND RATIONALE:

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
Name of Sponsor updated to Novartis Pharma Services AG	Synopsis header	Study conducted in EU, so sponsor needs to be Novartis Pharma Services AG
Visit windows updated in Time and Events Table: Screening window extended to -28 days, -1/+1 windows applied to Reminder Calls and Phone Calls for Visits 3, 4, 7, and 8	Time and Events Table (Table 2)	Site requested additional screening time and windows for phone calls to better facilitate scheduling of subjects.
INR removed as safety lab assay.	Table of Contents, Section 3.5.3, Section 6.7	INR was erroneously kept in list of safety labs; it is linked to PT, which was previously removed.
<p>Clarified Stopping/Pausing Guidelines:</p> <p>Reordered so SAEs are listed first, then Grade 4 AEs, then Grade 3 AEs.</p> <p>Added that review of SAEs will be done by DMC, IRB/EC, and health authorities. Review of Grade 4 AEs and Grade 3 AEs will be done by the DMC.</p> <p>Grade 3 AEs guideline now also refers to Appendix A</p>	Section 3.6	Clarified guidelines so they are more easily understood.

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
and Appendix B.		
Vaccine Group names updated to align with other study documents. All three cohorts now have Groups A, B, and C. All placebo groups (in all three cohorts) are named Group D.	Table 5.1	Updating Vaccine Groups to 1A, 1B, 1C, 1D, 2A, 2B, 2C, 2D, 3A, 3B, 3C, and 3D allowed for simplified kit design and aligned with other documentation.

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CLINICAL STUDY PROTOCOL AMENDMENT

Study Number: V122_01

Protocol Title: A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults

Amendment Number 3

Revised Protocol version 4.0 issued on 16 DEC 14

The present amendment reflects changes to the Revised Protocol version 3.0 issued on 26 AUG 14

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DESCRIPTION OF CHANGE(S) AND RATIONALE:

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
<p>Exclusion criteria #14 has been updated from “Individuals with any abnormal safety laboratory result at the screening visit” to “Individuals with any clinically significant abnormal safety laboratory result, as judged by the investigator.”</p>	<p>Section 4.2</p>	<p>The investigator must be able to decide if an abnormal safety lab result (based on standard toxicity tables) in an otherwise healthy individual is clinically significant and therefore should exclude the subject from the study. This language is similar to what has been used in other trials.</p>
<p>Safety review of data by the DMC has been changed from Visit 9 to Visit 6, prior to proceeding with enrollment of the subsequent cohort.</p>	<p>Section 3.2.3</p>	<p>As a precedent, DMC reviews within and between cohorts will be performed based on safety data from 7 days after each vaccination. The Stopping/Pausing guidelines defined in Section 3.6 will of course still apply.</p>
<p>Reference to “28 days after each vaccination” has been removed.</p>	<p>Section 6.9</p>	<p>As described above for Section 3.2.3, the DMC review will be performed at 7 days after each vaccination.</p>
<p>In the Times and Events Table, Visit Number row was moved up so it is next to Study Day row.</p>	<p>Table 2</p>	<p>The rows were reordered to make the table easier to read.</p>
<p>References to “major” and “minor” protocol deviations have been updated to “reportable” and “not reportable,” respectively.</p>	<p>Section 7.3.8</p>	<p>Novartis SOP now defines protocol deviations as either CSR reportable or non-reportable.</p>

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CLINICAL STUDY PROTOCOL AMENDMENT

Study Number: V122_01

Protocol Title: A Phase 1 Randomized, Observer Blind, Placebo Controlled, Dosage-Escalation Single Center Study to Evaluate the Safety and Immunogenicity of an RSV Fusion Glycoprotein (F) Subunit Vaccine in Healthy Adults

Amendment Number 4

Revised Protocol version 5.0 issued on 12 APR 16

The present amendment reflects changes to the Revised Protocol version 4.0 issued on 16 DEC 14

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DESCRIPTION OF CHANGE(S) AND RATIONALE:

CHANGE	LOCATION(S) OF CHANGE	RATIONALE FOR CHANGE
Update to sponsor name	Throughout the entire document	Sponsorship change due to acquisition of Novartis Vaccines by GSK
Removal of reference to interim analysis	Synopsis (page 13) Section 3.1 Section 3.3 Section 7.5	Alignment with internal decision made within organization

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