STUDY TITLE

A Phase 2 Randomized, Placebo- and Active-Controlled, Human Influenza A/California/04/2009 (H1N1) Challenge Study Following Administration of an Oral H1N1 HA Adenoviral-Vector Based Seasonal Influenza Vaccine and dsRNA Adjuvant (VXA-A1.1) to Healthy Adult Volunteers

Protocol Number:	VXA-CHAL-201
Phase:	2
Investigational Product:	**VXA-A1.1 Oral Vaccine**
Indication	Prevention of Seasonal Influenza
Sponsor:	Vaxart, Inc. 385 Oyster Point Blvd, Suite 9A South San Francisco, California 94080
Sponsor Contact:	Shaily Jaini Garg, PMP, CCRA Vice President, Clinical Development & Regulatory Affairs Vaxart, Inc. P: 650-521-4496; email: sgarg@vaxart.com
Sponsor Chief Medical Officer:	Dave Liebowitz, MD, PhD Chief Medical Officer Vaxart, Inc. P: 408-340-8605; email: dliebowitz@vaxart.com
Study Medical Monitor:	Michelle Abada, MD Study Medical Monitor WCCT Medelis, Inc. P: (714) 252-0700 Ext 1068; email: Michelle.Ababa@wcct.com

Protocol Date and Version:	**June 8, 2017 (Amendment 3)**
Replaces:	September 27, 2016 (Amendment 2)
	July 27, 2016 (Amendment 1)
	June 28, 2016 (Original)

INVESTIGATOR'S STATEMENT

I agree to conduct the study as outlined in the Protocol entitled:

A Phase 2 Randomized, Placebo- and Active-Controlled, Human Influenza A/California/04/2009 (H1N1) Challenge Study Following Administration of an Oral H1N1 HA Adenoviral-Vector Based Seasonal Influenza Vaccine and dsRNA Adjuvant (VXA-A1.1) to Healthy Adult Volunteers

Amendment 3

in accordance with the guidelines and all applicable government regulations (21 CFR 50, 54, 56, 312; ICH E6), and the Declaration of Helsinki. I have read and understand all sections of the protocol, including Section 9, Administrative Items, and the Investigator's Brochure.

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

I understand that Vaxart, its representatives, and regulatory agencies, shall have access to any source documents relevant to this study including documents that demonstrate protocol and regulatory compliance.

Investigator Printed Name	Signature	Date

STUDY NUMBER	**VXA-CHAL-201 (NCT: 02918006)**
STUDY TITLE	**A Phase 2 Randomized, Placebo- and Active-Controlled, Human Influenza A/California/04/2009 (H1N1) Challenge Study Following Administration of an Oral H1N1 HA Adenoviral-Vector Based Seasonal Influenza Vaccine and dsRNA Adjuvant (VXA-A1.1) to Healthy Adult Volunteers**
SPONSOR	**Vaxart, Inc.**
PHASE	2
STUDY POPULATION	180 healthy adult volunteers age 18 to 49 years (~180 subjects will be vaccinated in Part A to ensure at least 150 subjects are available to participate in the challenge phase in Part B)
NO. OF SITES	1 qualified U.S. Site (with isolation unit)
DURATION OF STUDY PARTICIPATION	One year following successful pre-screening, screening and enrollment, to include: • **PART A:** Vaccination and initial safety follow-up phase (Study Day 1 – Day 90) • **PART B:** Influenza challenge phase (Study Day 90) and long-term safety follow-up (through Day 365 post-vaccination). Enrollment, vaccination, challenge and follow-up visits will be staggered into approximately 6 cohorts of subjects enrolled at approximately 30-day intervals. Individual subject involvement in study activities is estimated to last ~16 months.
INVESTIGATIONAL VACCINE	VXA-A1.1 is an E1/E3-deleted replication-defective Adenovirus serotype 5 vaccine vector for prevention of seasonal influenza H1N1. The vaccine vector encodes for a full-length hemagglutinin (HA) gene from the influenza A/California/04/2009 (H1N1) and a double-stranded RNA (dsRNA) adjuvant (TLR3 agonist). The adjuvant is a short hairpin RNA, expressed as a 21 nucleotide sequence as a tandem sequence in forward and reverse orientations separated by 6 nucleotides that comprise the loop of the RNA. The suspension cell-based production process utilizes a lineage of HEK 293 cells adapted to a protein-free medium. VXA-A1.1 final drug product (DP) is an oral enteric-coated tablet formulation. Subjects will be dispensed 7 vaccine tablets (1.5×10^{10} IU/tablet) to administer a dose of 1.05×10^{11} IU [1×10^{11} IU ±0.5 log]
CONTROL PRODUCTS	Active Comparator: • Fluzone Quadravalent (Fluzone®) influenza vaccine (QIV) suspension for intramuscular (IM) injection (2015-2016 formula, Lot # U1440AB) o The potency of Fluzone® is being continuously monitored by SRID assay to ensure that the vaccine remains within specification during the dosing period of the study. o Stability testing (time 0) was initiated prior to the labeled expiry date (June 30, 2016) of the Fluzone® Lot # U1440AB Placebo: • Oral tablets similar in appearance and number to active oral vaccine tablets • 0.9% Sodium Chloride for Injection, USP

CHALLENGE VIRUS STRAIN AND DOSE	Wild-type Influenza virus (A/California/H1N1 2009) human challenge strain (BB-MF 15915 and BB-MF 15942; WCCT Global). WCCT Global has submitted Letters of Authorization for FDA to refer to and incorporate by reference, any and all information contained in the two Master Files for the use of the influenza challenge strain product described in Protocol VXA-CHAL-201.
RESCUE TREATMENT	Rescue treatment with oseltamivir will be available to any subject who develops severe influenza illness post-challenge, as judged by the Investigator. Otherwise, challenged subjects will receive a 5-day course of therapy if symptomatic at discharge, based on Investigator discretion.

STUDY OBJECTIVES	**Primary Objectives:**
	• Further establish the safety and tolerability of the VXA-A1.1 oral vaccine
	• Determine the clinical efficacy of VXA-A1.1 to protect against illness caused by the homologous A strain influenza virus challenge 3 months following a single immunization in comparison to placebo and QIV
	Secondary Objectives:
	• Determine the ability of VXA-A1.1 to modify disease severity compared to QIV and placebo
	• Determine the correlation of immunogenicity parameters with clinical outcome
	• Determine the quantity and duration of influenza virus shedding by qRT-PCR
	• Evaluate the immunogenicity of VXA-A1.1:
	  • Assess the ability of VXA-A1.1 to elicit serological and cellular immune responses against the matched challenge strain in comparison to licensed QIV
	  • Assess the ability of VXA-A1.1 to elicit more broadly cross- reactive serological immune responses in comparison to licensed QIV
	Exploratory Objectives:
	• Determine the resulting vaccine specific immune response by CyTOF (Mass flow cytometry) and antibody repertoire sequence analysis, comparing oral VXA-A1.1 to the inactivated QIV vaccine for homing, memory, and activation of B and T cells.

STUDY DESIGN, REGIMEN AND DOSING	This is a Phase 2 randomized, placebo- and active-controlled, two-part study in which healthy adult volunteers with low or undetectable pre-existing antibodies against A/California/7/2009(H1N1) pdm09-like virus will be challenged with an influenza A/H1N1 human challenge strain approximately 90 days (+30 day window) after vaccination with a single dose of H1N1 HA Adenoviral-vector based seasonal influenza vaccine and dsRNA adjuvant (VXA-A1.1), an injectable QIV vaccine (Fluzone®), and/or placebo (double-dummy design). An independent Safety Monitoring Committee (SMC) will convene at regular intervals during the influenza challenge period, and also *ad hoc* as needed during the vaccination and challenge periods, to oversee the safety of the study. To accommodate the limited size of the isolation unit that will be utilized for the challenge and post-challenge sequestration period, subjects will move through the study (enrollment, vaccination and challenge) sequentially in a total of 6 cohorts. Each cohort will randomize approximately 30 subjects to obtain approximately 25 subjects per cohort for the challenge phase. Subjects will be randomized in a ratio of 2:2:1 (VXA-A1.1: Fluzone®: Placebo).

STUDY DESIGN, REGIMEN AND DOSING (cont.)	The study will be conducted in two parts. **Part A:** Subjects will be randomized in a double-blinded manner to receive a single administration of one of three treatment arms: • Arm 1: VXA-A1.1 oral vaccine tablets [$1x10^{11}$ IU±0.5 log] + placebo IM injection (n=72 subjects) • Arm 2: Fluzone® IM injection + oral placebo tablets (n=72 subjects) • Arm 3: Placebo IM injection + oral placebo tablets (n=36 subjects) Subjects will return to the site for ~8 visits and be contacted remotely at defined time points to be followed for immunogenicity and safety during study Part A (see Study Schedule section below as well as Schedule of Study Events, Table 1). **Part B:** Following confirmation of eligibility, subjects will be challenged with a wild-type influenza A H1 virus strain (A/H1N1 pdm09) 90 days (+30 day window) following vaccination (~25 subjects/ cohort). Blood samples will be collected to evaluate immunogenicity at pre-challenge. Subjects will remain in the isolation unit for at least 6, and up to 9 days post-challenge (see Study Schedule section below as well as Schedule of Study Events, Table 1). Following challenge, influenza symptoms will be systematically collected using the Flu-PRO questionnaire, which will be completed by the subject once daily in the evening through 14 days post challenge. Additionally, signs of influenza will be assessed by the Investigator once daily via a targeted physical exam during the sequestration period. Blood samples, nasopharyngeal swab samples and tissue paper weights will also be collected (Table 1). Vital signs will be measured approximately every 4 hours, starting at the time of challenge administration. After release from the isolation unit subjects will return to the site 30 days post-challenge for a final efficacy and routine safety (unsolicited AEs) follow-up visit. Part B will continue for purposes of collecting long term safety follow-up (SAEs and AESIs/NOCIs) via monthly phone contacts through 1 year post-vaccination (Day 365).

STUDY SCHEDULE	The following study visits and telephone contacts will be conducted during the study (also see Tables 1a and 1b, Schedule of Study Events): **Part A Visits (Vaccine-related):** • Pre-Screening Period (within 120 days prior to vaccination) [for purposes of ascertaining subjects with low or undetectable pre-existing HAI antibodies (≤10) to A/California/7/2009 (H1N1)-like virus] • Screening Period (within 45 days prior to vaccination) • Day 1 Visit (Baseline assessments; day of randomization and vaccination) • Day 3 Visit (Safety labs and cellular immunological sample collection) • Day 8 Visit (Cellular immunological sample collection) • Day 30 Visit (Serological sample collection) • Day 60 Visit (Safety checks) • Day 90 Visit (Serological and cellular immunological sample collection) In Part A subjects will be followed via phone call on Days 2, 4 to 7, 15 and then every two weeks between site visits on Days 45 and 75 for safety. **Part B Visits (Challenge-related):** **Starting at Day 90 post vaccination (Day of Challenge is Day B1)** • Day 90 (B1) (+30 day window; subjects challenged with influenza virus) • Days 89 – 95+ (B[-1] – B6+) (Subjects sequestered) • Day 120 (B30) (Post-Challenge; End of Active/Interventional Period) • Day 365 (End of Study Follow-up Period) In Part B subjects will be sequestered one day prior to viral challenge at Day 89 (B[-1]), with challenge at Day 90 (+30 days) at Day B1, and remain sequestered for 6 to 9 days. They will return to the site 30 days following their challenge, for their final efficacy and routine safety (unsolicited AEs) site visit. Daily clinical assessments, Flu-PRO questionnaire by subjects (symptoms of influenza), targeted physical exam by physician (signs of influenza), blood collection for cellular immunological, nasopharyngeal swabs and tissue paper weight (nasal secretions) collection will be performed routinely during the sequestration period per the schedule of events in Table 1b. Flu-PRO questionnaire will continue to be completed once daily by the subject from discharge to Day 14 post challenge. Blood samples for safety laboratory assessments (chemistry and hematology) will also be collected at Days B1, B3 and B6 post challenge, and more often if clinically indicated, per the discretion of the Investigator. Clinical assessments for safety and safety laboratory and cellular immunological blood sample collection will also be performed on the Day B30 post challenge clinic visit Subjects will also be contacted monthly between their Day B30 post-challenge visit and Day 365 for long-term safety follow-up (SAEs and AESIs/NOCIs).

INCLUSION CRITERIA	1. Male or female volunteers aged 18 – 49 years, inclusive 2. Able to give written informed consent 3. Low or undetectable pre-existing antibodies to the study vaccine as determined by an HAI titer of ≤10 against A/California/7/2009 (H1N1)-like viruses, measured within 120 days prior to vaccination 4. Healthy (no clinically significant health concerns), as determined by medical history, physical examination, 12-lead ECG, and vital signs at screening 5. Safety laboratory values within the following range criteria at screening: a. Normal range or < grade 1 elevation from normal (or decreased with NCS) for ALP, ALT, AST and bilirubin; b. Normal range for neutrophils (neutropenia) or elevated with NCS; c. Normal range (or abnormality with NCS) for: albumin, magnesium, total protein, hemoglobin, lymphocytes, white blood cells, platelets; phosphorous, amylase, BUN, CPK, creatinine, eosinophils, calcium, glucose, potassium and sodium; d. Negative laboratory value (or positive with NCS) for urine protein and blood urine 6. Body mass index between 17 and 35 at screening 7. Comprehension of the study requirements with ability and willingness to complete all assessments and comply with scheduled visits and contacts 8. Female participants must have a negative pregnancy test at screening and baseline and fulfill one of the following criteria: a. At least one year post-menopausal; b. Surgically sterile; c. Use of oral, implantable, transdermal or injectable contraceptives for 30 days prior to immunization and until 30 days after challenge (Day B30); • A reliable form of contraception approved by the Investigator (e.g. double barrier method, Depo-Provera, intrauterine device, Norplant, oral contraceptives, contraceptive patches, abstinence)
EXCLUSION CRITERIA	1. Receipt of any influenza vaccine within two years prior to study vaccination or plans to receive influenza vaccine during the active/interventional period of the study (i.e., through Day B30) 2. Administration of any investigational vaccine or investigational adjuvanted vaccine within 8 weeks preceding vaccination, or planned use of the above stated during the study through the 12-month safety follow-up period 3. Use of any investigational drug or device the greater of: within 4 weeks preceding vaccination, or planned use of the above stated during the study through the study active period (Day B30) OR within 5 half-lives of an investigational drug product 4. Administration of any licensed vaccine within 30 days prior to vaccination or planned use of the above stated during the active/interventional period of the study (i.e., through Day B30) 5. Presence of significant uncontrolled medical or psychiatric illness (acute or chronic) including institution of new medical/surgical treatment or significant dose alteration for uncontrolled symptoms or drug toxicity within 3 months of screening and reconfirmed at baseline 6. Any one of the following ECG findings within 30 days prior to vaccination: a. QTc F (interval duration > 450 msec (male) or > 470 msec (female), b. QRS interval greater than 120 msec,

EXCLUSION CRITERIA (cont.)	c. PR interval greater than 220 msec, d. Clinically significant ST-T wave changes or pathologic Q waves 7. Positive serology for HIV-1 or HIV-2, or HBsAg or HCV antibodies 8. Cancer, or treatment for cancer, within past 3 years (excluding basal cell carcinoma, squamous cell carcinoma, and cervical cancer in situ) 9. Presence of immunosuppression or medical condition possibly associated with impaired immune responsiveness or increased risk of severe influenza illness, including diabetes mellitus 10. Administration of any medications or treatments that may adversely affect the immune system such as allergy injections, immune globulin, interferon, immunomodulators, cytotoxic drugs or other drugs known to be associated with significant major organ toxicity, or systemic/inhaled corticosteroids during 3 months prior to vaccination or planned use of the above stated through 30 days post-challenge. Topical corticosteroids allowed 11. Presence of household members who have received the Ad4 or Ad7 vaccines within 2 months prior to vaccination 12. Presence of household members who are neonates, pregnant women, or hematopoietic stem cell transplant or solid organ transplant recipients or who are immunocompromised including those who are HIV positive. 13. History of drug, alcohol or chemical abuse within 1 year prior to vaccination 14. Receipt of blood or blood products 6 months prior to vaccination or planned administration during the follow-up study period 15. Donation of blood or blood products within 4 weeks prior to vaccination or planned donation during the follow-up study period 16. Acute disease within 72 hours prior to vaccination defined as the presence of a moderate or severe illness with or without fever (as determined by the Investigator through medical history and physical examination) or any acute respiratory or gastrointestinal illness even with mild symptoms occurring within 7 days of dosing the vaccine and within 7 days of entering the challenge phase 17. Presence of a fever ≥ 38ºC measured orally at baseline 18. Stool sample with occult blood at screening 19. Positive urine drug screen for drugs of abuse at screening or baseline 20. Positive breath or urine alcohol test at screening or baseline 21. Consistent/habitual smoking (> 2 cigarettes/week) within 2 months prior to vaccination 22. History of serious reactions to any vaccination such as anaphylaxis, respiratory problems, Guillain-Barre syndrome, hives or abdominal pain 23. History of documented egg allergy 24. History of a hypersensitivity or allergic reaction to any component of the investigational vaccine or placebo, including but not limited to fish gelatin 25. History of a hypersensitivity or allergic reaction to any component of the licensed QIV comparator vaccine (Fluzone®), including but not limited to eggs 26. Diagnosed bleeding disorder or significant bruising or bleeding difficulties that could make blood draws problematic or those with poor venous access that will make venipuncture difficult 27. History of irritable bowel disease or inflammatory digestive or gastrointestinal condition that could affect the distribution / safety evaluation of an orally administered vaccine targeting the mucosa of the small intestine. Such conditions may include but are not limited to: a. Esophageal Motility Disorder

EXCLUSION CRITERIA (cont.)	b. Malignancy c. Malabsorption d. Pancreaticobiliary disorders e. Irritable bowel syndrome f. Celiac Disease g. Inflammatory Bowel Disease h. Surgical Resection with the exception of appendectomy or a minor resection that is deemed acceptable by investigator and sponsor i. GERD j. Hiatal Hernia k. Peptic Ulcer (History of cholecystectomy is not exclusionary) 28. Any condition that resulted in the absence or removal of the spleen 29. History of any form of angioedema 30. Use of concomitant medications such as those listed below unless the subject can safely discontinue these medications for the rest of the study; none of these can be taken within 30 days of vaccination**:** a. Proton pump inhibitors b. Over-the-counter probiotics c. H2-blockers d. Anti-seizure medications e. Pain relief medications (chronic use) f. Cardiovascular drugs g. Imidazoles, Triazoles, and Thiozole antifungals h. Antidiarrheals i. Antibiotics i. Use of a brief ($\leq$ 10 days) course of oral or topical antibiotic for minor URI, UTI, dental work, or skin infection allowed within the screening period, but must be completed 7 days prior to vaccination j. Influenza antiviral medications (from screening to viral challenge) 31. The use of stable doses of prior and concomitant medications for the below indications is allowed: a. Dyslipidemia: No new agents for treatment or changes in dose of agents already in use within 3 months prior to screening (includes niacin obtained without prescription) b. Thyroid medications: Must be on a stable dose of medication (eg, levothyroxine) for at least 3 months prior to screening 32. Any condition that, in the opinion of the Investigator, might interfere with ability to assess the primary study objectives 33. Adulthood history of seasonal hay fever or a seasonal allergic rhinitis or perennial allergic rhinitis or chronic or nasal or sinus condition such as chronic sinusitis, abnormal nasal structure including septal deviation and nasal polyps unless the allergic rhinitis has been asymptomatic for the last 5 years. 34. Asthma, bronchiectasis or chronic obstructive pulmonary disease 35. Any known allergy or intolerance to oseltamivir
SAFETY, IMMUNOGENICITY, AND EFFICACY ASSESSMENTS	**Safety:** **PART A: Post-Vaccination (Day 1 to Day 90)** Safety and tolerability will be evaluated by: • Solicited symptoms listed below through Day 8: o Gastrointestinal reactions (nausea, vomiting, diarrhea and abdominal pain) for

SAFETY, IMMUNOGENICITY, AND EFFICACY ASSESSMENTS (cont.)	oral component o Injection site reactions (localized pain, tenderness, erythema and induration) for IM component, o Symptoms/signs of systemic reactogenicity (malaise/fatigue, myalgia, anorexia, fever and headache) • Unsolicited AEs will be recorded through 120 days post-immunization (90 days in Part A and 30 days post challenge in Part B) • SAEs and AESIs/NOCIs will be recorded through Day 365 • Physical exams, routine urinalysis, complete blood counts and serum chemistries will be collected pre-dose at Screening and Baseline and at Study Days 3, 8, 30, 90 (pre-challenge) • Vital signs will be recorded at all study visits • Female participants will have serum/urine pregnancy tests at screening, baseline, Day 30 and pre-challenge (Day 89 or Day 90). **PART B: Challenge Phase:** **Safety:** • Unsolicited AEs through 30 days post-challenge (Day B30 Visit) • Pre-challenge and daily throughout the sequestration period post-challenge: o Pulmonary function using a spirometer to assess forced vital capacity (FVC), (forced expiratory volume in one second) FEV_1, peak expiratory flow (PEF), and the FEV_1/FVC ratio; o Vital signs (blood pressure, heart rate (HR), respiratory rate, oral body temperature, and oxygen saturation (pulse ox) approximately every 4 hours, starting at the time of challenge administration) o 12-lead electrocardiograms (ECGs) o Assessment of safety laboratory panels (chemistry, hematology, and urinalysis) on Days B1, B3 and B6. Additional time points may be collected if clinically indicated per the discretion of the Investigator. • Follow-up clinic evaluations on 30 days post-challenge at Day B30 Visit **Immunogenicity/Efficacy:** **Post-vaccination (See** Table 1**, Part A)** Cellular and/or humoral immune function assays from blood samples obtained at baseline (pre-dose) and at protocol-specified Study Days 8, 30 and 90 (pre-challenge), depending on the assay. In addition, a final evaluation for immune status may be performed 30 days post-challenge. HA antibody response will be determined using HAI assay, a microneutralizing (MN) assay and by ASC assay. Anti-Adenovirus 5 immune responses will be measured at baseline (pre-immunization) and on Day 30. T cell responses will be measured by an HA-specific interferon-γ (IFN-γ) / granzyme B (GrB) dual ELISpot assay **Challenge Phase (See** Table 1**, Part B)** Solicitation of acute influenza symptoms post-challenge once each evening by the subject (Flu-PRO) through to Day 14 post-challenge, and signs of influenza once daily by the investigator during the sequestration period. Daily Pulmonary function using a spirometer to assess forced vital capacity (FVC), (forced expiratory volume in one second) FEV_1, peak expiratory flow (PEF), and the FEV_1/FVC ratio Nasopharyngeal swabs for multiplex molecular testing for respiratory viruses by Biofire FilmArray System or Luminex; influenza virus shedding 2 times daily during the sequestration period by quantitative RT-PCR

	Nasopharyngeal swabs to determine absence of viral shedding at baseline and prior to discharge: o A rapid influenza test will be used to access for the absence of viral shedding starting on Day 5 post challenge; once the rapid influenza test becomes negative on or after day 5, the subject's negative shedding status will be confirmed on that same day and the subsequent day using Biofire or Luminex to document negative PCR for 2 consecutive days. Oseltamivir therapy (5 days) will be begun at discharge in subjects that are symptomatic at discharge, per the discretion of the investigator. Tissue paper weights (nasal secretions) will be determined once each day, in the morning during the sequestration period
HALTING RULES	Halting Rules for study Part B are listed below. For each individual cohort, if any of these conditions are met during the sequestration period post viral challenge, no new enrollment or further challenge virus administration in subsequent cohorts will be initiated pending a full SMC safety review. 1. One or more subjects has a body temperature ≥103 °F (39.4°C) that is not responsive to acetaminophen; 2. One or more subjects has a reduction of forced expiratory volume in one second (FEV1) below 70% of the baseline value, difficulty breathing, oxygen saturation <90% while awake, or other evidence of lower respiratory tract involvement if deemed clinically significant by the investigator; 3. One or more subjects experience a treatment-related SAE or grade 4 clinical AE, or two or more subjects experience the same treatment-related grade 3 clinical AE or treatment-related grade ≥3 laboratory abnormality. The SMC will provide study oversight throughout the duration of the trial interventional and safety follow-up period (Day 0 through Day 365 post-vaccination). The IRB and CBER will be notified if the study is halted for safety concerns.

STUDY ENDPOINTS	Efficacy: Primary: The primary efficacy endpoint is the occurrence of illness (attack rate) caused by the homologous A strain influenza virus challenge at 3 months. Attack rate is defined as subjects experiencing at least one day with: 1) Two symptoms present of at least "A little bit", "1 time" or "Rarely" or one symptom present of at least "Somewhat", "2 times" or "Sometimes" among the acute influenza symptoms on the Flu-PRO (Appendix 2) and 2) laboratory-confirmed infection (≥2 positive quantitative RT-PCR results while sequestered) Comparisons will be performed in: 1) all challenged subjects in each respective vaccine group; and then 2) infected subjects (as defined by ≥2 positive quantitative RT-PCR results while sequestered) in each vaccine group Secondary: Flu-PRO symptom severity score (Appendix 2) Number of subjects with detectable shedding by qRT-PCR Virus shedding AUC by qRT-PCR Duration of shedding as detected by qRT-PCR Tissue paper weights Number of symptoms per Flu-PRO (Appendix 2) Number of signs per physician assessment (Appendix 3) Correlations between individual immunogenicity parameters (see below) and clinical efficacy Safety – Number (%) of subjects reported solicited (through Day 8), unsolicited adverse events (through Day 120), and SAEs and AESIs through Day 365; findings of clinical

	laboratory tests; findings of vital signs; and findings of physical examination and pulmonary function compared to placebo and QIV. Secondary Immunological: IgA and/or IgG ASC at Day 8, T cell (IFN-γ and GrB) responses at Day 8 against matched strain compared to licensed QIV and placebo Geometric mean titers of HAI, MN against matched strain compared to licensed QIV and placebo at Day 30 Geometric mean titers of HAI, MN against mismatched strains when compared to licensed QIV at Day 30 Geometric mean titers of HAI against matched strain compared to licensed QIV and placebo at Day 90 (pre-challenge) for all challenged subjects Exploratory: Determine the resulting vaccine specific immune response by CyTOF (Mass flow cytometry) and antibody repertoire sequence analysis, comparing oral VXA-A1.1 to the inactivated QIV vaccine for homing, memory, and activation of B and T cells.
STATISTICAL CONSIDERATIONS	This is a proof-of-concept study to determine whether VXA-A1.1 may enhance efficacy and/or immunogenicity when compared with licensed QIV. All analyses will be descriptive in nature, in which results for various endpoints described above will be compared among VXA-A1.1, QIV and placebo groups. No formal evaluation of sample size was performed with respect to these endpoints. Group sizes of 60, 60 and 30 are generally larger than those in previous influenza challenge studies in which subjects have received prior vaccination, and therefore should be large enough to detect any signals suggestive of improved efficacy or immunogenicity. No statistical adjustments will be made to account for multiplicity. In doing the analysis, any subjects who had elevated HAI titers greater than 1:10 on the pre vaccination (baseline) blood sample should be excluded from statistical analysis and should be excluded from the challenge phase analysis if they are enrolled.

Table 1 Schedule of Events

Part A (Vaccination Period)

Assessments	Pre-Screen[1] (Day -120 to Screen)	Screen (Day -45 to Day -1)	Baseline2 (Day -2 to Day 1 Visit)	Day 3 Visit	Days 2, 4-7 and Day 15* Contact	Day 8 (Week 1) Visit	Day 30 Visit	Day 60 Visit	Day 45 and Day 75 Contact	Day 90 Visit (Non-Challenge Subject)	Early Term (Day 0 – 30)	Early Term (Day 31 –90 pre-challenge)
Windows	**N/A**	**N/A**	**N/A**	**+1 day**	**±2 days***	**None**	**±2 days**	**±1 week**	**±1 week**	**+30 days**	**N/A**	**N/A**
Serum for Prescreening HAI Titer	**X[1]**											
Informed Consent	**X**	**X**										
Demographics		**X**										
Review Eligibility Criteria		**X**										
Medical History		**X**										
Physical Examination		**X**	**X**	**X**		**X**	**X**	**X**		**X**	**X**	**X**
Vital Signs		**X**	**X**	**X**		**X**	**X**	**X**		**X**	**X**	**X**
12-Lead ECG		**X**								**X**		
Spirometry										**X**		
Blood Sample – safety labs (fasting)		**X**	**X [2]**	**X**		**X**	**X**			**X**	**X**	
Blood Sample – HIV ELISA, HCV ELISA, HBV sAg		**X**										
Urinalysis		**X**	**X [2]**	**X**		**X**	**X**			**X**	**X**	
Urine Drug Screen		**X**	**X**							**X**		
Breath or Urine Alcohol Test		**X**	**X**							**X**		
Serum/Urine Pregnancy Test		**X**	**X**				**X**				**X**	**X**
Immunogenicity: Serum Assays – anti-Ad5 [3], HAI, MN			**X [2]**				**X**			**X**	**X**	**X**
Immunogenicity: PBMC Assays – ASCs, CyTOF, Ab Sequencing)			**X [2]**			**X**				**X**	**X**	
Immunogenicity: Fixed Whole Blood – CyTOF, cytocines) [heparinized tubes]			**X [2]**	**X**		**X**	**X**			**X**	**X**	
Gene Expression: RNA (PAXgene Blood RNA® Tubes)			**X [2]**			**X**				**X**		
Stool Sample for Occult Blood		**X**										
Vaccination			**X**									

Assessments	Pre-Screen[1] (Day -120 to Screen)	Screen (Day -45 to Day -1)	Baseline2 (Day -2 to Day 1 Visit)	Day 3 Visit	Days 2, 4-7 and Day 15* Contact	Day 8 (Week 1) Visit	Day 30 Visit	Day 60 Visit	Day 45 and Day 75 Contact	Day 90 Visit (Non-Challenge Subject)	Early Term (Day 0 – 30)	Early Term (Day 31 –90 pre-challenge)
Windows	**N/A**	**N/A**	**N/A**	**+1 day**	**±2 days***	**None**	**±2 days**	**±1 week**	**±1 week**	**+30 days**	**N/A**	**N/A**
Dispense Diary Card(s) [4]			**X**			**X**	**X**	**X**				
Collect Diary Card(s)						**X**	**X**	**X**		**X**	**X**	**X**
Review of Concomitant Medications		**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Assess for AEs[5], SAEs and AESIs/ NOCIs & Review Diary Cards			**X [8]**	**X [8]**	**X [8]**	**X [8]**	**X**	**X**	**X**	**X**	**X**	**X**

[1] Pre-screening for low or undetectable pre-existing antibodies to the study vaccine as determined by an HAI titer of ≤10 against A/California/7/2009 (H1N1)-like viruses
[2] Baseline blood samples may be drawn at up to two days (Day -2) of vaccination. Baseline safety labs do not need to be repeated if screening safety labs collected within two days of vaccination.
[3] Anti-Ad5 assay is not performed on serum collected at Day 90
[4] (1) Symptom Diary Card: solicited symptoms of reactogenicity from Day 1 – Day 8 Visit; (2) General Diary Card: clinical symptoms/events and con meds from Day 8 – Day 120 (B30) Visit; (3) AESIs/NOCIs Diary Card: AESIs/NOCIs from Day 120 (B30) Visit – Day 365 Contact
[5] Monitor AEs for acute symptoms of conjunctivitis, URI, loose stools/diarrhea within 7 days post-vaccination to evaluate for Ad5 infection. If reported, collect throat and rectal swabs and forward to test lab.

Part B (In-Patient Challenge Period)

Study Visits/Quarantine	Assessment Period (Days)									
Study Day (Part B)	**B(-1) (Day 89)**	**(B1) (Day 90)**	**(B2)**	**(B3)**	**(B4)**	**(B5)[1]**	**(B6)[1]**	**(B7)[1]**	**(B8)[1]**	**(B9)[1]**
Windows	**-2 days**	**+ 30**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**
Out Patient Period										
Quarantine Period	X	X	X	X	X	X	X	X	X	X
Informed Consent [2]	X[5]	X[5]								
Review Eligibility Criteria	X[5]	X[5]								
Vital Signs [3]	X[5]	X	X	X	X	X	X	X	X	X
12-lead ECG and Spirometry [4]	X[5]	X[5]	X	X	X	X	X	X	X	X
Physical Examination	X[5]	X[5]	X	X	X	X	X	X	X	X
Immunogenicity: Serum Assays – anti-Ad5 [6], HAI, MN	X[5]	X[5]					X			
Immunogenicity: PBMC Assays – ASCs, CyTOF, Ab Sequencing)	X[5]	X[5]		X			X			
Immunogenicity: Fixed Whole Blood – CyTOF, cytokines) [heparinized tubes]	X[5]	X[5]	X	X	X		X		X	
Gene Expression: RNA (PAXgene Blood RNA® Tubes)	X[5]	X[5]		X						
Serum/urine Pregnancy Test	X[5]	X[5]								
Serum/urine Drug and Breath/urine Alcohol Tests	X[5]	X[5]								
Blood Sample – safety labs (fasting)	X[5]	X[5]		X			X			
Urinalysis	X[5]	X[5]		X			X			
Viral Challenge		X[7]								
Influenza Symptom and Signs Scoring [8]	X[5]	X[5]	X	X	X	X	X	X	X	X
Nasopharyngeal Swabs for Viral Shedding/efficacy (qRT-PCR) [9]	X[5]	X[5]	X	X	X	X	X	X	X	X
Nasopharyngeal Swabs (rapid influenza test & Biofire/Luminex) [10]	X[5]	X[5]				X	X	X	X	X

Study Visits/Quarantine	Assessment Period (Days)									
Study Day (Part B)	**B(-1) (Day 89)**	**(B1) (Day 90)**	**(B2)**	**(B3)**	**(B4)**	**(B5)[1]**	**(B6)[1]**	**(B7)[1]**	**(B8)[1]**	**(B9)[1]**
Windows	**-2 days**	**+ 30**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**	**N/A**
Tissue Paper Weights (nasal secretions)		X[11]	X	X	X	X	X	X	X	X
Oseltamivir Administration [12]										X[12]
Review Concomitant Medications	X	X[11]	X	X	X	X	X	X	X	X
Assess for Adverse Events (including SAEs and AESIs/NOCIs)	X	X[11]	X	X	X	X	X	X	X	X
Dispense (at discharge) /Collect General Symptom Diary Card		X[11]						X	X	X
Query for SAEs and AESIs. Review AESIs/NOCIs Diary Card [13]										

[1] Subjects will have rapid influenza test daily starting on Day B5. If rapid influenza is test negative subjects will have confirmatory test (Biofire or Luminex) performed the same day and the subsequent day. Subjects will be discharged based on the Biofire/Luminex test results, their influenza signs and symptoms and any other available data at the discretion of the Investigator. Assessments will only be made once unless the subject is not fit to leave the unit. If subjects are not discharged on Day B6, the same assessments will be performed on Days 7 through 9, except immunological testing. Subjects who withdraw consent prior to absence of viral shedding will be requested to remain in quarantine but will only undergo assessments to determine absence of viral symptomology and shedding before discharge.

[2] In addition to the main Informed Consent Form (ICF), subjects will confirm consent prior to challenge in Part B

[3] Vital signs include blood pressure, heart and respiratory rate and body temperature. Blood pressure and heart rate will be measured in seated position after 5 min rest. Body temperature will b measured orally.
On Day B1, the morning vital signs measurement will be performed pre-challenge. From Day B1 to the day of discharge, vital signs will be performed approximately every 4 hours, starting at the time of challenge administration.

[4] ECG and spirometry will be measured once daily in supine and seated position, respectively, after 5 min of rest.

[5] Pre-challenge assessment. May be completed at Day B-1 or Day B1 pre-challenge.

[6] Anti-Ad5 assay is not performed on serum collected at Day 90

[7] Administration of challenge virus will be in the morning.

[8] Solicitation of acute influenza symptoms (Flu-PRO) will be completed by the subject in the morning prior to challenge and then every evening (after 3pm) and signs will be assessed by the investigator/study staff daily during the sequestration period. Subjects will continue to complete the Flu-PRO once daily following discharge to Day 14 post-challenge.

[9] Nasopharyngeal swabs for viral shedding (qRT-PCR) will be collected twice daily approximately 12 hours apart from Day B1 to day of discharge. Swabs should be collected from alternating nostrils (left or right) at each collection time point. The initial swab at Day B1 will be collected pre-challenge.

[10] A second nasopharyngeal swab will be collected for testing by Biofire/Luminex at baseline visit. Subsequent testing by rapid influenza test starting at Day B5 with confirmation of negative results by Biofire/Luminex test starting at Day B6. Data will be reviewed before discharging subject from unit; subjects with 2 consecutive negative values by Biofire/Luminex test may be discharged starting at Day B6.

[11] Pre-challenge

[12] Oseltamivir (5-day course) will be administered to symptomatic subjects starting on day of discharge per the discretion of the investigator.

[13] AESIs/NOCIs Diary Card will be dispensed at the Day B30 Visit (end of active period/last efficacy visit) to assist subjects in recording symptoms of AESIs/NOCIs through Day 365. Prior to Day B30. AESIs/NOCIs will be collected from Part A, Day 1 (vaccination) to Part B, Day B30 via site visits and phone contacts.

Follow-up Period

Study Visits/Quarantine	Assessment Period (Days)				
Study Day (Part B)	**Day B30 (D120)/ ET**	**Monthly Contact (Thru Day 365)**	**Early Term (Day B2-B9)**	**Early Term (Discharge–B30)**	**Early Term (B30 – End of Safety Follow-up)**
Windows	**±2 days**	**±1 week**			
Out Patient Period	X	X			
Vital Signs [1]	X				
12-lead ECG and Spirometry [2]	X				
Physical Examination	X				
Immunogenicity: Serum Assays – anti-Ad5, HAI, MN	X		X	X	
Immunogenicity: PBMC Assays – ASCs, CyTOF, Ab Sequencing)	X				
Serum/urine Pregnancy Test	X		X	X	
Blood Sample – safety labs (fasting)	X		X	X	
Urinalysis	X		X	X	
Oseltamivir Administration [3]			X [3]		
Review Concomitant Medications	X		X		
Assess for Adverse Events (including SAEs and AESIs/NOCIs)	X		X	X	
Dispense (at discharge) /Collect General Symptom Diary Card	X		X	X	
Query for SAEs and AESIs. Review AESIs/NOCIs Diary Card [4]		X			X

1 Vital signs include blood pressure, heart and respiratory rate and body temperature. Blood pressure and heart rate will be measured in seated position after 5 min rest. Body temperature will be measured orally

2 ECG and spirometry will be measured in supine and seated position, respectively, after 5 min of rest

3 Oseltamivir (5-day course) will be administered to symptomatic subjects starting on day of discharge or termination per the discretion of the investigator.

4 AESIs/NOCIs Diary Card will be dispensed at the Day B30 Visit (end of active period/last efficacy visit) to assist subjects in recording symptoms of AESIs/NOCIs through Day 365. Prior to Day B30. AESIs/NOCIs will be collected from Part A, Day 1 (vaccination) to Part B, Day B30 via site visits and phone contacts