IGHID 11911 - Cross-reactive N. gonorrhoeae Immune Responses Induced by a N. meningitidis Vaccine

NCT number NCT04094883
Document Date 06/24/2020

IGHID 11911 - Cross-reactive N. gonorrhoeae Immune Responses Induced by a N. meningitidis Vaccine

Funding Sponsor: North Carolina Translational & Clinical Sciences (NC TraCS) Institute
Funding Mechanism: Duke/UNC CTSA Consortium Collaborative Translational Research Grant

Protocol Date: 06/24/2020
Protocol Version: 1.2

Principal Investigator:
Joseph Alex Duncan, MD, PhD
Associate Professor of Medicine
Division of Infectious Diseases
University of North Carolina
Chapel Hill, NC 27599-7030
Phone: 919-966-6547
Fax: 919-843-1015
E-mail: jaduncan@med.unc.edu

Co-Investigator:
Marcia Hobbs, PhD
Professor of Medicine and
Microbiology Immunology
Division of Infectious Diseases
University of North Carolina
Chapel Hill, NC 27599-7030
Phone: 919-966-5050
Fax: 919-843-1015
E-mail:
marcia_hobbs@med.unc.edu

IGHID 11911 - Cross-reactive N. gonorrhoeae Immune Responses Induced by a N. meningitidis Vaccine

Protocol Date: 06/24/2020
Protocol Version: 1.2

SIGNATURE PAGE

The signature below documents the review and approval of this protocol and provides the necessary assurances that this study will be conducted according to the protocol, including all statements regarding confidentiality, and according to national, regional, and local legal and regulatory requirements.

_____
Site Principal Investigator Name (Print)

_____
Site Principal Investigator Signature

_____
Date

TABLE OF CONTENTS

LIST OF ABBREVIATIONS

Abbreviation	Definition
4CMenB	4 component *Neisseria meningitidis* serogroup B vaccine (trade name Bexsero™)
AE	Adverse Event
CI	Confidence Interval
CTRC	Clinical and Translational Research Center
ID	Identification
Ig	Immunoglobulin
IL	Interleukin
IRB	Institutional Review Board
ISM	Independent Safety Monitor
LOS	Lipo-oligosaccharide
MAb	Monoclonal Antibody
MBRB	Medical Biomolecular Research Building
mL	Milliliter
Ng	*Neisseria gonorrhoeae*
Nm	*Neisseria meningitidis*
OMV	Outer Membrane Vesicles
PBMC	Peripheral Blood Mononuclear Cells
TGF	Transforming growth factor
Th	T helper cell
Treg	Regulatory T cell
UNC	University of North Carolina at Chapel Hill
UNCH	UNC Hospitals

PROTOCOL SUMMARY

Protocol Title	Cross-reactive *N. gonorrhoeae* Immune Responses Induced by a *N. meningitidis* Vaccine
Study Design	Single arm, prospective clinical trial to assess immunologic responses induced by immunization with 4CMenB vaccine that cross react with *N. gonorrhoeae*. Participants will receive two-doses of an FDA-approved vaccine that provides protection from *N. meningitidis* infection according to the recommended dosing schedule. The participants will provide samples of blood as well as mucosal surface derived samples (urine and/or swabs) at four separate visits.
Study Population	Individuals 18-25 years of age that are not pregnant, HIV negative, have no history of congenital immunologic disorder, and are not taking immune suppressive medications
Number of subjects	15
Number of sites	1
Clinical Samples	Blood, pharyngeal swab, urine (male subjects), vaginal swab (female subjects)
Estimated Start of Enrollment	07/31/2019
Estimated Time to Completion	12/31/2020
Protocol Duration	Each individual will participate in the study for between 6 and 8 weeks.
Regimen or Intervention	Each participant will receive two doses of 4CMenB vaccine 5 weeks between doses.

1.0 HYPOTHESIS AND STUDY OBJECTIVES

1.1 Hypothesis

Immunization with 4CMenB induces cross reactive immune responses against N. gonorrhoeae

1.2 Primary Objectives

1. Determine whether immunization with 4CMenB induces serum antibodies against *N. gonorrhoeae* surface antigens
2. Determine whether polyfunctional CD4+ lymphocytes directed against *N. gonorrhoeae* surface antigens are induced by immunization with 4CMenB

1.3 Exploratory Objectives

1. Determine whether immunization with 4CMenB induces mucosal surface antibodies against *N. gonorrhoeae* surface antigens
2. Determine whether immunization with 4CMenB induces serum and/or mucosal surface antibodies against *N. gonorrhoeae* porB or mtrE
3. Determine whether immunization with 4CMenB induces serum and/or mucosal surface antibodies against *N. gonorrhoeae* antignens other than porB and mtrE
4. Determine whether polyfunctional CD8+ lymphocytes directed against *N. gonorrhoeae* surface antigens are induced by immunization with 4CMenB
5. Determine quantity and phenotype of CD4+ Tfh cells lymphocytes induced by immunization with 4CMenB
6. Determine inflammatory cytokine production by N. gonorrhoeae surface antigen-stimulated PBMC from 4CMenB-immunized humans.
7. Identify and clone B-cell receptors that recognize *N. gonorrhoeae* antigens after immunization with 4CMenB

2.0 INTRODUCTION

2.1 Background

***N. gonorrhoeae* poses global public health threats because of emerging antibiotic resistance.** The WHO estimates over 60 million new cases of *N. gonorrhoeae* (Ng) infection occur each year worldwide, making it one of the most common bacterial sexually transmitted infections[1]. Most cases of Ng infection are limited to the lower genital tract, causing urethritis in men and cervicitis in women. Antibiotic resistance in Ng presents a major problem in the control of this pathogen[2]. In 2018, three cases of extensively drug resistant Ng were identified in Australia and the United Kingdom after treatment failure with currently recommended dual antibiotic therapy[3]. Effective preventative strategies, particularly vaccines, are urgently needed to reduce the public health burden of Ng infections throughout the world.

Ng infection in humans does not lead to protective adaptive immune responses. Uncomplicated Ng infections are associated with increased risk of recurrent infections. Re-infections with the same Ng strain occur at a higher frequency than with other circulating strains in the community, indicating there is little strain-specific immunity to Ng[4]. Moreover, male subjects in experimental human Ng infection studies show no evidence of immunity upon rechallenge[5].

OMV-based meningococcal vaccines appear to provide cross-species protection against Ng infection. Effective vaccines for the most prevalent serogroups of *N. meningitidis* (Nm) have been in use in humans for decades. Meningococcal vaccines were first generated against polysaccharide capsular antigens of Nm serogroups A,C,Y, and W. Nm serogroup B (NmB) capsule is a poorly immunogenic disaccharide in humans which has made the development of vaccines targeting NmB

challenging and prevented the use of capsular polysaccharicde antigen for this purpose[6]. Recently, vaccines containing several proteins and lipoproteins have been generated from NmB OMV[7,8]. These vaccines are safe and have been used in mass vaccination campaigns in the setting of NmB epidemics in several countries[9-11]. STI surveillance in the setting of vaccine campaigns in Cuba and Norway showed reduced rates of Ng infections, which suggested Nm-OMV-based vaccines may offer cross-species protection against Ng[12,13]. STI education campaigns could have contributed to decreased Ng rates in these settings. However, a recent OMV-based vaccine campaign in New Zealand demonstrated decreased gonococcal infection rates, and surveillance data showed that infection rates for *Chlamydia trachomatis* (another sexually transmitted bacterial pathogen that causes urethritis and cervicitis) did not mirror the decrease in gonorrhea rates. This, and the fact that the effect was greater in vaccinated than unvaccinated persons, strongly supports the hypothesis that the Nm vaccine provided specific protection from Ng infection[14]. Data from New Zealand suggest that the Nm-OMV-based vaccine offered ~30% protection against Ng infection. Researchers at the Uniformed Services University of Health Sciences have demonstrated that mice vaccinated with the FDA-approved four component NmB OMV vaccine (4CMenB), delivered subcutaneously (SC) or intraperitoneally (IP), substantially accelerated Ng clearance and reduced vaginal Ng burdens vs. unvaccinated or adjuvant only-treated animals, supporting the ecologic data in humans that suggests Nm-OMV can induce immune responses that protect from Ng. Serum pooled from 10 individual humans that were immunized with 3 doses of 4CMenB and one individual who received the currently US FDA-approved 2 dose schedule of 4CMenB was recently shown to have increased antibody titers against whole Ng bacteria, demonstrating 4CMenB does indeed induce immunologic responses that cross react with Ng[15].

2.2 Rationale

Immunologic responses associated with protection from Nm disease are well-characterized. During a Nm outbreak at a US military base, a lack of detectable bactericidal antibodies against the outbreak strain of Nm was associated with high-rates of invasive infection in exposed individuals[16]. Bactericidal antibodies to Nm develop naturally during invasive infection and during nasopharyngeal carriage. Vaccination with capsular polysaccharide, which induced bactericidal antibodies, was shown to protect against invasive Nm disease[16-18]. In contrast to Nm, infection with non-encapsulated Ng weakly induces antibodies; anti-Ng antibody concentrations in vaginal secretions and sera are not different in individuals with current or past Ng infection vs. those with no history of infection[19]. There are few published studies of Ng-directed T cell responses in patients with Ng, but one small study showed that T cells from 30 Ng-infected patients exhibited weak proliferative responses to gonococcal PorB not significantly greater than proliferation without antigen[20]. Interleukin 17 (IL-17), the main cytokine from Th17 lymphocytes that stimulates neutrophil (PMN) recruitment and causes local inflammation, increases during Ng infections in humans and mice[21-23]. However, Ng is resistant to PMN killing, leading Russell et al. to suggest that stimulation of adaptive Th17 responses to Ng may partially explain the lack of protective immunity to Ng in humans[22].

Identification of the immunologic correlates of protection against *N. gonorrhoeae* would provide critical information that would allow for development and testing of vaccines for the prevention of *N. gonorrhoeae* infection. Characterization of the anti-gonococcal immune responses that develop in response to 4CMenB immunization will provide preliminary data that defines the immunologic parameters that can be assessed in the setting of a clinical trial designed to test the efficacy of 4CMenB or other vaccines for protection against *N. gonorrhoeae* infection. There is currently no data regarding the identity of gonococcal outermembrane antigens recognized by human antibodies after 4CMenB immunization as well as no documented reports of the cellular

immune response evoked by this vaccine. Consequently, this proposed pilot study will provide a crucial first step to defining the immunologic correlates of immunity to *N. gonorrhoeae* infection. Although the 4CMenB vaccine has been FDA approved, it is not currently required for enrollment as a student at the University of North Carolina at Chapel Hill. 4CMenB vaccine uptake has been estimated at 14% based on teen National Immunization Survey data; however, uptake rates between 14% and 98% have been found at Universities that have implemented vaccination campaigns in the setting of NmB disease outbreaks[24,25]. Thus, it is timely to conduct this study while prior MenB immunization rates in the target population are relatively low.

3.0 STUDY DESIGN

This study is a single center, single arm, interventional pilot study in which participants will receive two-doses of the 4CMenB vaccine according to the recommended administration schedule and will provide blood, pharyngeal swabs, and urine or self-collected vaginal swabs at each of four study visits.

The study population will **include 15 individuals** aged 18-25 years with no contraindication to vaccination and no known immune compromising medical condition or medication**.**

Participants will be seen for informed consent and eligibility screening. Enrolled participants will be given 1 dose of 4CMenB at enrollment and a second dose at week 5. Participants will be seen at entry, weeks 5, 6, and 7 for blood collection, pharyngeal swabs, and provide urine (male participants) or self-collected vaginal swabs collected for secondary screening and baseline immunologic testing.

4.0 SELECTION AND ENROLLMENT OF PARTICIPANTS

4.1 Inclusion Criteria

1. Able to understand and give informed consent
2. Willingness to undergo all study procedures.
3. Males or females between the ages of 18 to 25 at screening (inclusive).
4. Good health, as determined by medical history and targeted physical exam.
5. Participants agree to abstain from vaccines from study entry to 30 days after receipt of the second 4CMenB vaccine.
6. Female participants of child bearing potential must have a negative urine pregnancy test at the screening visit, and prior to receipt of vaccines at study entry and week 5 visits. Self-reported history is acceptable documentation of hysterectomy, bilateral oophorectomy, tubal ligation, or tubal micro-inserts which eliminate child bearing potential of participant. If participating in sexual activity that could lead to pregnancy, women must agree to use a form of contraceptive until 28 days after completion of the vaccine series. At least one of the following methods must be used appropriately: Condoms (male) with or without spermicidal agent, Diaphragm or cervical cap with spermicide, Intrauterine device (IUD), or Hormone-based contraceptive. Self-report of a monogamous male partner who has a vasectomy is also acceptable.

4.2 Exclusion Criteria

1. Known allergy/sensitivity or any hypersensitivity to latex or any of the components of the study product or its formulation (see section 5.2 for a list of components).
2. Participants who have received any vaccine directed against *N. meningitidis* serogroup B
3. Serious illness or injury requiring hospitalization within 21 days prior to study entry.

4. Current or prior history of a medical condition resulting in impaired immunity (such as HIV infection, inborn or acquired immunodeficiency syndromes, all cancers including leukemia or lymphoma, or the of use antineoplastic drugs or radiation treatment).
5. Known active infection with HIV, HCV, or HBV. This information will be obtained verbally from the participant.
6. History of excessive alcohol consumption, drug abuse, psychiatric conditions, social conditions or occupational conditions that in the opinion of the investigator would preclude compliance with the study.
7. Hemophilia or other bleeding diatheses.
8. Receipt of anticoagulants (aspirin or nonsteroidal anti-inflammatory drugs (NSAIDS) are acceptable) within 14 days prior to study entry.
9. Autoimmune disorders; mild autoimmune disorders, such as eczema, are not exclusionary and will be determined by the investigator.
10. Use of any systemic immunomodulatory treatment, systemic corticosteroids, (inhaled and topical corticosteroids acceptable), investigational products, interleukins, interferons, growth factors, or intravenous immunoglobulin (IVIG) within 45 days prior to study entry.
11. Pregnant women and nursing mothers or women who are planning to become pregnant or breastfeed within 28 days after receipt of their second 4CMenB vaccine.
12. Have received any licensed vaccine within 30 days prior to study vaccination.
13. Have donated blood or blood products within 30 days before study vaccination, plan to donate blood at any time during the study and up to 30 days after the last blood draw.
14. Any condition in the opinion of the investigator that would interfere with the proper conduct of the trial.

4.3 Study Recruitment and Enrollment Procedures

Potential participants will be recruited from the UNC-Chapel Hill University and surrounding area population. Potential participants will be recruited through flyers posted on the campus and through the UNC email listserv and through Join the Conquest at UNC. Potential participants will contact the study team via telephone or email and a study team member will provide information about the study and review eligibility criteria. Participants will receive a total of $130 over the course of the study to compensate for time and providing samples related to the study in addition to receiving the benefit of protection from invasive N. menigtidis infection as a result of receiving the FDA-approved 4CMenB vaccine over the course of the study. The payments for participation are $20 provided at screening visit, $20 at entry visit, and $30 each at week 5 visit, week 6 visit, and week 7 study completion visit. Participants will be paid at the end of each study visit.

To promote a roughly even balance in the sex at birth of participants, enrollment of additional participants of one sex at birth will be discontinued when enrollment of participants of that sex at birth reaches 8 participants.

Once a candidate has been identified, study details will be carefully discussed with the participant by the study coordinator or study investigator. The participant will be asked to read and sign the IRB approved informed consent form. For participants from whom a signed informed consent form has been obtained, a screening visit will be performed that will include obtaining information related to eligibility, completion of a targeted physical examination, and a point of care urine pregnancy test for female participants of child bearing potential. For participants from

whom informed consent has been obtained, but who are deemed ineligible or who do not enroll into initial immunization step, a Screening Failure Results form must be completed.

5.0 STUDY TREATMENT

Study treatment is defined as 4-component *Neisseria menigitidis* group B vaccine, 4CMenB (trade name: Bexsero™)

5.1 Regimens, Administration, and Duration

All participants will receive the 4CMenB vaccine, 0.5 mL, at entry (Day 0) and at week 5. The study product will be administered as supplied, intramuscularly in the deltoid region of the upper arm or the higher anterolateral area of the thigh in the clinic by a licensed study nurse, nurse practitioner, or study physician with experience administering vaccines using aseptic technique during preparation and administration. All injections should preferably be given in the deltoid region and in the non-dominant arm; however, if this is not feasible, the dominant arm may be used. The vaccine must not be injected intravascularly, subcutaneously, or intradermally. Vaccination should be deferred at the discretion of the site investigator if the participant is febrile or acutely ill. Following each vaccination, a licensed study nurse, nurse practitioner, or study physician with experience administering vaccines will observe the participant for 15 minutes for any adverse reactions/events.

5.2 Study Product Formulation and Preparation

4-Component Meningococcal Group B Vaccine
Each 0.5 mL dose of 4CMenB contains approximately:

• recombinant Neisserial adhesin A protein	50 mcg
• recombinant factor H binding protein	50 mcg
• recombinant Neisserial Heparin Binding Antigen protein	50 mcg
• Outer Membrane Vesicles	25 mcg
• aluminum hydroxide	1.5 mg
• Sodium chloride	3.13 mg
• L-histidine	0.78 mg
• sucrose	10 mg
• Water	quantity sufficient

5.3 Pharmacy: Study Product Acquisition/Distribution and Accountability

5.3.1 Study Product Acquisition/Distribution

4CMenB Vaccine is an FDA-approved product. Study vaccine will be purchased form the manufacturer through UNC Investigational Drug Service (IDS) Pharmacy.

5.3.2 Study Product Accountability

The site pharmacist is required to maintain complete records of all study product received from the manufacturer and subsequently dispensed. All unused study product will be distributed from the UNC Investigational Drug Service (IDS) Pharmacy to the primary investigator's laboratory for use in in vitro experiments after the study is completed or terminated.

5.4 Concomitant Medications

5.4.1 Required Medications

None

5.4.2 Prohibited Medications

- anticoagulants (aspirin or nonsteroidal anti-inflammatory drugs (NSAIDS) are acceptable)
- systemic corticosteroids (inhaled or topical corticosteroids are acceptable)
- systemic antineoplastic drugs
- systemic immunomodulatory drugs (including but not limited to immune modulating monoclonal antibodies, calcineurin inhibitors, antiproliferative agents, mTOR inhibitors, interleukins, interferons, and growth factors)
- investigational or approved vaccines (other than study product)
- intravenous immunoglobulin (IVIG)
- other investigational products

5.4.3 Precautionary Medications

None

6.0 CLINICAL AND LABORATORY EVALUATIONS

6.1 Schedule of Events

Evaluation	Screening	Entry	48-72 hr post injection	Post-Entry Evaluations			
				Week 5	48-72 hr post injection	Week 6	Week 7
Medical & medication history	X	X		X		X	X
Acute Illness Assessment		X		X			
Vital signs/targeted physical exam	X	X		X		X	X
Pregnancy test[1a]	X	X		X			
Phlebotomy collected blood (up to 60 mL per visit)		X		X		X	X
Stored PBMCs[2]		X		X		X	X
Stored serum[2]		X		X		X	X
Pharyngeal swab collection[2]		X		X		X	X
Self-collected vaginal swab[1,2]		X		X		X	X
Urine collection[2,3]		X		X		X	X
Vaccine injection		X		X			
Telephone contact			X		X		

[1] female participants only

[1a] female participants of child bearing potential

[2] mucosal surface swabs, urine, serum, and PBMC preparations will be used for immunologic analysis that will be conducted on batched samples.

[3] male participants only

6.2 Timing of Evaluations

6.2.1 Screening Evaluations

Screening evaluations include: medical history, medication history within the past 60 days, vital signs, targeted physical exam, and pregnancy test for female participants of child bearing potential). Screening visit must be conducted no greater than 30 days prior to enrollment visit. The screening evaluation and enrollment visit can all occur on the same day. Participants may be re-screened one time.

6.2.2 Entry Evaluations

Entry evaluations will be collected PRIOR to phlebotomy, specimen collection, and vaccine administration:

Acute Illness Assessment will be completed before phlebotomy, specimen collection, and vaccine administration. Participants who have an acute illness may be rescheduled for their phlebotomy, specimen collection, and vaccine within their screening visit window.

Update medical history and concomitant medications. Conduct a targeted physical exam and collect vital signs [heart rate (HR), blood pressure (BP), oral temperature (Temp), respiration rate (RR), and weight] prior to phlebotomy to collect up to 60 mL of blood, specimen collection, and vaccine administration.

For females of reproductive potential, negative urine pregnancy testing results must be available before phlebotomy, specimen collection, and vaccine administration.

Samples to be stored for immunologic testing will be batch run at the end of the study: pharyngeal swabs, urine sample (from male participants), self-collected vaginal swabs (from female participants), serum, and PBMC.

6.2.3 Post-Entry Evaluations

Week 5 Evaluations:

Acute Illness Assessment will be completed before phlebotomy, specimen collection, and vaccine administration. Participants who have an acute illness or who have taken exclusionary anticoagulant medications may be rescheduled for their vaccine within their screening visit window.

Update medical history and concomitant medications before phlebotomy and vaccine administration. Conduct a targeted physical exam and collect vital signs (HR, BP, Temp, RR) before phlebotomy to collect up to 60 mL of blood, specimen collection, and vaccine administration.

For females of reproductive potential, negative urine pregnancy testing results must be available before phlebotomy, specimen collection, and vaccine administration.

Week 5 laboratory evaluations will be collected PRIOR to vaccine administration:

Samples to be stored for immunologic testing will be batch run at the end of the study: pharyngeal swabs, urine sample (from male participants), self-collected vaginal swabs (from female participants), serum, and PBMC.

Week 6 and Week 7 Evaluations:

Update medical history and concomitant medications prior to phlebotomy to collect up to 60 mL of blood and specimen collection.

Samples to be stored for immunologic testing will be batch run at the end of the study: pharyngeal swabs, urine sample (from male participants), self-collected vaginal swabs (from female participants), serum, and PBMC.

6.2.4 Discontinuation Evaluations

Participants who elect not to receive or cannot receive the initial immunization or second immunization within the appropriate study time window are permanently discontinued from the study and will have no further evaluations conducted upon discontinuation. Participants who remain eligible but elect to prematurely discontinue form the study will have no additional evaluations performed from the point at which they discontinue. Participants who experience a grade 3 or 4 Adverse Event related to the vaccine or study procedures prior to the second immunization will be discontinued from the study and will return for evaluation at week 6 and/or 7 to ensure resolution but will not provide specimens for immunologic evaluation.

6.3 **Instructions for Evaluations**

6.3.1 Medical & Medication History

At screening, a standard medical history and medication history will be obtained and documented. Specifically, information will be obtained regarding:

- Known allergy/sensitivity or any hypersensitivity to latex or any of the components of the study product or its formulation (see section 5.2 for a list of components).
- Serious illness or injury requiring hospitalization within 21 days prior to study entry.
- A history of a medical condition resulting in impaired immunity (including complement deficiency, antibody deficiency, chronic granulomatous disease, or HIV infection).
- Autoimmune disorders; mild autoimmune disorders such as eczema is not exclusionary after assessment by the investigator.
- Known active infection with HIV, HCV, or HBV.
- History of all cancers including leukemia or lymphoma
- History of excessive alcohol consumption, drug abuse, psychiatric conditions, social conditions, or occupational conditions that in the opinion of the investigator would preclude compliance with the study.
- Hemophilia or other bleeding diatheses.
- Have donated blood or blood products within 30 days before study vaccination, plan to donate blood at any time during the duration of subject study participation, or plan to donate blood within 30 days after the last blood draw.
- Pregnancy in women and nursing mothers or women who are planning to become pregnant within the study duration.
- Prior history of gonorrhea, chlamydia, or syphillis

The medication history will include inquiry and documentation of all medications used within the last 60 days and the participants will be asked specifically about the use of the following agents:

- anticoagulants other than aspirin or nonsteroidal anti-inflammatory drugs (NSAIDS) within 14 days prior to study entry.
- any systemic antineoplastic or immunomodulatory treatment, systemic corticosteroids, (inhaled or topical corticosteroids acceptable), investigational products, interleukins, interferons, growth factors, or intravenous immunoglobulin (IVIG) within 45 days prior to study entry.

The medication history will also include inquiry and documentation of vaccine history and the participants will be asked specifically about the use of the following:

- Previous immunization with a MenB vaccine
- Have received any licensed vaccine within 30 days prior to study vaccination.
- Have planned vaccination with any vaccine within 30 days of the anticipated date of second study immunization (approximately 65 days from study entry).

6.3.2 Targeted Physical Exam & Vital Signs

A physical exam performed at each visit will include at a minimum an examination of the skin, head, mouth, and neck; auscultation of the chest; cardiac exam; abdominal exam; and examination of the lower extremities for edema. A targeted physical exam and vital signs (oral temperature, pulse, respiration rate, and blood pressure) including weight will be collected at each visit.

6.3.3 Laboratory Evaluations

Pregnancy Test for females of child bearing potential

A point of care pregnancy test is performed and results need to be recorded only in source document at screening visit, at entry, and at week 5 visit prior to vaccine administration.

6.3.4 Immunologic Studies

Stored PBMCs

PBMCs for cell-surface marker phenotyping, *N. gonorrhoeae*-antigen stimulated cytokine production, and B-cell receptor cloning will be isolated and stored per the SOE (Schedule of Events). Cryopreserved PBMC will be sent to Duke Immune Profiling Core at Duke University for flow cytometry and cellular immune analysis; the shipped samples will have an identifying sample number (code) derived from the participant's assigned PID and the visit number; no identifying information on the participant's samples will be transmitted to the Duke Immune Profiling Core and leftover samples wil be destroyed or returned to UNC for storage if the participant has agreed to storage of their study specimens. The PI at UNC will keep a master log which links the codes on the stored specimens to the participants. Leftover PBMC from consenting participants may be used for future *N. gonorrhoeae* research.

N. gonorrhoeae reactive antibody studies/Stored Serum

Pharyngeal and vaginal swabs, urine, and serum for measures of *N. gonorrhoeae*-directed antibodies will be collected per the SOE. Antibodies eluted from swabs or present in urine and present in serum from samples collected at entry, week 5, week 6, and week 7 will be tested for *N. gonorrhoeae* antigen-specific antibodies using ELISA. Antibody containing samples will be sent to Regional Biocontainment Laboratory (RBL) Immunology Unit at Duke University for ELISA analysis. The shipped samples will have an identifying sample number (code) derived from the participant's assigned PID and the visit number; no identifying information on the participant's samples will be transmitted to the Duke RBL Immunology Unit and leftover samples will be destroyed or returned to UNC for storage if the participant has agreed to storage of their study specimens. The PI at UNC will keep a master log which links the codes on the stored specimens to the participants. Leftover sera from consenting participants may be used for future N. gonorrhoeae research.

6.3.5 Vaccine/ Injection

Every effort should be made to administer vaccines within the protocol-defined visit window. Participants who do not receive their first vaccine within 30 days of screening or who do not receive the week 5 vaccine will be prematurely discontinued from the study.

The week 5 visit should be conducted 35 days (± 7 days) after the first vaccination (enrollment visit). If the week 5 visit is not within the protocol-specified window, then the vaccine can be given as late as 8 weeks post enrollment.

The week 6 visit should occur 5-9 days after the second dose of vaccine; if week 6 visit samples are not collected, the participant may continue in the study and attend the week 7 visit 12-16 days after receiving the second dose of vaccine. If the week 7 visit is not collected between day 12-16 after the second dose of vaccine, participant will be discontinued from the study.

Vaccination should be deferred at the discretion of the site investigator if the participant is febrile with reported or observed temperature greater than or equal to 38 °C or acutely ill with symptoms (ongoing or present within 7 days of planned vaccination) consistent with infection of the respiratory tract, gastrointenstinal tract, urinary tract, or skin/soft tissues.

Participants will remain at the clinic for a 15-minute post-injection observation period following each vaccination to observe for syncope or any immediate adverse reactions. Participants will be provided instructions on self-assessment of adverse reactions related to vaccination.

6.3.6 Telephone Contact

The study staff will contact each participant by telephone 48 to 72 hours after each vaccination at the entry visit and at the week 5 visit to determine whether any side effects have occurred. Information from the telephone contact will be recorded on the electronic case report form (eCRF). If a participant experiences any side effects that are Grade 3 or 4, including edema (swelling), erythema (redness), or induration of the injection site, the participant should be scheduled to come into the research clinic for evaluation, including a targeted physical exam focused on the area involved, as soon as possible.

NOTE: If telephone contact is not possible, then in person is permissible.

7.0 CLINICAL MANAGEMENT ISSUES

7.1 Dose Modification

No modification of the study vaccine dose is allowed.

7.2 Toxicity Management

7.2.1 Management of Injection Site and Allergic Reactions

Local and systemic reactions will be graded according to the DAIDS Adverse Event (AE) Grading Table, Corrected Version 2.1, July 2017, found on the DAIDS RSC Web site: http://rsc.tech-res.com/safetyandpharmacovigilance/.

Injection Site Reactions

- Swelling, induration, or redness at the injection site
- Pain or tenderness, or other reactions at the injection site

Systemic reactions occurring within 48 hours after vaccination

- Fever or chills
- Malaise or fatigue
- Headache or pain, e.g., myalgia or arthralgia
- Nausea or vomiting
- Allergic reactions, e.g., rash, hives, dyspnea, pruritis

For any Grade 3 or 4 reactions thought definitely, possibly, or probably related to vaccination, further vaccines should not be given to that participant. The participant will be asked to return to the site for an evaluation and followed until AE has been resolved.

7.2.2 Other Adverse Events

For toxicities not specifically addressed above, the following guidelines should be used for the management of AEs that are felt to be at least possibly related to vaccines:

Grade 1 or 2 Toxicity/AE:

Participants who develop a Grade 1 or 2 AE may continue in the study and receive the second dose of vaccine (which may be postponed as directed in section 6.3.5) and return for evaluation study visits as noted in section 6.2.3. For participants experiencing Grade 1 or 2 AEs who choose to discontinue study treatment or study participation prior to receiving the second vaccine dose, see section 6.2.4.

Grade 3 or 4 Toxicity/AE:

Participants who develop a Grade 3 or 4 AE that is at least possibly related to vaccine should be re-evaluated for that toxicity until the AE returns to Grade ≤2. Participants who experience Grade 3 or 4 AE that is at least possibly related to vaccine should not receive the second immunization if the AE occurs prior to the week 5 study visit and their participation in the study should be discontinued. Participants who experience a grade 3 or 4 AE that is considered by the investigator to be unrelated to vaccine or that occurs after the second immunization may continue in the study and study specimens may be obtained for week 6 and 7 visits if the AE has returned to Grade ≤2. For instructions regarding participants experiencing Grade 3 or 4 AEs that are at least possibly related to vaccine that result in premature discontinuation of study vaccine and/or study participation, see section 6.2.4. These participants should be followed until resolution of the AE Toxicity.

7.3 Pregnancy

Pregnant participants are discontinued from the study at the time pregnancy is discovered.

8.0 ADVERSE EVENT REPORTING

8.1 Definitions

An adverse event (AE) is any untoward medical occurrence in a patient or clinical investigation participant administered a pharmaceutical product and which does not necessarily have a causal relationship with this treatment. An adverse event can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not related to the medicinal (investigational) product.

An unexpected (unlisted) adverse event is an adverse reaction, the nature or severity of which is not consistent with the applicable product information (e.g., Investigator's Brochure for an unapproved investigational product or package insert/summary of product characteristics for an approved product).

A serious adverse event (SAE) is any untoward medical occurrence that:

- results in death;
- is life-threatening;
- requires inpatient hospitalization or prolongation of existing hospitalization;
- results in a persistent or significant disability/incapacity;
    - or
- is a congenital anomaly/birth defect

Other important medical events as assessed by medical and scientific judgment may also be considered SAEs by the investigator and should be reported in an expedited fashion.

An Unanticipated Problem (UP)/New Safety Information (NSI) is defined by the UNC IRB as any incident, experience, or outcome that:

- is unexpected (in terms of nature, severity, or frequency) given (a) the research procedures that are described in the protocol-related documents, such as the IRB-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- is related or possibly related to a participant's participation in the research; and
- suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) related to the research than was previously known or recognized.

AE attribution will be assigned according to the following scale:

- Not related: The AE is clearly not related to the study procedures (i.e., another cause of the event is most plausible and/or a clinically plausible temporal sequence is inconsistent with the onset of the event).
- Possibly related: An event that follows a reasonable temporal sequence from the initiation of study procedures, but that could readily have been produced by a number of other factors.
- Related: The AE is clearly related to the study procedures.

8.2 **Reporting**

The protocol-defined expedited event reporting period for this protocol is the entire study duration for an individual participant (from study enrollment until study completion or discontinuation of the participant from study participation for any reason).

All AEs defined below must be recorded on the eCRFs if any of the following criteria have been met:

- All grade ≥3 AEs
- All AEs that led to a change in study treatment/intervention regardless of grade
- All AES meeting SAE definition

For all of these adverse events reported or observed, the information should be recorded in the electronic Case Report Form (eCRF) for that participant and will include a full description of the event, its seriousness, its severity or toxicity grade (intensity), the relationship to the study drug, and the treatment, outcome, and sequelae of the event.

Any serious or unexpected event, which occurs to any participant in the course of his/her treatment on this study or within 30 days following cessation of treatment, must be reported within 24 hours to the principal investigator. The immediate reports should be followed promptly by detailed, written reports. Copies of these reports should be sent to:

- UNC IRB if it meets the UNC IRB UP/NSI reporting requirements
- Events that are reportable to the UNC IRB will also be submitted to the appropriate representative of the study funding source/sponsor (NC TraCS): Marie Rape, RN, BSN, Associate Director of Regulatory, NC TraCS (marie_rape@med.unc.edu)

Reporting is required of all unanticipated problems/new safety information to the UNC IRB, including those which may occur after the participant has completed or has withdrawn from the study, including after study closure.

Clinically significant AEs and other problems related to vaccines will also be reported to the Vaccine Adverse Event Reporting System (VAERS), which is maintained by the FDA and CDC (http://vaers.hhs.gov/).

All individual AE shall be maintained by the Principal Investigator. For those events that are not reportable to the UNC IRB, a summary (i.e., not individual reports) of all adverse events that have occurred within the last approval period will be submitted to the UNC IRB at the time of continuing review.

9.0 STATISTICAL CONSIDERATIONS

9.1 **General Design Issues**

This study is a single center, single arm interventional pilot study in which participants will receive two-doses of the US FDA-approved 4CMenB vaccine according to recommended administration schedule. Blood and mucosal surface specimens will be obtained at 4 study visits. The study population will include 15 individuals aged 18-25 years desiring immunization against group B *N. meningitidis* with no contraindication to vaccination and no known immune compromising medical condition or medication. Enrolled individuals who are discontinued will be replaced.

9.2 **Endpoints**

9.2.1 Primary Endpoints:

- Geometric mean concentration of IgG binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human serum at entry visit (prior to first vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human serum at entry visit (prior to first vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human serum at entry visit (prior to first vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Frequency of CD4+ T cells expressing at least two different activation markers (IFN-g, TNF-a, IL-2, and CD107a) after in vitro stimulation with *N. gonorrhoeae* strain FA1090 Outer-membrane vesicles in circulating PBMC obtained at entry visit (prior to first vaccination) and week 7 visit (2 weeks after second vaccination) determined by flow cytometry

9.2.2 Exploratory Endpoints:

- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human pharyngeal secretions at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human vaginal secretions (female participants) at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (Outer-membrane vesicles) in human urine (male participants) at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (PorB, and MtrE) in human serum at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (PorB, and MtrE) in human pharyngeal secretions at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (PorB, and MtrE) in human vaginal secretions (female participants) at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA

- Geometric mean concentration of IgM, IgG, and IgA binding to *N. gonorrhoeae* strain FA1090 surface antigens (PorB, and MtrE) in human urine (male participants) at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by ELISA
- Qualitative assessment of the presence of human IgG directed against *N. gonorrhoeae* strain FA1090 antigens (Outer-membrane vesicles, PorB, and MtrE) in human serum at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by immunoblot.
- Frequencies of CD8+ T cells expressing functional IFN-g, TNF-a, IL-2, and CD107a after in vitro stimulation with *N. gonorrhoeae* strain FA1090 Outer-membrane vesicles in circulating PBMC obtained at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by flow cytometry
- Frequencies of memory, activated, and proliferating CD4+ Tfh cells specific in circulating PBMC obtained at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by flow cytometry
- The quantity of IFN-g and IL-17 secreted after in vitro stimulation with *N. gonorrhoeae* strain FA1090 Outer-membrane vesicles from circulating PBMC obtained at entry visit (prior to first vaccination), week 5 visit (4-6 weeks after first vaccination), week 6 visit (1 week after second vaccination), and week 7 visit (2 weeks after second vaccination) determined by Bio-Plex Multiplex immunoassay
- The sequence of B-cell receptors found to associate with FA1090 Outer-membrane vesicles through flow cytometry-based cell sorting will be determined using sequencing of PCR amplicons generated using primers for non-variable regions of the B cell receptor

9.3 Sample Size and Accrual

Due to the majority of the target population likely to be undergraduate students, we anticipate that enrollment will be limited to ~1 participant each month in July and August (months when student population residing in Chapel Hill is low) and that the remaining participants will be enrolled at ~6/month in September and October. All Serum and PBMC will be stored until a complete specimen set is obtained from at least 8 subjects; the serum and PBMC specimens will then be transferred to Dr. Andrew MacIntyre's laboratory at Duke University in batch for immunologic assays. We plan to enroll up to 15 subjects so we can obtain a complete specimen set at all 4 study visits for at least 8 subjects.

9.4 Monitoring

Oversight of the trial is provided by the Principal Investigator (PI). The PI will be responsible for safety monitoring throughout the study. The PI will assure that informed consent is obtained prior to performing any research procedures, that all subjects meet eligibility criteria, and that the study is conducted according to the IRB-approved research plan. Study data are accessible at all times for the PI to review. The PI will review study conduct including: accrual, drop-outs, protocol deviations after 5 participants are enrolled and complete the study protocol and then on an annual basis until study is complete. The PI reviews AEs individually in real-time and in aggregate after 5 individuals have completed the protocol and then annually until study is complete. The PI reviews and reports all serious adverse events (SAEs) and unanticipated events as defined in

section 8.0 in real-time. The PI ensures all protocol deviations, AEs, and SAEs are reported to the NC TraCS and the IRB according to the applicable reporting requirements.

9.5. Analyses

9.5.1 Statistical Methods:

Descriptive statistics such as age and gender will be reported in means, standard deviations, frequencies, and proportions when appropriate. As recently recommended for pilot studies, ours will focus on point estimates of immuse responses and their 80% confidence intervals (CIs) in place of 95% CI used for traditional statistical inferences[26] .The geometric mean concentration (anti-log of the arithmetic mean of log transformed concentrations) of antibodies while the arithmetic mean of T cell population frequencies and secreted cytokine levels from in vitro antigen-stimulated PBMC will be identified. Comparisons between measurements of immune response in pre- and post-immunization specimens (as specified for primary outcome measurements) will be completed using paired Student's t-test for normally distributed data or Wilcoxon rank signed test for non-normally distributed data. For repeated outcome measurements at 4 time points (as specified in exploratory outcomes), we will use generalized estimating equations (GEE) with unstructured covariance matrix to estimate the time difference. The link function for GEE will be chosen depending on the data type.

9.5.2 Sample Size and Power:

There is only one published report describing the anti-gonococcal antibody titers in 10 humans who received 3 immunizations with 4CMenB. The geometric mean titer of serum antibodies directed against whole cell *N. gonorrhoeae* (strain 1291) demonstrated a statistically significant increase between pre (95% CI : 25,906–90,811) and post (95% CI : 49,228–126,115) immunization samples[15]. We do not have preliminary data and there is no published data regarding cross- reactive antibodies in 4CMenB-immunized humans against the *N. gonorrhoeae* strain FA1090 (the strain used by our center in both human challenge model and preclinical laboratory studies) or specific FA1090 outer-membrane antigens. Further, there is no published data on anti-gonococcal antibody subtype or levels of these antibodies in mucosal surface secretions in any 4CMenB-immunized humans. Finally, there is no published data regarding cellular immune responses to gonococcal antigens in 4CMenB-immunized individuals. Thus, the proposed study is a pilot study and the power to detect significant immune responses towards *N. gonorrhoeae* antigens is unknown. The study will provide baseline values and variability in measurements for non-immune individuals as well as post immunization measurements for the studied immunologic parameters. We will enroll up to 15 individuals, including similar numbers of men and women; this sample size is proposed based on size of potential participant pool and available funding for the study. The data generated will provide valuable information for how to deploy these assays in human populations receiving 4CMenB immunization for testing efficacy in preventing *N. gonorrhoeae* infection in the future.

10.0 STUDY MANAGEMENT

10.1 Institutional Review Board (IRB) Review and Informed Consent

This protocol and the informed consent document and any subsequent modifications (amendments) will be reviewed and approved by the IRB or ethics committee responsible for oversight of the study.

It is expected that the IRB will have the proper representation and function in accordance with federally mandated regulations. The IRB should approve the consent form(s) and protocol.

In obtaining and documenting informed consent, the principal investigator should comply with the applicable regulatory requirement(s), and should adhere to Good Clinical Practice (GCP) and to ethical principles that have their origin in the Declaration of Helsinki.

Before recruitment and enrollment onto this study, the participant will be given a full explanation of the study and will be given the opportunity to review the consent form(s). Each consent form must include all the relevant elements currently required by the FDA Regulations and local or state regulations, which include elements such as the purpose of the study, the procedures to be followed, and the risks and benefits of participation. Once this essential information has been provided to the participant and the investigator is assured that the participant understands the implications of participating in the study, the participant will be asked to give consent to participate in the study by signing an IRB-approved consent form.

Prior to a participant's participation in the trial, the written informed consent form(s) should be signed and personally dated by the participant or the participant's legally authorized representative (legal guardian or person with power of attorney for participants who cannot consent for themselves), and by the person who conducted the informed consent discussion.

A copy of the consent form will be given to the participant or legal guardian, and this fact will be documented in the participant's record.

10.2 Registration Procedures

All participants must be registered with the Global HIV Prevention and Treatment Clinical Trials Unit Office at the University of North Carolina before enrollment onto study. Prior to registration, eligibility criteria must be confirmed with the Study Coordinator.

Each participant will be assigned a participant identification (PID) number prior to study entry. Therefore, all participants are identified by a PID number and not by name on all case report forms.

10.3 Criteria for Discontinuation

Removal of participants from study can occur for the following reasons:

10.3.1 Premature Treatment Discontinuation

- Treatment-related AEs as discussed in section 7.2.
- Pregnancy or breastfeeding.
- Clinical reasons believed life-threatening by the site principal investigator, even if not addressed in section 7.2.

10.3.2 Premature Study Discontinuation

- Request by the participant to discontinue treatment prior to receiving second immunization or to withdraw from study.
- Request of the primary care provider if s/he thinks the study is no longer in the best interest of the participant.
- Failure to start study vaccination within 30 days after screening or to receive second vaccination within 8 weeks of initial dose.
- Failure to attend week 6 or week 7 study visits within the indicated time window.

- At the discretion of the IRB, NC TraCS, industry supporter, FDA, Office for Human Research Protections (OHRP), investigator, or other country-specific government agencies as part of their duties to ensure that research participants are protected.

10.4 **Deviations from the Protocol**

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

a) IRB for review and approval
b) to the sponsor for agreement and, if required
c) to the appropriate regulatory authority(ies)

10.5 **Clinical Site Monitoring and Record Availability**

Personnel from the Global HIV Prevention and Treatment Clinical Trials Unit at the University of North Carolina (UNC) will monitor the trial for quality assurance. These monitors will review all source documents used in the preparation of the case report forms to assure proper conduct of the trial and proper collection of the data.

10.6 **Required Documentation**

Before the study can be initiated, the following documentation must be provided to the IGHID Regulatory Office at the University of North Carolina.

- A copy of the official IRB approval letter for the protocol and informed consent(s)
- IRB membership list
- CVs and medical licensure for the principal investigator and any associate investigators who will be involved in the study
- A copy of the IRB-approved consent form(s)

10.7 **Adherence to the Protocol**

Except for an emergency situation in which proper care for the protection, safety, and well-being of the study participant requires alternative treatment, the study shall be conducted exactly as described in the approved protocol. Any deviation from the protocol must have prior approval by the Principal Investigator and must be recorded and explained.

10.8 **Amendments to the Protocol**

Should amendments to the protocol be required, the amendments will be originated and documented by the Principal Investigator. The written amendment will be approved by the principal investigator and must be submitted to the IRB at the principal investigator's site for approval. It should also be noted that when an amendment to the protocol substantially alters the study design or the potential risk to the participant, a revised consent form might be required.

10.9 **Data Management**

Data to be obtained during the study includes directed medical history provided by participant self-report, personally identifying information (including name, contact information, results of directed physical examination, and point of care pregnancy tests administered to female participants, as well as results of research tests to monitor immunologic response to immunization. Medical history and personal identification information will be recorded with identifiers and kept in participants' research charts. After written informed consent is provided, consecutive study code numbers will

be assigned to the participants to ensure confidentiality. This study code number will be used to identify research specimens and the results of research tests. At the end of each individual's participation in the trial, research charts will be reviewed by the study coordinator and stored in locked cabinets in the PI's office. At the completion of the study, the research charts and all identifying information related to participants will be archived in an off-site storage facility.

De-identified data entered into computerized files will be stored in a password-protected, encrypted file on a UNC-owned server with access provided only to authorized personnel directly involved with the study. Only authorized personnel directly involved with the study will have access to individually identifiable private information about participants. All data entered into the study database will be coded and unlinked to personal identifiers. Data from the trial to be stored electronically or shared will be unlinked to all identifiable information.

10.10 **Record Retention**

Study documentation includes all Case Report Forms, data correction forms or queries, source documents, Sponsor-Investigator correspondence, monitoring logs/letters, and regulatory documents (e.g., protocol and amendments, IRB correspondence and approval, signed participant consent forms).

Source documents include all recordings of observations or notations of clinical activities and all reports and records necessary for the evaluation and reconstruction of the clinical research study.

Government agency regulations and directives require that all study documentation pertaining to the conduct of a clinical trial must be retained by the study principal investigator. Study documents will be kept on file until three years after the completion and final study report of this investigational study.

10.11 **Obligations of Investigators**

The Principal Investigator is responsible for the conduct of the clinical trial at the site in accordance with Title 21 of the Code of Federal Regulations and/or the Declaration of Helsinki. The Principal Investigator is responsible for personally overseeing the treatment of all study participants. The Principal Investigator must assure that all study site personnel, including co-investigators and other study staff members, adhere to the study protocol and all FDA/GCP regulations and guidelines regarding clinical trials both during and after study completion.

The Principal Investigator will be responsible for assuring that all the required data will be collected and entered onto the Case Report Forms (CRFs). At the completion of the study, all case report forms will be reviewed by the Principal Investigator and will require his/her final signature to verify the accuracy of the data.

The site principal investigator will make study documents (e.g., consent forms, drug distribution forms, CRFs) and pertinent hospital or clinic records readily available for inspection by the local IRB, the site monitors, the FDA, the NCATS, the OHRP, or NC TraCS for confirmation of the study data.

10.12 **Participant Confidentiality**

All laboratory specimens and accompanying evaluation forms, reports, and other records that are sent to Duke University for immunologic analysis will be de-identified to maintain participant confidentiality. All records will be kept locked in secure areas/secure offices at UNC. All

computer entry and networking programs at UNC will be done with coded numbers only. Clinical information will not be released without written permission of the participant, except as necessary for monitoring by the IRB, the NCATS, the OHRP, or NC TraCS.

10.13 **Study Discontinuation**

The study may be discontinued at any time by the IRB, NCATS, the OHRP, NC TraCS or other government agencies as part of their duties to ensure that research participants are protected.

11.0 BIOHAZARD CONTAINMENT

As the transmission of HIV and other blood-borne pathogens can occur through contact with contaminated needles, blood, and blood products, appropriate blood and secretion precautions will be employed by all personnel in the drawing of blood and shipping and handling of all specimens for this study, as currently recommended by the Centers for Disease Control and Prevention and the NIH.

All dangerous goods materials, including diagnostic specimens and infectious substances, must be transported using packaging mandated by CFR 42 Part 72. Please refer to instructions detailed in the International Air Transport Association (IATA) Dangerous Goods Regulations.

12.0 REFERENCES

1 Tapsall, J. W. Antibiotic resistance in Neisseria gonorrhoeae. *Clin Infect Dis* **41 Suppl 4**, S263-268, doi:10.1086/430787 (2005).

2 Unemo, M. *et al.* High-level cefixime- and ceftriaxone-resistant Neisseria gonorrhoeae in France: novel penA mosaic allele in a successful international clone causes treatment failure. *Antimicrob Agents Chemother* **56**, 1273-1280, doi:10.1128/AAC.05760-11 (2012).

3 European Centre for Disease Prevention and Control. Extensively drug-resistant (XDR) Neisseria gonorrhoeae in the United Kingdom and Australia. (2018).

4 Fox, K. K. *et al.* Longitudinal evaluation of serovar-specific immunity to Neisseria gonorrhoeae. *Am J Epidemiol* **149**, 353-358 (1999).

5 Schmidt, K. A. *et al.* Experimental gonococcal urethritis and reinfection with homologous gonococci in male volunteers. *Sex Transm Dis* **28**, 555-564 (2001).

6 Finne, J., Leinonen, M. & Makela, P. H. Antigenic similarities between brain components and bacteria causing meningitis. Implications for vaccine development and pathogenesis. *Lancet* **2**, 355-357 (1983).

7 Naess, L. M. *et al.* Human IgG subclass responses in relation to serum bactericidal and opsonic activities after immunization with three doses of the Norwegian serogroup B meningococcal outer membrane vesicle vaccine. *Vaccine* **17**, 754-764 (1999).

8 Peeters, C. C. *et al.* Phase I clinical trial with a hexavalent PorA containing meningococcal outer membrane vesicle vaccine. *Vaccine* **14**, 1009-1015 (1996).

9 de Moraes, J. C. *et al.* Protective efficacy of a serogroup B meningococcal vaccine in Sao Paulo, Brazil. *Lancet* **340**, 1074-1078 (1992).

10 Holst, J. *et al.* Properties and clinical performance of vaccines containing outer membrane vesicles from Neisseria meningitidis. *Vaccine* **27 Suppl 2**, B3-12, doi:10.1016/j.vaccine.2009.04.071 (2009).

11 Nokleby, H. *et al.* Safety review: two outer membrane vesicle (OMV) vaccines against systemic Neisseria meningitidis serogroup B disease. *Vaccine* **25**, 3080-3084, doi:10.1016/j.vaccine.2007.01.022 (2007).
12 Pérez, O. *et al. Mucosal approaches in Neisseria Vaccinology*. (2009).
13 Whelan, J., Klovstad, H., Haugen, I. L., Holle, M. R. & Storsaeter, J. Ecologic Study of Meningococcal B Vaccine and Neisseria gonorrhoeae Infection, Norway. *Emerg Infect Dis* **22**, 1137-1139, doi:10.3201/eid2206.151093 (2016).
14 Petousis-Harris, H. *et al.* Effectiveness of a group B outer membrane vesicle meningococcal vaccine against gonorrhoea in New Zealand: a retrospective case-control study. *Lancet* **390**, 1603-1610, doi:10.1016/S0140-6736(17)31449-6 (2017).
15 Semchenko, E. A., Tan, A., Borrow, R. & Seib, K. L. The serogroup B meningococcal vaccine Bexsero elicits antibodies to Neisseria gonorrhoeae. *Clin Infect Dis*, doi:10.1093/cid/ciy1061 (2018).
16 Goldschneider, I., Gotschlich, E. C. & Artenstein, M. S. Human immunity to the meningococcus. I. The role of humoral antibodies. *J Exp Med* **129**, 1307-1326 (1969).
17 Goldschneider, I., Gotschlich, E. C. & Artenstein, M. S. Human immunity to the meningococcus. II. Development of natural immunity. *J Exp Med* **129**, 1327-1348 (1969).
18 Gotschlich, E. C., Rey, M., Triau, R. & Sparks, K. J. Quantitative determination of the human immune response to immunization with meningococcal vaccines. *J Clin Invest* **51**, 89-96, doi:10.1172/JCI106801 (1972).
19 Hedges, S. R., Mayo, M. S., Mestecky, J., Hook, E. W., 3rd & Russell, M. W. Limited local and systemic antibody responses to Neisseria gonorrhoeae during uncomplicated genital infections. *Infect Immun* **67**, 3937-3946 (1999).
20 Simpson, S. D., Ho, Y., Rice, P. A. & Wetzler, L. M. T lymphocyte response to Neisseria gonorrhoeae porin in individuals with mucosal gonococcal infections. *J Infect Dis* **180**, 762-773, doi:10.1086/314969 (1999).
21 Feinen, B., Jerse, A. E., Gaffen, S. L. & Russell, M. W. Critical role of Th17 responses in a murine model of Neisseria gonorrhoeae genital infection. *Mucosal Immunol* **3**, 312-321, doi:10.1038/mi.2009.139 (2010).
22 Liu, Y. & Russell, M. W. Diversion of the Immune Response to Neisseria gonorrhoeae from Th17 to Th1/Th2 by Treatment with Anti-Transforming Growth Factor {beta} Antibody Generates Immunological Memory and Protective Immunity. *MBio* **2**, doi:10.1128/mBio.00095-11 (2011).
23 Gagliardi, M. C. *et al.* Circulating levels of interleukin-17A and interleukin-23 are increased in patients with gonococcal infection. *FEMS Immunol Med Microbiol* **61**, 129-132, doi:10.1111/j.1574-695X.2010.00759.x (2011).
24 Walker, T. Y. *et al.* National, Regional, State, and Selected Local Area Vaccination Coverage Among Adolescents Aged 13-17 Years - United States, 2017. *MMWR Morb Mortal Wkly Rep* **67**, 909-917, doi:10.15585/mmwr.mm6733a1 (2018).
25 Soeters, H. M. *et al.* University-Based Outbreaks of Meningococcal Disease Caused by Serogroup B, United States, 2013-2018. *Emerg Infect Dis* **25**, 434-440, doi:10.3201/eid2503.181574 (2019).
26 Lee, E. C., Whitehead, A. L., Jacques, R. M. & Julious, S. A. The statistical interpretation of pilot trials: should significance thresholds be reconsidered? *BMC Med Res Methodol* **14**, 41, doi:10.1186/1471-2288-14-41 (2014).